

# State reconstruction of a model of microalgae growth based on one continuous-time and one discrete-time measurements

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**Abstract:** An observer for the reconstruction of the state of the phenomenological two time-scale model of the microalgae growth, so-called photosynthetic factory model formally described as a bilinear system, is designed. Three states of the model form the probability vector and while one of the states is measurable in real time the second is available only in integrated quantities. The observer designed here uses both information channels to increase precision of the reconstruction. The results are demonstrated on simulations.

*Keywords:* Nonlinear observer, photosynthetic factory, bilinear system,

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## 1. INTRODUCTION

The phenomenological two time-scale model of the microalgae growth, so-called photosynthetic factory model - further **PSF model**, was firstly presented by Eilers and Peeters, see Eilers and Peeters (1988, 1993). The PSF model describes only the most important processes, i.e. the "fast" light and dark reactions and the "slow" photoinhibition, thus it is well suited to model the growth of microalgae in biotechnological cultivation systems, especially in bioreactors where the light and flow regime causes either constant or periodic (intermittent) illumination as well. (The bioreactor is a device where a liquid - water with dispersed microbial cells (e.g. microalgae) and chemicals (nutrients)- is kept moving according to a flow pattern so as to achieve best production rate of the required product.)

Processes in the bioreactors are modelled using various types of differential equations, partial or ordinary ones. The functions solving these equations have also various meanings - they might be concentrations of various substances but also some artificially introduces quantities as in the case of PSF model, see Merchuk and Wu (2003) and references therein or also Lara-Cisneros (2012). To control the processes in the bioreactor, these functions - states of the system - must be known. However, a direct measurement is possible only in the case of a limited number of quantities. Hence the need for the observer. Design of the observer has some difficulties as the processes

are usually nonlinear having often fast and slow dynamics. Concerning a bioreactor, one sees that only a small fraction of the states of the system can be measured. This challenge originates in the physical nature of the state (which cannot be measurable at all, like ratio of cells in the process of growing) or sensors for their measurements are not available (this applies for the case when the state is a concentration of a reactant or metabolite). This, in turn, brings benefits for the control of the biological systems as more accurate information for the control is available.

The PSF model as lumped parameter model is composed from the three-state bilinear system and one integral equation. This model is presented here in detail. Concerning other approaches, let us mention the one of Su et al. (2003) where the states of another model are estimated using the extended Kalman filter.

The PSF model turns out to be bilinear. This means, one of the terms in the right-hand of the differential equations describing this system has the form  $uMx$ , where  $x$  denotes the state of the system, the variable  $u$  stands for its control and  $M$  is a square matrix of appropriate dimension. Other terms in this differential equation are linear.

Bilinear systems have a significant importance for various fields of applications. Besides other areas, they have long been useful for description of biological systems Bruni et al. (1974); Sontag et al. (2009). It is thanks to two reasons: first, they are fairly simple as they are in fact one step ahead of linear systems, yet, they can describe phenomena that do not occur in the linear systems theory. Moreover, many real biological systems, e.g. the microalgae cultures, are naturally described by bilinear systems.

In accordance to the enormous importance of bilinear systems there is a vast number of results concerning controllability, observability and identifiability of bilinear systems.

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The question of observability is of utmost interest here as not all quantities are measurable. In fact, one has to rely on measurement of some metabolites and dissolved CO<sub>2</sub> or O<sub>2</sub>, in case of heterotrophic or autotrophic microorganisms, respectively. The other variables must be calculated from the measured data. One of early papers concerning observability of bilinear systems with a special regard of biological systems is Williamson (1977). Observers for bilinear systems were proposed e.g. in Gauthier and Kazakos (1986) or Gauthier et al. (1992), the latter with special regards to bioreactors which is the direction we pursue in the presented paper as well.

In biological and biotechnological systems, not all quantities can be measured in real time. Usually, some measurement takes certain time to obtain the results as the laboratory analysis of samples might be necessary. Hence, we aim to propose an observer suited especially for reconstructing one state of the model of microalgae growth (see below), when one of the states is measurable in real time while the other one is measurable in discrete time instants only (being the time integrals over a certain period the result). Our objective is to exploit as much information about the observed process as possible. Hence we combine two different observers to achieve this goal. Demonstration of how one can extract information about this specific process (with rather difficult dynamics as the eigenvalues have different magnitudes) and how to adapt the general nonlinear observer theory to this practical problem is the contribution of our paper.

The paper is organized as follows: the phenomenological model called the "photosynthetic factory" is introduced in the second section. Third section contains the design of the observer while the results of the simulations are contained in the fourth section. The simulation contain real-world data, namely those published in Merchuk and Wu (2003). Finally, the paper is concluded.

## 2. PHOTOSYNTHETIC FACTORY MODEL

### 2.1 Original form of PSF model

Three-state PSF model, see Fig. 1, has been thoroughly studied in biotechnological literature Eilers and Peeters (1988, 1993); Kmet et al. (1993); Merchuk and Wu (2003). According to Eilers and Peeters (1988), the microalgae cells are supposed in one of three states: the resting state  $R$ , the activated state  $A$  and the inhibited state  $B$ . The model actually evaluates probabilities the "photosynthetic factory" being in the corresponding state  $R$ ,  $A$  or  $B$ . Thus the PSF model was in fact considered to be a Markovian model, hence the sum of the three states equals one. The probabilistic interpretation was later replaced, e.g. Papáček et al. (2006), the states representing now the molar fractions of microbial cells in the resting, activated and inhibited states respectively.

The state vector  $x$  of the PSF model is thus three dimensional, namely,  $x = (x_R, x_A, x_B)^T$ , where  $x_R + x_A + x_B = 1$  holds. The PSF model has to be completed by an equation connecting the hypothetical states of the PSF model with some quantity related to the cell growth. This quantity is

the specific growth rate  $\mu$ .<sup>3</sup> The rate of photosynthetic production is proportional to the number of transitions from the activated to the resting state, i.e.  $\gamma x_A(t)$ , see Fig. 1. Hence, for the average specific growth rate we have the relation:

$$\mu = \frac{\kappa\gamma}{t_f - t_0} \int_{t_0}^{t_f} x_A(t) dt, \quad (1)$$

where  $\kappa$  is a new dimensionless constant – the fifth PSF model parameter. Equation (1) reveals the reason why PSF model can successfully simulate the microalgae growth in high-frequency fluctuating light conditions: the growth is described through the "fast" state  $x_A$ , hence the sensitivity to high-frequency input fluctuations is reached, see e.g. flashing light experiments Nedbal et al. (1996).

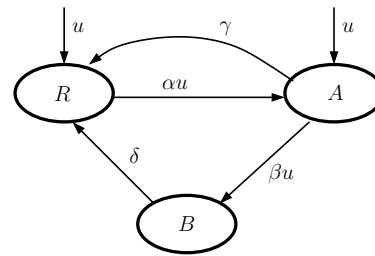


Fig. 1. Three states and four transition rates (with four PSF model parameters) of the photosynthetic factory – Eilers and Peeters PSF model.

The PSF model yields very good results, namely description of the phenomena occurring during the flashing light experiments (experimental measurements of photosynthesis during intermittent light), see e.g. Papáček (2006) where the model and its control is described into detail, together with analysis of the equations from the mathematical point of view. The main point is the investigation of the dependence of the algae growth on the frequency of the intermittent light. These experiments using the intermittent or flashing light can be also used for parameter identification, see Reháček et al. (2008). Integrals of the state  $x_A$ , according to (1), obtained for various values of the mean light intensity and frequency were measured while the same quantity was calculated using the PSF model. Both values were compared and their difference was used to adjust the value of the parameter in the PSF model. In fact, the observed quantity is the real-time value of the state  $x_A$  as their measurements - the integrals - cannot be considered to be a sufficient knowledge.

### 2.2 Reformulation of PSF model

In this paper we aim to present an algorithm for state estimation of the PSF model. The algorithm is based on the continuous measurement of the "slow" state  $x_B$  combined with discrete-time measurement of integrals of

<sup>3</sup>  $\mu := \dot{c}/c$ , where  $c$  is the microbial cell density. The notation used is the most usual in biotechnological literature, cf. Dunn et al. (1992).

the quantity  $x_A$ . This method allows us to extract most of the information contained in the measurements and, consequently, to provide accurate results. Because the "third" state  $x_R$  can be always evaluated if necessary, one can deal only with two states:  $x_A$  and  $x_B$ . For the sake of completeness, the model in reformulated form is introduced in detail. Discussion and further explanations can be also found in Čelikovský et al. (2010) and references there.

Hence using the substitution  $x_R = 1 - [x_A + x_B]$ , the reformulated PSF model has the following form:

$$\dot{x} = Ax + Bu + uDx \quad (2)$$

where the matrices in the above equation (2) are:

$$A = \begin{pmatrix} -\gamma & 0 \\ 0 & -\delta \end{pmatrix}, \quad B^\top = (\alpha \ 0)$$

$$D = \begin{pmatrix} -(\alpha + \beta) & -\alpha \\ \beta & 0 \end{pmatrix},$$

with  $x \in \mathbb{R}^2$  being the state of the system,  $x = (x_A, x_B)^\top$  and the input  $u$  is the light intensity.

Note that the control  $u$  is not a feedback control in the control-engineering sense. The nature of the process generally does not require such a feedback. Rather, the light intensity is a feedforward. One might look at this as the light intensity generator is an "exosystem" as used in the output regulation theory. But again, we do not need the information of the state of the photosynthetic factory for control purposes. That is why one does not need to ask about properties of the plant under other than periodic signals  $u$  as in practice, only these can be used as this signal. In practice, one uses sinusoidal (with a mean value added to avoid negative light intensity) or piecewise constant light intensity signals.

The paper Papáček (2006); Reháček et al. (2008); Merchuk and Wu (2003) proves that  $x_A + x_B \in [0, 1], x_A > 0, x_B > 0$ .

The species specific values of the parameters  $\alpha, \beta, \gamma, \delta$  of the PSF model has to be determine experimentally and for some microalgae strains can be found in literature, e.g. Wu and Merchuk (2001); Merchuk and Wu (2003). The reliable methodology for PSF model parameter estimation was proposed in Reháček et al. (2008); Papáček et al. (2010). Using the values published in Merchuk and Wu (2003), one the values of the constants  $\alpha, \beta, \gamma$  and  $\delta$  are as follows:

$$\alpha = 1.935 * 10^{-3} \mu E^{-1} m^2, \quad \beta = 5.785 * 10^{-7} \mu E^{-1} m^2,$$

$$\gamma = 0.146 s^{-1}, \quad \delta = 4.796 * 10^{-4} s^{-1}.$$

Using this, the matrices attain values

$$A = \begin{pmatrix} -0.1460 & 0 \\ 0 & -0.0005 \end{pmatrix} B^\top = (1.935 * 10^{-3} \ 0)$$

$$D = \begin{pmatrix} -0.0019356 & -0.001935 \\ 5.785 * 10^{-7} & 0 \end{pmatrix}$$

The input signals we are going to deal with are periodic, with minimum equal to 0, maximum at 250. Therefore, the mean value (denoted by  $\bar{u}$ , in our case 125) is nonzero. Thus, one can define the new input  $\tilde{u}$  by  $u = \bar{u} + \tilde{u}$  and, consequently, to introduce the new matrix  $\tilde{A} = A + \bar{u}D$ . Then (2) can be rewritten into the form

$$\dot{x} = \tilde{A}x + Bu + \tilde{u}Dx. \quad (3)$$

This system is observable with outputs  $C_1 = (1, 0)$  as well as  $C_2 = (0, 1)$ . This property will be essential in the following text.

The first output corresponds to the measurement of the state  $x_A$  which is possible only in discrete time instants and the results are in fact integrals over a certain time period. This quantity is in fact bounded and corresponds to the photosynthetic oxygen production rate and specific growth rate  $\mu$ , see (1), as well. Contrary to this, real-time measurement methods for the "slow" state  $x_B$  exist. Namely, this is so-called chlorophyll fluorescence measurement method which is widely used and yields fairly reliable results. In connection to PSF model, it was proposed in Wu and Merchuk (2001) in order to enhance the accuracy of PSF model parameters estimation.

### 3. OBSERVER DESIGN

The design of the continuous-time observer takes advantage from the fact that the PSF model is a bilinear system. Moreover, the input - the light intensity - is a signal with bounded magnitude. These features enable us to use existing results containing observer design for such systems. Various kinds of observers for bilinear systems were proposed (Mechmeche and Nowakowski (1997); Derese (1979)). The observer described in Mechmeche and Nowakowski (1997) proved to be most suitable thanks to its simple and straightforward implementation and also thanks to its easy-to-satisfy assumptions.

The discrete-time measurements (to be precise, the measurements of the integrals of the state  $x_A$ ) are considered to be auxiliary, thus only a linear observer was designed for them. This observer is in fact a Kalman filter for discrete-time systems.

As said above both states  $x_A$  and  $x_B$  are observable, but both in different ways. While it is in theory sufficient to reconstruct the state of the system just by using only one output, in practice it seems to be advantageous to take as much information from the measurements as possible. This leads us to the idea of designing two observers and to combine the results of both. To be precise, if the first observer estimates the state  $x(t)$  by  $\hat{x}^1(t)$  while the second one provides the estimate  $\hat{x}^2(t)$  then using  $\alpha_o \in (0, 1)$  the value

$$\hat{x}(t) = \alpha_o \hat{x}^1(t) + (1 - \alpha_o) \hat{x}^2(t)$$

is used as the estimate of the state. The parameter  $\alpha_o$  is a design parameter. There is no rule how to tune it, on the other hand, if both the continuous and the discrete-time filters are properly designed its value can be chosen to a large extent arbitrarily.

#### 3.1 Continuous observer

As the photosynthetic factory yields a bilinear system an observer for such class of systems was implemented. Among the observers for bilinear systems the one developed in Mechmeche and Nowakowski (1997) was chosen. To design this observer one has to ensure that the input of the system is bounded which is indeed the case.

For the sake of completeness the basic facts are repeated here. The observer design methodology is much more

general and can be used even for systems subjected to disturbances of various kinds. Here, the design procedure is introduced in a reduced form so that not necessary terms do not appear here.

The task is to define an observer for the system

$$\dot{x} = Ax + Bu + uDx, \quad y = Cx \quad (4)$$

where  $x \in R^n, y \in R^p$  and the matrices have appropriate dimensions.

The observer is sought in the form

$$\dot{z} = Hz + Ly + Ju + B_1uy + E_1uz, \quad \hat{x} = Mz + Py. \quad (5)$$

The approximation of the state of the system is the variable  $\hat{x}$ , the matrices must be found.

The cited paper enables to define an observer in presence of disturbances. As these are supposed to be negligible, one has  $P = 0, J = B, L = K_h^T$  (this one to be defined later).

First, define (using the matrices introduced above)

$$K_e = DC^T(CC^T)^{-1}, \quad E = D - K_eC, \quad M = K_e.$$

Then, compute

$$\gamma_o = \sqrt{2\|E^TE\|}$$

and find a matrix  $K_h$  such that the eigenvalues  $\lambda_i(H), i = 1, \dots, n$  of the matrix  $H = \tilde{A} - K_h^TC$  satisfy for all  $i$ :

$$Re(\lambda_i(H)) < 2\gamma_o.$$

Under this condition the observer is defined by

$$\dot{\hat{x}} = H\hat{x} + K_h^Ty + Bu + \tilde{u}My + Ey\hat{x}$$

For our purpose, the measurable output is the state  $x_B$ . Thus  $y = (0, 1)x$ .

### 3.2 Discrete-time observer

In this subsection another observer based on measurements in discrete time-instants is defined. We call this "discrete-time observer" as opposed to the previous case where the measurements are done continuously in spite of the fact that even in this case, the problem might be reformulated using differential equations. To design this observer, only the linearization of the system (in our case, the system without the bilinear term) was taken into account.

Let  $T \in N$ . Assume the measurement of the quantity  $y$  is done through integration over an interval of length  $T_i$ . The measured output was

$$y_T = \int_0^{T_i} x_A(T + \tau) d\tau. \quad (6)$$

First, for all  $t > 0$  one has

$$x(t + T) = \exp(At)x(T) + \int_0^t B \exp(A\tau)u(\tau) d\tau \quad (7)$$

The discrete-time observer is evaluated only at the time instants  $T \in N$ . Therefore

$$x(T + 1) = \exp(AT)x(T) + \int_0^T B \exp(A\tau)u(\tau) d\tau \quad (8)$$

The equations (6) and (8) are the key to the observer design. Using (7), (6) and by defining  $C = [0, 1]$  one infers

$$y(T) = C \left( \int_0^{T_i} \exp(At)x(T) + \int_0^t B \exp(A\tau)u(\tau) d\tau dt \right) \quad (9) \\ = CA^{-1}(\exp(AT_i) - I)x(T) + \varphi(u)$$

The term  $\varphi(u)$  contains the influence of the control. It is not necessary to express its exact form as it is not required for the observer design. In contrast, the matrix  $\Gamma = CA^{-1}(\exp(AT_i) - I)$  plays a crucial role.

As one deals with linear approximation of the system one can see the following lemma where, for simplicity of the notations, the symbol Obsv is used to denote the observability matrix:

$$\text{Obsv}(C, A) = (C^T, (CA)^T, \dots, (CA^{n-1})^T)^T$$

when matrices  $C$  and  $A$  have suitable dimensions.

**Lemma:** Assume the matrix  $\tilde{A}$  is regular. Then the asymptotic observer can be constructed if the condition

$$\text{rank}(\text{Obsv}(\Gamma, \exp(\tilde{A}T))) = n$$

is satisfied. Moreover, this condition is satisfied if

$$\text{rank}(\text{Obsv}(C, \exp(\tilde{A}T))) = n$$

The main statement follows from the way how the discrete system is defined, the second condition is due to the fact that the matrices  $\tilde{A}^{-1}(\exp(\tilde{A}T_i) - I)$  and  $\exp(\tilde{A}T)$  commute.

The robust observer was constructed using the method described in Lu (2006). A detailed description and proof of convergence can be found there. For the sake of brevity, only a simplified version of the algorithm is outlined here.

The algorithm finds an observer for the system

$$x(t + 1) = \bar{A}x(t) + \bar{B}\Phi(t, x(t), u(t)), \quad y(t) = Cx(t). \quad (10)$$

where  $x(t) \in R^n, y(t) \in R^m, \bar{A}, \bar{B}$  are matrices with appropriate dimensions and  $\Phi$  is a nonlinear function whose properties are determined later. The initial condition is  $x(0) = x_0$ .

The observer is sought in form

$$\hat{x}(t + 1) = \bar{A}\hat{x}(t) + \bar{B}\Phi(t, \hat{x}(t), u(t)) - L(y(t) - C\hat{x}(t)). \quad (11)$$

It is assumed there exist a matrix  $M_1$  and a scalar  $\alpha_1 > 0$  such that the nonlinearity  $\Phi$  satisfies the Lipschitz condition

$$\|\Phi(t, x_1, u) - \Phi(t, x_2, u)\| \leq \alpha_1 \|M_1(x_1 - x_2)\|.$$

As usual in the robust control theory, one defines the controlled output. Here, it is  $z(t) = Ce(t) = C(x(t) - \hat{x}(t))$ .

The problem is solved by finding a solution of a minimization problem described by a set of linear matrix inequalities (LMI). One has to find symmetric positive definite matrices  $P, Q \in R^{n \times n}$ , a (general) matrix  $X \in R^{n \times m}$  and a scalar  $\gamma > 0$  solving the problem

$$\begin{aligned} & \text{minimize } \gamma \\ & \text{subject to} \\ & \begin{pmatrix} -P + Q + M_1^T M_1 + C^T C & * & * & * & * \\ 0 & Q & * & * & * \\ 0 & 0 & -\eta_1 J & * & * \\ 0 & 0 & 0 & -\gamma J & * \\ PA + X\bar{C} & 0 & P\bar{B} & 0 & -P \end{pmatrix} < 0 \end{aligned}$$

The matrix is symmetric, the values of the terms denoted by  $*$  were omitted.  $\eta_1$  is a given positive constant,  $J$  denotes the identity matrix of appropriate dimension. The method can handle more general problems, so that time delays can be handled easily. The observer gain is then

$$L = P^{-1}X. \quad (12)$$

Note that the disturbance attenuation is  $\|z\|_{[0,\infty)} \leq \gamma\|w\|_{[0,\infty)}$  where  $w$  is a disturbance acting upon the system (see Lu (2006)). For simplicity, we do not consider this.

For our purpose, the matrix  $\bar{C}$  was defined as  $\bar{C} = CA^{-1}(\exp(AT_i) - I)$ ,  $\bar{A} = \exp(A * T_i)$ ,  $B = A^{-1}(\exp(A * T_i) - I)B$ . The bilinear term  $uDx$  is treated via the function  $\Phi$  and is estimated as follows: Let  $T = kT_i$  with  $k$  being an integer and  $x_1(T), x_2(T) \in R^2$ . Compute the value  $|x_1(T + T_i) - x_2(T + T_i)|$  if both are subject to the same control  $u(t)$ . Then

$$\begin{aligned} |x_1(T + T_i) - x_2(T + T_i)| &\leq |e^{AT_i}(x_1(T) - x_2(T))| \\ &+ \int_0^{T_i} |e^{A(T_i-\tau)}u(\tau)D(x_1(T + \tau) - x_2(T + \tau))|d\tau \\ &\leq \kappa_1 e^{\omega T_i} |x_1(T) - x_2(T)| \\ &+ \int_0^{T_i} \kappa_2 e^{\omega(T_i-\tau)} \max |u| |x_1(\tau) - x_2(\tau)| d\tau. \end{aligned}$$

Using the Gronwall inequality, one has for some positive constants  $\kappa_1, \kappa_2$ :

$$\begin{aligned} |x_1(T + T_i) - x_2(T + T_i)| &\leq |e^{AT_i}(x_1(T) - x_2(T))| \\ &+ \int_0^{T_i} |x_1(T) - x_2(T)| \kappa_1 \kappa_2 e^{\omega\tau} e^{\omega(T_i-\tau)} \\ &\times \max |u| e^{\kappa_2 \int_\tau^{T_i} \kappa_2} \max |u| e^{\omega(T_i-\xi)} d\xi d\tau \end{aligned}$$

Hence the nonlinearity is Lipschitz. Using the values for the system it turns out that the estimate  $M_1 = \text{diag}(0.0022, 0.0022)$  holds.

#### 4. EXAMPLE AND SIMULATIONS

The continuous-time observer of the system was designed such that values of the above introduced were:

$$\begin{aligned} K_h &= \begin{pmatrix} -0.10844 & 0.31573 \end{pmatrix} \\ H &= \begin{pmatrix} -0.388 & -0.133 \\ 0.0000723 & -0.3162 \end{pmatrix} \\ K_e = M &= \begin{pmatrix} -0.001935 \\ 0 \end{pmatrix} \\ \gamma_o &= -0.0306 \end{aligned}$$

This choice of matrices yields eigenvalues of the matrix  $H^T H$  to be equal to 0.08 and 0.19.

The discrete-time observer that uses the integrated values of the state  $x_B$  as measurements was designed as follows: one assumes the period elapsed between two measurements equals 1 while integration of the quantity  $x_B$  takes 0.1 s. This is to model that the evaluation of the measurement takes the rest of the period. The robust discrete-time filter yields the observer gain  $L_d = 10^{-3}(-0.84, 0.85)$ .

The results of the two filters were combined, the convex combination  $\hat{x} = 0.5(\hat{x}_{continuous} + \hat{x}_{discrete})$  was used.

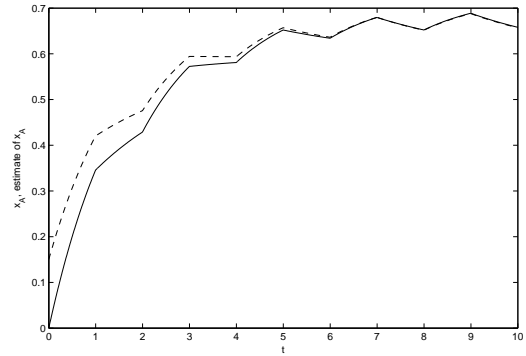


Fig. 2. The state  $x_A$

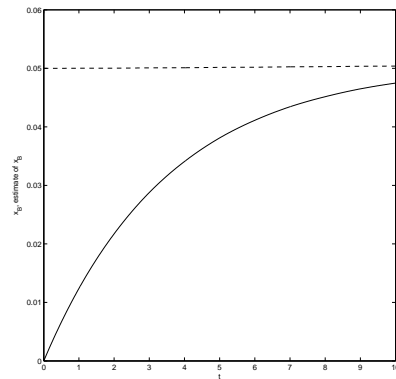


Fig. 3. The state  $x_B$

There were two input signals (light intensity)  $u$  used: The first input signal was equal to 250 on intervals  $(2k, 2k + 1)$ ,  $k \in N$  and equal to 0 on intervals  $(2k + 1, 2k + 2)$ ,  $k \in N$ .

The figures show how the fast dynamics is recovered. Note that the observer was not designed with the constraint that the states of the system need to be nonnegative and less than one. This requirement must be met by tuning the design parameters of the filter as they cannot be taken into account. Both the controllers were capable of reconstructing the dynamics fairly satisfactorily, so there is no significant difference when the parameter  $\alpha$  is changed. Due to this, we decided to keep  $\alpha = 0.5$  as mentioned above.

The last figure shows the system's behavior on large time scale. Here, one can hardly distinguish between the state of the observer and the real system. The system was fed by the rectangular signal with period 8000, amplitude varying between the values 2 in the first half of the period and 0 in the remaining half of the period. This figure was included to demonstrate the slow and fast dynamics of the PSF model.

#### 5. CONCLUSIONS

A design method for the construction of the observer for the photosynthetic factory - PSF model was proposed. The observer consists of two parts - the first one is composed of an continuous-time observer while the latter is based

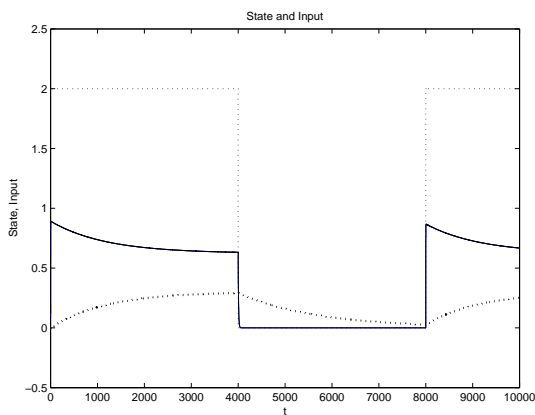


Fig. 4. The state and the input of the PSF model

on a discrete-time observer that uses the measurements of the state  $x_A$  that are available only in form of time integrals. A reliable continuous-time observer was sought out among many bilinear system observers. The results were illustrated by simulations that were carried out on a two time-scale model of PSF.

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