Kinetic Modeling of the Light-Dependent Photosynthetic Activity of the Green Microalga *Chlorella vulgaris*

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Abstract: Light-dependent photosynthesis of *Chlorella vulgaris* was investigated by using a novel photosynthesis measurement system that could cover wide ranges of incident light and cell density and reproduce accurate readings. Various photosynthesis models, which have been reported elsewhere, were classified and/or reformulated based upon the underlying hypotheses of the light dependence of the algal photosynthesis. Four types of models were derived, which contained distinct light-related variables such as the average or local photon flux density (APFD or LPFD) and the average or local photon absorption rate (APAR or LPAR). According to our experimental results, the LPFD and LPAR models could predict the experimental data more accurately although the APFD and APAR models have been widely used for the kinetic study of microalgal photosynthesis. © 2003 Wiley Periodicals, Inc. *Biotechnol Bioeng* **83:** 303–311, 2003.
Keywords: microalgae; photosynthesis; light; modeling; *Chlorella vulgaris;* photosynthesis measurement system

INTRODUCTION

Photoautotrophic microalgal mass culture has been extensively studied with various purposes such as production of biomass as a source of fine chemicals or foods (Becker, 1988, 1994; Glombitza and Koch, 1989) and wastewater treatment (Aziz and Ng, 1992; Chevalier and de la Naue, 1985; de la Naue and Proulx, 1988; Yun et al., 1999). Recently, microalgal photosynthesis was considered as an effective means to reduce the emission of carbon dioxide, a major greenhouse gas, to the atmosphere (Benemann, 1997; Karube et al., 1992; Yun and Park, 1997a,b; Yun et al., 1996). As the potential of microalgal mass culture increases, kinetic modeling of microalgal photosynthesis and/or growth has become of significant importance because an accurate model is a prerequisite for designing an efficient photobioreactor, predicting process performance, and optimizing operating conditions (Aiba, 1982; Ever, 1991; Rabe and Benoit, 1962). In addition, the mechanistic modeling approach can be useful for confirming or rejecting the hypotheses on mechanisms underlying the models.

Light is the most important factor affecting microalgal photosynthesis kinetics (Aiba, 1982; Becker, 1994). In general, most microalgal mass culture systems are limited by light. Light cannot penetrate deeply into dense microalgal suspensions because it is absorbed and scattered by the microalgal cells (Yun and Park, 2001). Therefore, light is spatially distributed along the light path inside of the photobioreactors. The light availability is expressed as either photon flux density ($\mu E\ m^{-2}\ s^{-1}$) or photon absorption rate ($\mu E\ kg^{-1}$ cell s^{-1}), which is different from that of common soluble substrates. Such distinct features of light make it difficult to mathematically describe the light dependence of the microalgal photosynthetic activity.

Many researchers have suggested various kinds of mathematical models on algal photosynthesis and/or growth kinetics (Aiba, 1982; Cornet et al., 1992b; Ever, 1991; Iehana, 1983; Prokop and Erickson, 1995; Van Liere and Mur, 1979). The underlying hypotheses of models can be classified into three types based upon the expression of the light dependence of microalgal photosynthesis. In the first type, algal photosynthesis is assumed to be dependent on the average photon flux density (APFD) that can be obtained by volume-averaging the spatially distributed photon flux density inside the photobioreactor (Molina Grima et al., 1994, 1997; Prokop and Erickson, 1995; Rabe and Benoit, 1962). The second type of model (Aiba, 1982; Iehana, 1983, 1990; Koizumi and Aiba, 1980; Van Liere and Mur, 1979) uses the volume-averaged photon absorption rate (average photon absorption rate, APAR). The third type (Cornet et al.,

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1992a, 1992b; Ever, 1991; Tamiya et al., 1953) is based upon a more sophisticated hypothesis that each algal cell responds to the photon flux density arriving at its position (local photon flux density, LPFD). However, it is not easy to measure the photosynthetic activity of each cell; the apparent activity (i.e., the oxygen production rate of whole algal culture) is generally monitored and compared with the model outputs. Finally, we can consider an additional hypothesis that the photosynthetic activity of a cell is related to the photon absorption rate by the cell (local photon absorption rate, LPAR). The LPAR model has not been examined yet.

We evaluate herein the accuracy of four types of mathematical models derived on the basis of different hypotheses respectively to describe the kinetics of light-limited algal photosynthesis in terms of the short-term oxygen generation rate of non-growing algal cells. A novel system of photosynthesis measurement was developed in order to obtain accurate experimental data sets. In the model evaluation, the prediction performance and the rationale of hypothesis were estimated. Although the kinetic models of short-term photosynthesis evaluated in this study cannot be directly applied for predicting the microalgal growth, the evaluation on these models is expected to give a basic clue to the light-dependence description for further developments of growth models predicting actual large-scale photobioreactor systems.

MODEL DEVELOPMENT

Four conceptual hypotheses were used as a starting point for the derivation of the mathematical model equations comprising the experimentally measurable terms. In order to precisely express the light attenuation in the algal suspensions, a modified Beer–Lambert model (Yun and Park, 2001) was used, taking into account the non-linearity between absorbance and cell density, especially in the high-density suspension. Symbols used in this work are summarized in Table I.

APFD Model

Rabe and Benoit (1962) suggested that the average photon flux density could be correlated with the specific growth rate. This implies that the average photon flux density could be a representative value of spatially distributed density within the photobioreactor. Such an approach has been most widely adopted in the algal biotechnology field (Molina Grima et al., 1994, 1997; Prokop and Erickson, 1995).

The average photon flux density can be calculated from the light distribution. As can be seen in Figure 1A, light is attenuated through absorption and scattering depending on light path length and cell concentration. The average photon flux density is the volume-averaged value of the position-dependent photon flux density. Therefore, the area filled with vertical lines should be the same as that filled with horizontal lines in the plot of photon flux density versus light path length (Fig. 1A).

The APFD model can be formulated mathematically as follows:

$$P_X(X,L) = \frac{P_m PFD_{ave}(X,L)}{K + PFD_{ave}(X,L)} - R_X, \quad (1)$$

Table I. Explanation of symbols used and their values.

Symbol	Description	Unit	Value
a	Constant in the light attenuation model [Eq. (2)]	m^{-1}	1041[a]
b	Half constant in the light attenuation model [Eq. (2)]	kg m^{-3}	1.03[a]
PFD	Local photon flux density calculated from Eq. (2)	$\mu E\ m^{-2}\ s^{-1}$	
PFD_o	Incident photon flux density	$\mu E\ m^{-2}\ s^{-1}$	
PFD_{ave}	Average photon flux density calculated from Eq. (3)	$\mu E\ m^{-2}\ s^{-1}$	
K	Half constant in the algal photosynthesis models	$\mu E\ m^{-2}\ s^{-1}$ or $\mu E\ kg^{-1}\ s^{-1}$	Parameter[b]
l	Light path length at a certain position	m	
L	Depth of the photobioreactor	m	0.018
$LPAR$	Local photon absorption rate calculated from Eq. (8)	$\mu E\ kg^{-2}\ s^{-1}$	
$LPAR_{ave}$	Average photon absorption rate calculated from Eq. (6)	$\mu E\ kg^{-2}\ s^{-1}$	
P_m	Maximum specific photosynthetic activity in the algal photosynthesis models	g O_2 kg^{-1} h^{-1}	Parameter[b]
P_X	Apparent specific photosynthetic activity	g O_2 kg^{-1} h^{-1}	
P'_X	Local specific photosynthetic activity	g O_2 kg^{-1} h^{-1}	
P_V	Apparent volumetric photosynthetic activity ($=XP_X$)	g O_2 m^{-3} h^{-1}	
R_X	Specific respiratory activity	g O_2 kg^{-1} h^{-1}	4.05[c]
X	Microalgal cell concentration	kg m^{-3}	

[a]Determined according to Yun and Park (2001).
[b]Adjustable parameters in the algal photosynthesis models and their values are listed in Table II.
[c]Determined by measuring the oxygen consumption rate in the dark condition.

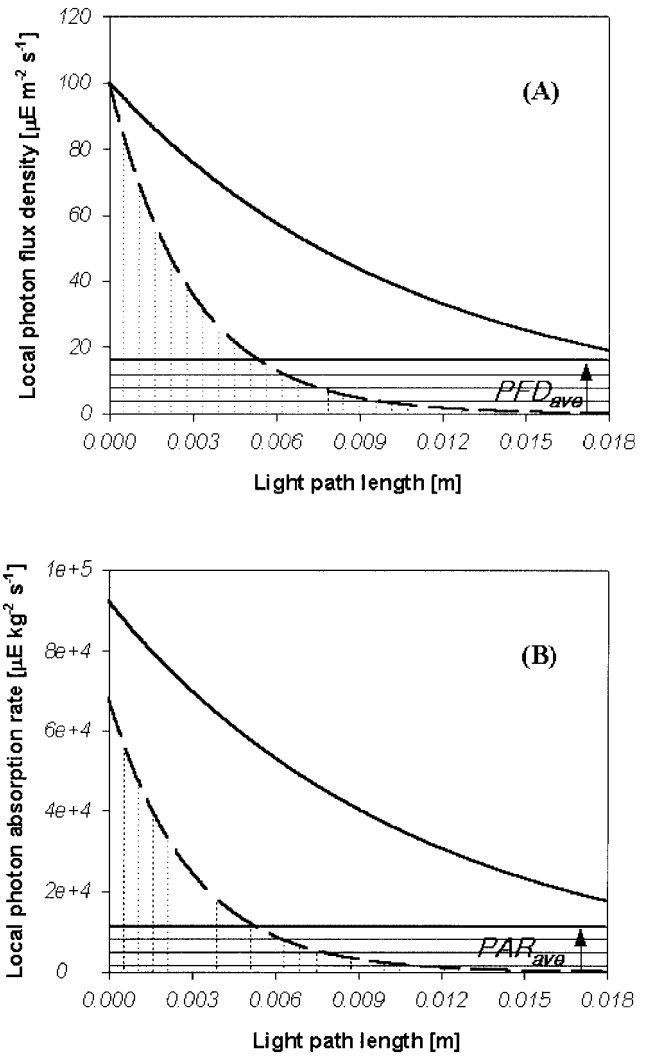

Figure 1. Spatial distributions of photon flux density (A) calculated using Eq. (2) and of photon absorption rate (B) calculated using Eq. (8). The cell densities were 0.1 kg m^{-3} (solid line) and 0.5 kg m^{-3} (dotted line), and the incident photon flux density was fixed at 100 μE m^{-2} s^{-1}. The average light intensity and the average light absorption rate are calculated using Eqs. (3) and (6), respectively.

where P_X is the photosynthetic activity, P_m is the maximum photosynthetic activity, K is the half constant, and R_X is the respiratory activity. The subscript X represents the specific activity (activity per unit dry weight of cell). Here, the average photon flux density (PFD_{ave}) can be calculated using Eq. (3) from the modified Beer–Lambert model [Eq. (2)]:

$$PFD(X,l) = PFD_o \exp\left(-\frac{aXl}{b+X}\right), \quad (2)$$

$$PFD_{ave}(X,L) = \frac{1}{L}\int_0^L PFD(X,l)dl = \frac{PFD_o\,(b+X)}{aXL}\left\{1-\exp\left(-\frac{aXL}{b+X}\right)\right\}, \quad (3)$$

where PFD_o is the incident light intensity, a and b are empirical constants, X is the cell density, l is the light path length at a certain position, and L is the depth of the photobioreactor. In this work, the configuration of the photobioreactor was designed to be parallel to the light beam (Fig. 2). Therefore, the integration for obtaining the volume-averaged light intensity can be carried out along the unidirectional path. Because the microalgal culture is assumed to be well mixed and thus uniform throughout the culture volume, the cell concentration could be regarded as a constant during the integration.

LPFD Model

In the LPFD model, the photosynthetic activity is assumed to be dependent on the local photon flux density. Therefore, the photosynthetic activity is also spatially distributed within the photobioreactor. In other words, each algal cell is considered to respond to the local photon flux density corresponding to its position. Some researchers used the LPFD model to describe the light dependence of algal photosynthesis (Cornet et al., 1992a,b; Ever, 1991; Tamiya et al., 1953).

According to the LPFD hypothesis, the local photosynthetic activity (P'_X) at a position (l) can be expressed as follows:

$$P'_X(X,l) = \frac{P_m\,PFD(X,l)}{K+PFD(X,l)} - R_X = \frac{P_m\,PFD_o\exp\left(-\frac{aXl}{b+X}\right)}{K+PFD_o\exp\left(-\frac{aXl}{b+X}\right)} - R_X, \quad (4)$$

where R_X is the respiration rate, which is assumed to be independent of the photon flux density. While the local photosynthetic activity (P'_X) cannot be easily measured, an apparent activity (generally in terms of oxygen production rate) of the total algal culture can be determined by measuring the change of dissolved oxygen concentration. The measured activity can be considered to be the volume-average value of the local activity. Therefore, in order to derive the model equation including the measurable activity term (P_X), Eq. (4) was integrated throughout the total volume of the photobioreactor as follows:

$$P_X(X,L) = \frac{1}{L}\int_0^L P'_X(X,l)dl = \frac{P_m(b+X)}{aXL}\ln\frac{PFD_o+K}{K+PFD_o\exp\left(-\frac{aXL}{b+X}\right)} - R_X. \quad (5)$$

APAR Model

Several researchers (Aiba, 1982; Koizumi and Aiba, 1980; Van Liere and Mur, 1979) found the linear relationship

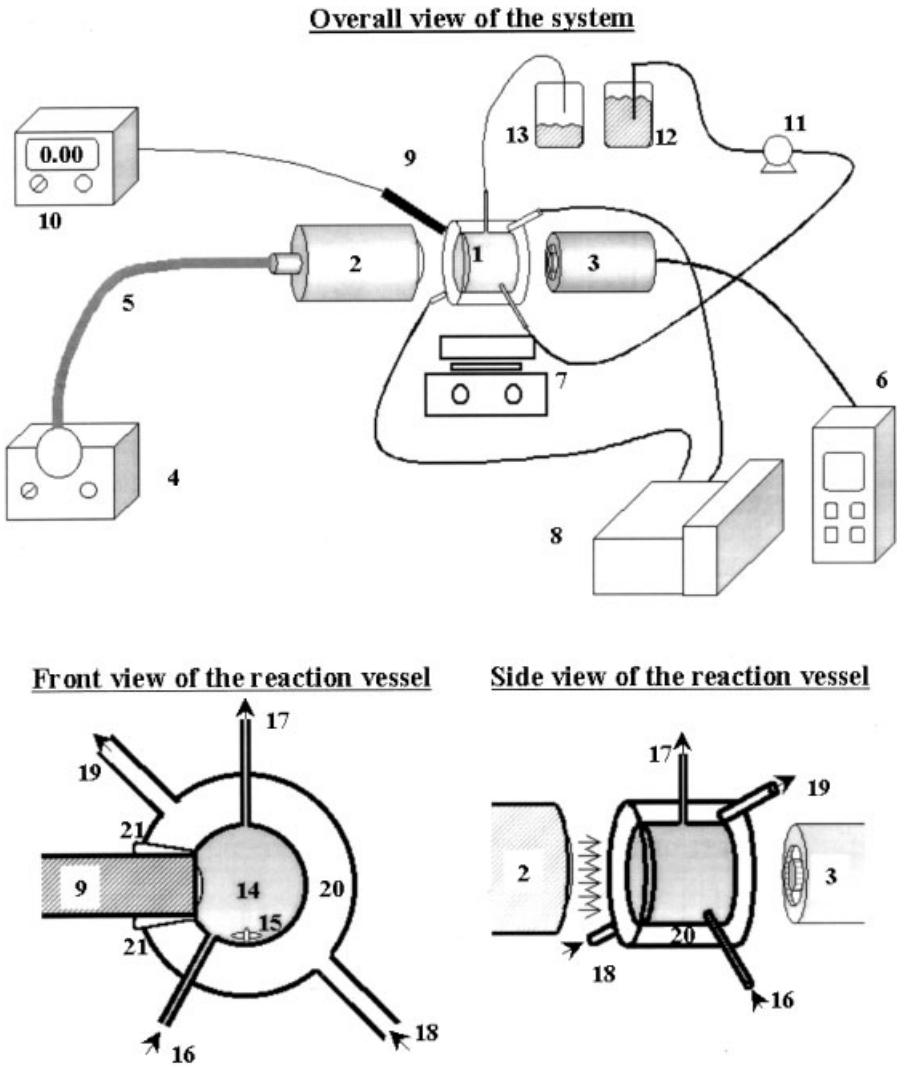

Figure 2. Schematic diagram of the photosynthetic activity measurement system and the reaction vessel: 1, reaction cell; 2, convex lens; 3, quantum sensor; 4, quartz halogen illuminator; 5, goose-neck-type optical fiber; 6, data logger; 7, magnetic stirrer; 8, water bath; 9, dissolved oxygen electrode; 10, dissolved oxygen meter; 11, peristaltic pump; 12, sample reservoir; 13, waste reservoir; 14, microalgal suspension; 15, magnetic bar; 16, inlet of sample; 17, outlet of sample; 18, inlet of cooling water; 19, outlet of cooling water; 20, cooling water jacket; and 21, septum.

between the average photon absorption rate by the whole algal culture and the specific growth rate, suggesting the use of average photon absorption rate for the kinetic analysis of microalgal activity. However, Iehana (1983, 1990) revealed non-linearity in this relationship at high cell concentrations. Therefore, a Monod-type expression, suggested by Iehana (1983, 1990), would be a more adequate approach able to cover a wide range of cell concentration.

As shown in Figure 1B, to calculate the average photon absorption rate, the local photon absorption rate should be also integrated throughout the total volume. However, instead of complicated integration procedure, the average photon absorption rate (PAR_{ave}) can be calculated more easily from difference between incident and exit photon flux densities with an assumption that the light beam penetrates straight without radial dispersion:

$$PAR_{ave}(X,L) = \frac{\{PFD_o - PFD(X,L)\}}{XL} = \frac{PFD_o}{XL}\left\{1 - \exp\left(-\frac{aXL}{b+X}\right)\right\}. \quad (6)$$

Therefore, the photosynthetic activity can be expressed as a function of the average photon absorption rate:

$$P_X(X,L) = \frac{P_m PAR_{ave}(X,L)}{K + PAR_{ave}(X,L)} - R_X = \frac{P_m PFD_o\left\{1 - \exp\left(-\frac{aXl}{b+X}\right)\right\}}{KXL + PFD_o\left\{1 - \exp\left(-\frac{aXL}{b+X}\right)\right\}} - R_X, \quad (7)$$

where the unit of K ($\mu E\ kg^{-1}\ s^{-1}$) is different from that of K in Eqs. (1) and (5) ($\mu E\ m^{-2}\ s^{-1}$).

LPAR Model

In addition to three types of models, we can consider another reasonable hypothesis that has not been studied. As can be seen in Figure 1B, the local photon absorption rate is also a function of position. The photosynthetic activity of an algal cell is probably dependent on the absorption rate of photon by the cell, which is designated as local photon absorption rate (LPAR) to stress its position dependence.

The local photon absorption rate (PAR) can be expressed with an assumption of non-dispersed light penetration as follows:

$$PAR(X,l) = \frac{1}{X}\left(-\frac{dPFD}{dl}\right) = \frac{aPFD_o}{b+X}\exp\left(-\frac{aXl}{b+X}\right). \quad (8)$$

Based upon the hypothesis, the position-dependent photosynthetic activity (P'_X) can be considered as a function of the local photon absorption rate:

$$P'_X(X,l) = \frac{P_m PAR(X,l)}{K + PAR(X,l)} - R_X = \frac{P_m aPFD_o \exp\left(-\frac{aXl}{b+X}\right)}{K(b+X) + aPFD_o \exp\left(-\frac{aXl}{b+X}\right)} - R_X. \quad (9)$$

Integrating Eq. (9) gives the apparent photosynthetic activity, which is an experimentally measurable term:

$$P_X(X,L) = \frac{1}{L}\int_0^L P'_X(X,l)dl = \frac{P_m(b+X)}{aXL}\ln\frac{aPFD_o + K(b+X)}{aPFD_o\exp\left(-\frac{aXL}{b+X}\right) + K(b+X)} - R_X. \quad (10)$$

It can be noted that the final model [Eqs. (1), (5), (7), and (10)] is quite different according to the hypotheses adopted as a starting point for the model derivation.

MATERIALS AND METHODS

Microalgal Strain and Culture

The green microalga *Chlorella vulgaris* UTEX 259 obtained from the Culture Collection of Algae at the University of Texas, Austin (Starr and Jeikus, 1993) was used as the model microalga in this study. The algae was cultivated in 0.25-L flasks with 0.1 L of sterilized nitrate-enriched medium (Yun and Park, 2001), which contained 2× higher concentration of potassium nitrate than the original N8 medium (Vonshak, 1986), in order to avoid nitrogen limitation in the high-density culture (Yun and Park, 1997b). The culture flask was agitated on a shaker at 150 rpm with air bubbling. Light was supplied continuously at 200 ± 50 $\mu E\ m^{-2}\ s^{-1}$ on average with twelve 20-W warm-white fluorescent tubes (Korea General Electric Co., Seoul, Korea) and temperature was controlled at 27°C. Subcultures were removed daily and replaced with 50% of culture broth with the fresh medium. The concentration of *C. vulgaris* before replacing the medium was maintained around 1.8 kg dry weight m^{-3}. The corresponding doubling time was estimated to be approximately 33 h.

Preparation of Microalgal Suspensions

The cultured algal cells were centrifuged at 3,000*g* for 15 min at room temperature and then washed with fresh medium. After three cycles of centrifugation and washing, the suspension was diluted in series to prepare various concentrations of algal suspension using the fresh medium. Dry cell weight was measured by drying 5 mL of the suspension at 90°C in a drying oven for 24 h after being filtered through a pre-dried and pre-weighed 0.45-μm cellulose nitrate membrane filter (Whatman, Ann Arbor, MI). The algal suspensions with different cell concentrations were shaken for 2 h at 150 rpm, 27°C, in dark conditions in order to remove any residual effects of previously exposed light, during which the dissolved oxygen was lowered due to respiration. The resulting suspensions were used for measuring the oxygen production rate.

Quantification of Light Attenuation

In order to evaluate the validity of the models for the photosynthetic activity, the light attenuation in microalgal suspensions was precisely described using a modified Beer–Lambert equation [Eq. (2)]. The parameters, a and b, in Eq. (2), were estimated from the attenuation coefficients at different cell concentrations. The attenuation coefficients were calculated from the absorption spectra of *C. vulgaris* and spectral irradiance of light source (Fig. 3). The absorption spectra were measured in a rectangular cuvette of 1-cm path length by using a spectrophotometer (UV-2401, Shimadzu, Japan). The spectral irradiance of the light source was analyzed using spectrometer (1000M, SPEX, Edison) and photomultiplier tube (R928, Hamamatsu, Japan). The methods for determining parameters were described in detail previously (Yun and Park, 2001).

Photosynthetic Activity Measurement

(1) Measurement System

The photosynthetic activity measurement system was designed and constructed as shown in Figure 2. The reaction vessel was double-jacket cylinder made of Pyrex glass. Magnetic stirring with a small bar (0.5 cm in length) in the

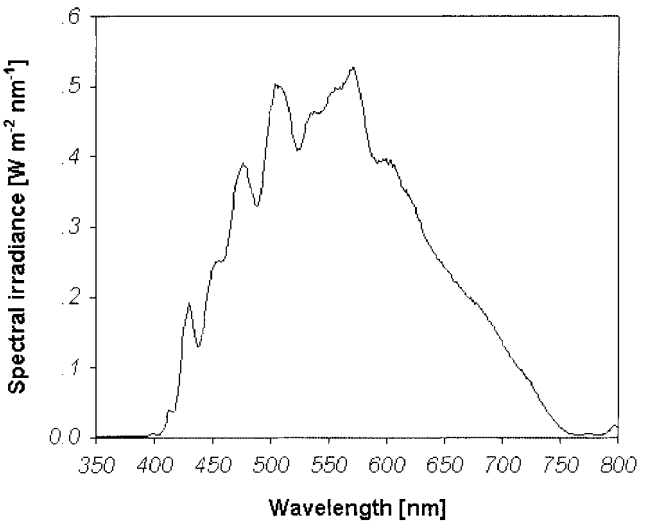

Figure 3. Optical property of light obtained by using a daylight-simulating filter.

vessel made the microalgal suspensions homogeneous. The working volume of the reaction vessel (photobioreactor) was 3.58 mL, and the light path length was 1.8 cm. Since the microalgal photosynthesis is known to be temperature sensitive, cooling water of 27°C was continuously circulated through the double jacket. The oxygen probe (Ingold, Urdorf, Switzerland) was equipped at the circular end of the vessel and used for measuring the concentration of dissolved oxygen (DO) produced by the algal photosynthetic activity. The light beam was supplied from the goose-neck-like optical fiber connection to one straight side of the reaction vessel. The convex lens was equipped in the head of optical fiber connection. By adjusting the position of the lens, we could make the light beam parallel to the axial direction without dispersion. It was confirmed that the DO probe, inlet/outlet gates, and stirring bar had negligible effects on the light penetration. A quantum sensor (LI-190A, Licor, Lincoln, USA) connected with a data logger (LI-1000, Licor) was located opposite to the illumination side to measure the transmitted light. The light source was a 150-W quartz halogen lamp in the optical fiber illuminator (A3200, Dolan-Jenner, Lawrence, USA). Use of a special optical filter (FLB32-165, Dolan-Jenner) between the lamp and optical fiber connection yielded a daylight-like spectrum (Fig. 3).

(2) Measurement Method

The incident photon flux density was correlated with the scale of illuminator aperture controlling the light output. The incident photon flux density was controlled according to this correlation. The light absorption by Pyrex glass, cooling water, and pure water was negligible compared to absorption by microalgal cells. The oxygen production was measured at various incident photon flux density using previously prepared microalgal suspensions. The algal suspension was filled in the reaction vessel in dark conditions. When the light was turned on, the DO value began to increase and the linearity between the DO and time was observed just after a few minutes. The volumetric oxygen production rate was measured from the slope of the DO change with time, and the specific oxygen production rate was calculated by dividing the volumetric oxygen production rate with the cell concentration. The measurement system reproduced consistent results.

Estimation of Model Parameters

Four models [Eqs. (1), (5), (7), and (10)] have light-transfer-related parameters (*a, b*) and photosynthesis-kinetics-related parameters (P_m, K, R_X). Parameters a and b were determined by separate experiments as described previously (Yun and Park, 2001). The respiration rate (R_X) was determined to be 4.05 ± 0.94 g O_2 g^{-1} cell h^{-1} by measuring the specific oxygen consumption rate of algal suspensions in the dark. The maximum photosynthetic activity (P_m) and the half constant (K) were estimated using the Marquardt–Levenberg nonlinear regression algorithm (Marquardt, 1963) as shown in Table II.

RESULTS AND DISCUSSION

Experimental Results

Algal photosynthetic activity was measured at various incident light intensities and cell concentrations (Fig. 4). The volumetric activity increased with increasing the incident light intensity and eventually reached the maximal value (Fig. 4A). However, no light inhibition (similar to substrate inhibition in heterotrophic microbial culture) was observed. The volumetric activity was significantly affected by the algal cell concentration. As increasing the algal concentration, the maximum volumetric activity increased. Additionally, a higher density suspension required a higher light intensity for its maximal activity.

As can be seen in the photosynthesis–irradiation (PI) curve (Fig. 4B), the specific activity reached the maximal value at the high photon flux density intensity, similar to the volumetric activity. However, as the incident photon flux density increased, the maximum specific activities at different cell concentrations converged into the same value (approximately 135 g O_2 kg^{-1} h^{-1}) while the maximum volumetric activity varied with the cell concentration. The spe-

Table II. Values of estimated parameters.[a]

Models	P_m [g O_2 kg^{-1} h^{-1}]	K [μE m^{-2} s^{-1}] or [μE kg^{-1} s^{-1}]	R^2
APFD	130.4 (3.5)	15.9 (2.7)	0.84
LPFD	136.4 (1.3)	49.0 (0.4)	0.98
APAR	132.7 (2.9)	12,600 (1,630)	0.89
LPAR	135.1 (1.5)	3160 (300)	0.97

[a]Standard errors are given in parentheses.

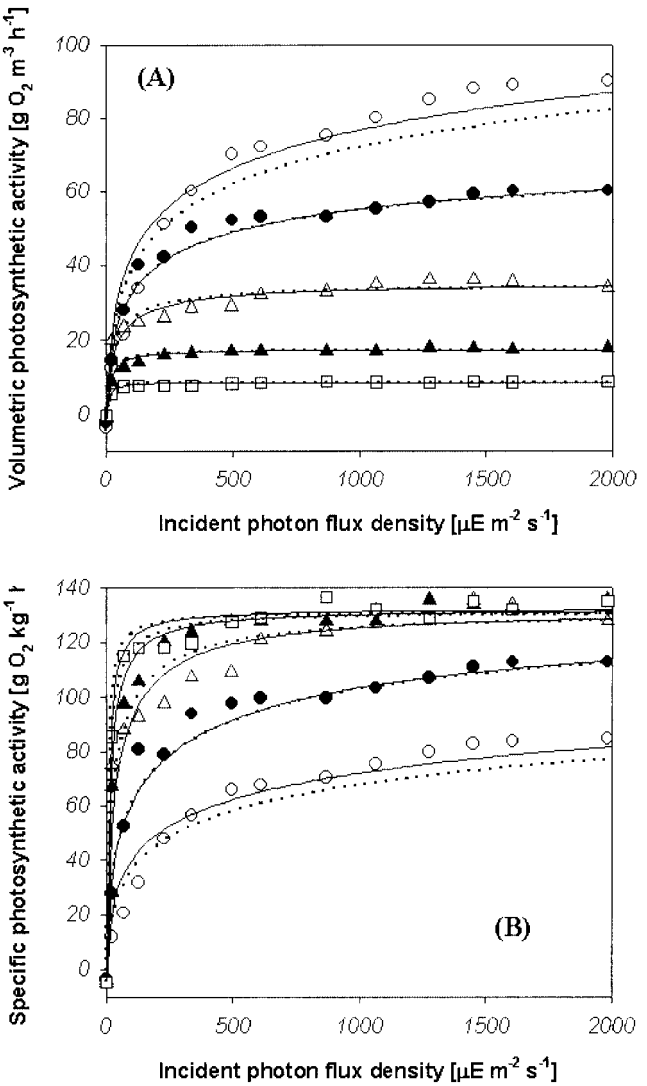

Figure 4. Volumetric photosynthetic activity (A) and the specific photosynthetic activity (B) as a function of incident photon flux density at various cell concentrations. The cell concentrations were 0.066 (□), 0.133 (▲), 0.266 (△), 0.532 (●), and 1.064 kg m^{-3} (○). The data points are average values of three replicated experimental results. The solid and dotted line represent the predicted values by the LPAR and LPFD model, respectively.

cific activity decreased as the cell concentration increased, which is opposite to the dependence of cell concentration on the volumetric activity. The volumetric and specific activities are likely to closely relate to the volumetric and specific growth rate of algal culture, respectively. As can be seen in Figure 4, high-density culture usually gives a higher volumetric productivity of biomass but does not have higher specific growth rates than low-density culture because the light is significantly attenuated at high cell concentrations (Iehana, 1983; Van Liere and Mur, 1979; Yun et al., 1999).

As can also be seen in Figure 4B, a dilute algal suspension (e.g., 0.066 kg m^{-3}) seemed to reach a light-independent state at a very low light intensity while a concentrated suspension (e.g., 1.064 kg m^{-3}) was still dependent on the light intensity even at as high as 1,000 μE m^{-2} s^{-1} of the incident photon flux density. Therefore, it should be noted that a photosynthesis–irradiance curve obtained at a fixed cell concentration could not be used for characterizing the light dependence of the algal strain.

The microalgal photosynthetic activities were largely affected both by the incident light intensity and the cell concentration. The focus of our work is on modeling such combined effects of light-related conditions upon the algal photosynthesis kinetics.

Performance of Models

In order to evaluate the validity of the models, we compared models with experimental results (Fig. 5). The estimated parameters are summarized in Table II. The APFD and APAR models were found to be poor in describing experimental results (Fig. 5A,C). Meanwhile, the LPFD and LPAR models were capable of predicting both volumetric and specific activities measured experimentally (Fig. 4A,B). Furthermore, the prediction of APFD or APAR model showed a deviation depending on cell concentration, i.e., overestimation at high cell concentrations and underestimation at low concentrations (Fig. 5A,C). Such a bias was not found in the LPFD and LPAR models (Fig. 5B,D). There was no significant difference between correlation coefficients of the LPFD and LPAR models (Table II), but, as can be seen in Figure 4, the LPAR model presented a better prediction performance than LPFD model, especially at a high algal concentration (1.064 kg m^{-3}), where the LPFD model showed a tendency to underestimate the volumetric activity (Fig. 4A) as well as the specific activity (Fig. 4B).

It should be noted that the APFD and APAR models showed the relatively poor prediction, although these models have been popularly used (Aiba, 1982; Iehana, 1983, 1990; Koizumi and Aiba, 1980; Molina Grima et al., 1994, 1997; Prokop and Erickson, 1995; Rabe and Benoit, 1962; Van Liere and Mur, 1979). Previous studies on the APFD or APAR model did not use experimental results obtained over broad ranges of incident light intensity and cell density. In fact, when we arbitrarily chose small data sets obtained from only two different cell concentrations, good fitness was found even by using the APFD or APAR model (data not shown), but the estimated parameters varied according to the data sets chosen. This implies that small sets of data obtained under a narrow range of conditions are insufficient for validating the models and determining the model parameters. The present study shows clearly that the LPFD and LPAR models provide better prediction estimates of the light-dependent microalgal photosynthesis than the APFD and APAR models.

Discussion on Validity of Models

Although the APFD and APAR models were proposed based upon empirical results, their rationale may be conceptualized as follows: since mixing is applied for algal culture in general, the algal cells move within the photo-

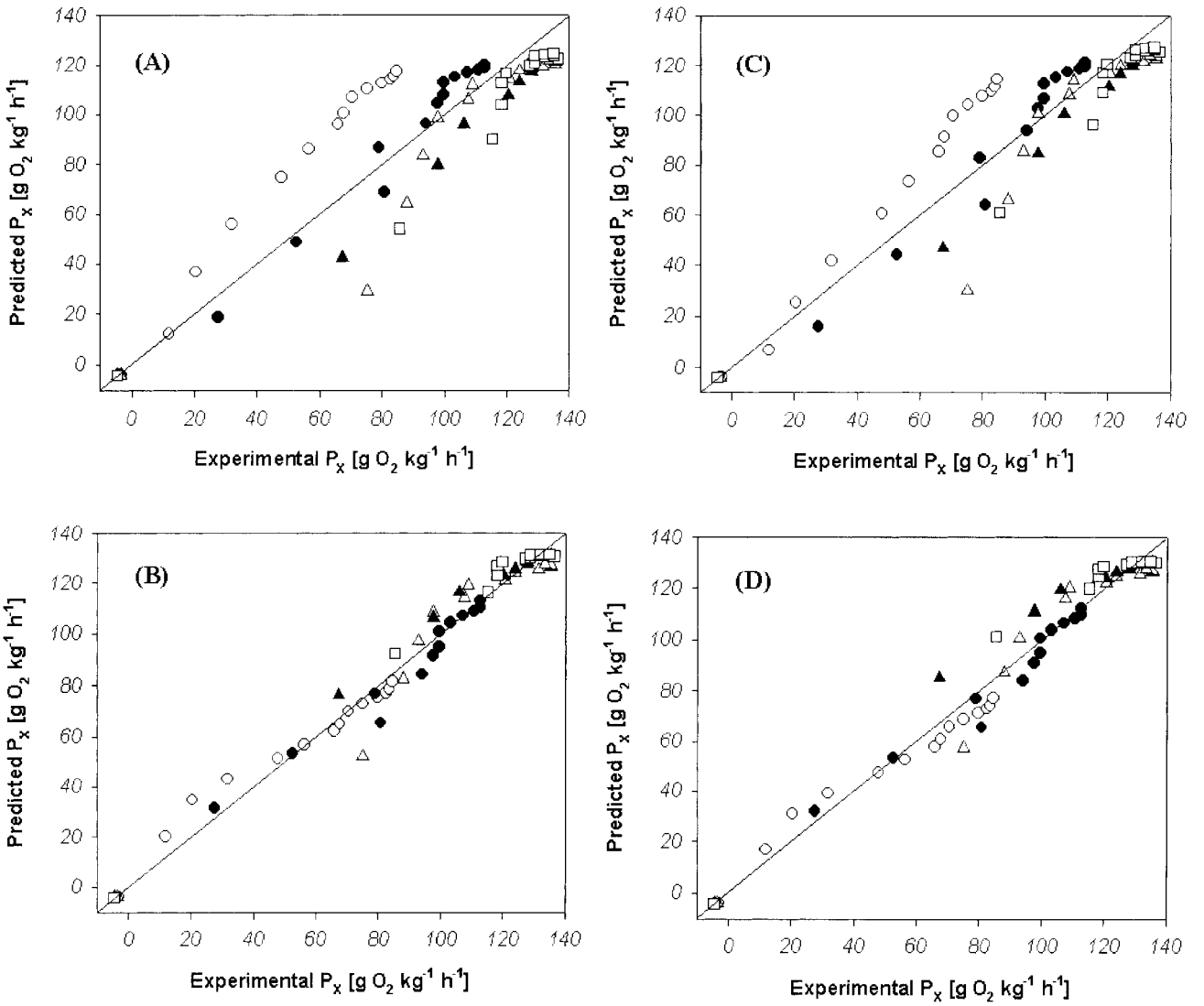

Figure 5. Comparison between experimental and predicted results of four models: APFD (A), LPFD (B), APAR (C), and LPAR (D). Cell concentrations were 0.066 (□), 0.133 (▲), 0.266 (△), 0.532 (●), and 1.064 kg m^{-3} (○).

bioreactor and the photosynthetic activity of each cell varies according to its instantaneous position (Fig. 1). However, at a time period longer than the mixing time, all cells can be considered to have the same time-averaged activity, which is the key basis of APFD and APAR models.

Meanwhile, the LPFD model (and also LPAR model) was developed from a different point of view. In a photobioreactor where light is spatially distributed (Fig. 1), a group of cells in a certain space under the same light condition has the same photosynthetic activity. Although the member of the group can be changed due to mixing, the space has the same number of cell population; in and out numbers are statistically same. Therefore, the total photosynthetic activity of entire photobioreactor can be considered as a volume-averaged value of activity of all spaces. This is the rationale of LPFD and LPAR models, which is quite different from that of APFD and APAR models.

In the LPFD and LPAR models, the algal photosynthetic activity is considered to be distributed inside the photobioreactor. Therefore, once the spatial light distribution is obtained, the photosynthetic activity can be predicted by using these models with regard to various type and size of photobioreactor. Furthermore, the LPFD and LPAR models are expected to describe the photosynthesis kinetics of a light-inhibited strain, such as *Spirulina* sp. (Vonshak and Guy, 1992). Such an algal strain is generally growth-inhibited by strong light in the region close to the illumination surface but would be light-limited if too far from the surface. Unlike APFD and APAR models, the LPFD and LPAR models could reflect this kind of position-dependent photosynthetic activity of light-inhibited strains.

Between the LPFD and LPAR models, no significant difference in the prediction performance was found except at a high cell concentration where the LPAR model was better. However, the LPAR hypothesis is likely to reflect the nature of light-dependent algal photosynthesis more reasonably than the LPFD hypothesis when the following situation is considered: for example, when the green algae are cul-

tured with a green light around 550 nm, the green cells cannot use actively the green light due to lack of pigment able to absorb this range of light. Therefore, the green light can penetrate more deeply without significant absorption (Yun and Park, 2001). In this case, although a high irradiation of green light can reach more algal cells, the algal photosynthetic activity is not expected to be high, which conflicts with the LPFD model. However, since the absorption of green light is low, the LPAR model can predict the low photosynthetic activity under the green light condition.

In this study, the local photon absorption rate was calculated regardless of light scattering to radial direction, because it was not possible to distinguish the absorption of light from the scattering. The attenuated photon flux was presumed to be absorbed by algal cells, which could result in overestimation of the photon absorption rate. Precise measurement of absorbed photon is believed to support the LPAR hypothesis more clearly, which is our on-going work.

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