UNIVERSIDAD DE CHILE
FACULTAD DE CIENCIAS FÍSICAS Y MATEMÁTICAS
DEPARTAMENTO DE INGENIERÍA QUÍMICA Y BIOTECNOLOGÍA

OPTIMAL SPATIAL ARRANGEMENTS OF THREE ECOSYSTEMS: MICROALGAE POND, ANAEROBIC DIGESTER AND AEROBIC WASTEWATER TREATMENT PLANT.

MEMORIA PARA OPTAR AL TÍTULO DE INGENIERO CIVIL EN BIOTECNOLOGÍA E INGENIERO CIVIL QUÍMICO

IGNACIO FRANCISCO LÓPEZ MUÑOZ

PROFESOR GUÍA:
FELIPE DÍAZ ALVARADO

MIEMBROS DE LA COMSIÓN:
OLIVIER BERNARD
J. CRISTIAN SALGADO HERRERA

SANTIAGO DE CHILE
2016

RESUMEN DE LA MEMORIA PARA OPTAR
AL TÍTULO DE INGENIERO CIVIL EN BIOTECNOLOGÍA E INGENIERO CIVIL QUÍMICO
POR: IGNACIO FRANCISCO LÓPEZ MUÑOZ
FECHA: 14/04/2016
PROF. GUÍA: FELIPE DÍAZ ALVARADO

OPTIMAL SPATIAL ARRANGEMENTS OF THREE ECOSYSTEMS: MICROALGAE POND, ANAEROBIC DIGESTER AND AEROBIC WASTEWATER TREATMENT PLANT

Actualmente existen diversos problemas medioambientales, dos de ellos corresponden a la futura crisis energética, debido al agotamiento de los combustibles fósiles, y a la existencia de aguas residuales con altas concentraciones de nitrógeno y carbono. Un microorganismo llamado microalga podría ser una potencial solución a estos problemas, ya que éstos consumen nitrógeno y carbono para acumular lípidos en su medio intracelular, el cual posteriormente es procesado para producir biocombustibles. Sin embargo, la producción de energía a partir de microlagas es aún muy cara, es por eso que se debe optimizar este proceso, en este trabajo se abordará la estrategia de optimización a través del acoplamiento de ecosistemas, los cuales corresponden a: Piscina de microalgas (PM), digestor anaeróbico (DA) y una planta de tratamiento de aguas (PTA). El objetivo general de esta investigación es encontrar el arreglo espacial óptimo entre ellos.

Se diseñaron y calibraron modelos matemáticos simples para el ecosistema PM y PTA. La calibración se llevó a cabo usando las curvas de nitrógeno y de demanda química de oxígeno (DQO) en paralelo, con una suma de errores de 22.5% para el PM y de 38.5% para el PTA. Los parámetros obtenidos son comparables a los encontrados en la literatura. Los ecosistemas fueron acoplados a través de sus flujos de DQO y de nitrógeno, y las siguientes funciones objetivo fueron definidas: 1) Maximizar el metano producido 2) Maximizar la energía total producida y 3) Maximizar las ganacias obtenidas gracias al valor del metano y de la biomasa producida. Además, fueron consideradas restricciones medioambientales, tales como la concentración de nitrógeno y la DQO en la salida del sistema.

Si lo que se maximiza es el metano, se obtienen 99.96 $[mol]$. El reactor anaeróbico es el más grande y recibe la alimentación más alta, el CH_4 generado corresponde al 72.4% del máximo teórico. En este caso, el arreglo espacial no puede ser presentado como una cadena de etapas porque el diagrama de flujos obtenido es uno circular y por lo tanto el orden entre cada ecosistema es irrelevante para el proceso. Si la ganacia y la energía total producida son maximizadas, se obtuvo 343.6 US$ y 115.53 $[kWh]$, respectivamente. El ecosistema de tratamiento de aguas recibe la alimentación más alta, el cual produce el mayor beneficio económicos y la mayor producción de energía. El metano producido es un 21% del máximo teórico. En ambos casos, el arraglo espacial obtenido son iguales, debido a la presencia de mínimos locales y a la similtud de las funciones objetivo. Además, es posible concluir que el primer paso del proceso debiese ser el ecosistema PTA, luego el DA y finalmente el PM. Así, se pudo cumplir el objetivo general de este trabajo. Finalmente, la metodología usada es capaz de cumplir los objetivos planteados, incluso es posible escalar el problema agregando otros ecosistemas o usarla en otras aplicaciones.

THESIS ABSTRACT TO GET
DEGREE OF BIOTECHNOLOGY ENGINEER AND CHEMICAL ENGINEER
BY: IGNACIO FRANCISCO LÓPEZ MUÑOZ
DATE: 14/04/2016
TUTOR: FELIPE DÍAZ ALVARADO

OPTIMAL SPATIAL ARRANGEMENTS OF THREE ECOSYSTEMS: MICROALGAE POND, ANAEROBIC DIGESTER AND AEROBIC WASTEWATER TREATMENT PLANT

In recent years several environmental problems have come about, two of them are an energy crisis as a result of fossil fuel exhaust and the waste water created with high nitrogen and carbon concentrations. A potential solution of the aforementioned problems is contained in the properties of microalgae, which is a microorganism that can accumulate lipids in its intracellular medium. These lipids can be processed and converted into biofuel by allowing microalgae to consume nitrogen and an organic source from the medium. However, energy production from microalgae is too expensive in comparison with fossil fuel and thus there is a need to optimize this process. The strategy of optimizing by coupling ecosystems will be carried out in this work. The ecosystems that will be coupled are: Microalgae pond (MP), anaerobic digester (AD) and wastewater treatment plant (WWT). The general objective of this research is to find the optimal spatial arrangement among them through mathematical modelling.

Simple mathematical models were designed and calibrated to MP and WWT ecosystem. Calibrations were carried out using nitrogen and chemical oxygen demand (COD) curves in parallel with a sum error of 22.5% in MP and 38.5% in WWT. Obtained parameters are similar to ones found in previous literature. Ecosystem were coupled through COD and nitrogen flows and the following objectives functions were defined: 1) Maximize methane produced 2) Maximize total energy produced and 3) Maximize profit due to the value of methane and microalgae biomass produced. Environmental constraints were considered, such as nitrogen and COD because they are in the output.

When the methane produced was maximized it reached 99.96 $[mol]$. The anaerobic reactor has the biggest size, it receives the highest input flow, and CH_4 generated is the 72.4% of maximum theoretical methane production. This result determined that the spatial arrangement can not be summarized by a chain of processes since the flowsheet obtained is a circular one and thus the order is irrelevant for the process. When profit and total energy produced are maximized, it was obtained 343.6 US$ and 115.53 $[kWh]$, respectively. Waste water ecosystem received the biggest input flow, which produced the majority of amount of revenues and energy. Methane produced is equal to 21% of maximum theoretical. In both cases, the spatial arrangements obtained are equals due to the presence of local minima and the similarity in the objective function. These calculations allow to conclude the best order of the ecosystems: WWT, AD, and finally MP. Finally the methodology is enough to reach the objectives of this work, even it is possible to scale the system adding more than one ecosystem or using this methodology in other fields.

"No hay nada como viajar para ensanchar la cultura. Pero también para afinar la sensibilidad. Conocí Israel, Egipto, Túnez, Marruecos. Al final de mis viajes volví con un solo convencimiento: no somos nada."
Roberto Bolaño, Los Detectives Salvajes

Agradecimientos

En primer lugar me gustaría agradecer a los miembros de la comisión. A Olivier Bernard por la confianza de aceptarme como pasante en el INRIA, por su simpatía y por hacerse el tiempo de reunirse a pesar de lo ocupado que siempre estaba. A Felipe Díaz por su constante apoyo moral a lo largo del trabajo, sobre todo en los momentos más frustantes, y no tan sólo por su ayuda en este trabajo de tesis, sino que también por los años que hemos compartido a lo largo de mi carrera, y por apoyar mi loca idea de hacer la memoria en el extranjero. Finalmente, un agradecimiento muy grande a Francis Mairet por su paciencia, disponibilidad y simpatía cada vez que iba a tocar su puerta para hacerle preguntas sobre el trabajo, sin lugar a dudas, sin él este trabajo habría sido mucho más difícil de realizar, pero no tan sólo eso, gracias a él me pude sentir un poco más como en casa estando tan lejos.

También quiero agradecer enormemente a mi viejo amigo Jesús Salas, por su ayuda con el inglés, y a Séverine por su paciencia con mi francés.

A todos mis amigos que me acompañaron en estos años de estudios, gracias a ellos podía desconectarme semana a semana del estrés de la universidad, en especial quiero agradecer a mis amigos del colegio: Jorge, Diego, Jonathan, Benjamín y Oscar, somos amigos hace muchos años y estoy seguro que lo seguiremos siendo. A Lazcano, a los Enfermos, a mi grupo Trisus, a los Bachis, a los chicos de la rama de tenis de mesa y a los compañeros de generación IQBT 2011 y 2012, y en general a toda las personas que conocí en el departamento, en particular a Gus, Fabián, Caro Rojas, Valentina Duarte, Federico, Caro Ross, Mariana, Camila Soriano, Carmen, Angelo, Juan Carrasco, Patricio, Willy, Fanny. A los funcionarios y profesores tanto de bachillerato como de IQBT, en especial al profesor Cristián Reyes.

A las personas que gracias a la vida conocí, a mis vecinas Francisca, Catalina, a Camila y Ale por sus años de incondicional amistad, a Valeria Millán por todos los buenos momentos y por toda la contención emocional de los años que nos hemos conocido, a Marta Luzynska por todas las alegrías que hemos vivido y por las que vendrán, y por el constante apoyo a distancia, a Constanza Sadino por todas las horas de trabajo, de estrés y de risas vividas desde que entramos como pequeños mechones a la especialidad. Y a las personas que conocí viviendo en el extranjero, a Thomas, Cristina, Benny, Alba y Rosa, Elsa, Camille y David.

Finalmente, y más importante, quiero agradecer a mis padres, hermano, primo, abuela, tíos y tías, por su apoyo incondicional, por todo el cariño que me entregan día a día. Sin ellos nada, absolutamente nada de lo que he hecho hasta ahora sería una realidad. Gracias por el ejemplo de amor a la gente y el esfuerzo que debo entregar en cada una de mis tareas. Estoy infinitamente agradecido de ustedes. Los quiero.

Contents

List of Tables

List of Figures

Chapter 1

Introduction

1.1 Preliminaries

In some years there will most likely be an energy crisis and that is why we as human beings will need to think new ways to get fuels. Presently, the current burn of fossil fuels is associated to global warming. These new strategies should provide energy and take care of the environment [1], otherwise there will be energy, but no planet on which to use it.

In this context, a suitable way to solve this problem is Non-conventional Renewable Energy (NCRE). In Chile this term is used to define energy whose sources are geothermal, wind, solar, biomass, small hydro, or other similar[2]. Among them, biofuel from microalgae qualify as NCRE. Its production is detailed below.

A second problem is the waste water constantly produced by human beings through the chemical industries and domestic use. This water must be treated, as it is specified by environmental regulations. Composition of these residues is characterized by high nitrogen concentration and a high Chemical Oxygen Demand (COD)[3].

Therefore, there are two main problems, which could find potential solutions through microalgae metabolism, as it will be explained below.

1.1.1 Biofuel from microalgae

Microalgae are phototrophic microorganisms which means they use photons as a source of energy to fix carbon dioxide (photosynthetic way, in which it is possible to mitigate CO_2), but some of them can also consume carbohydrate (so they actually are mixotroph). Under certain environmental conditions this consortium is able to accumulate neutral lipids[4]. These lipids are processed, involving a transesterification reaction, and converted into biofuel.

Since microalgae also need a source of nitrogen, it is possible to think about coupling microalgae culture with wastewater, which contains nutrients [3]. As a result, the global

amount of pollution would decrease.

It is noteworthy that both problems mentioned could have solutions because of the natural microalgae metabolism. However, this source of energy is still not competitive in comparison with fossil fuels because of the high costs of nutrients to feed the microalgae and the instrumentation needed to control variables in these reactors, in order to keep the microorganisms alive. That is why current researchers are looking for different strategies to optimize this process to make this alternate source of energy more competitive. For instance, a promising alternative is to couple an anaerobic digester to a microalgae culture to produce more energy because of the methane (biogas) generated and, on the other hand, if the residues of the digester are recirculated, it is possible to recover nutrients of microalgae, achieving a cheaper production of biodiesel[5].

It could be an interesting alternative to couple ecosystems in order to optimize the production of biodiesel from microalgae, minimize costs or the global amount of residues in the environment.

The possibilities mentioned raise the question of what would happen if another ecosystem were added to the process. This idea will be detailed in the next sections, which creates new uncertainties like the increase in productivity or if the coupling arrangement could have an impact. It is noteworthy to say that until now there are no published works in this field. It means that the optimal arrangement between more than two ecosystem in order to produce biofuels and treat wastewater is an open question.

1.2 Objectives

From these questions the objectives of this work are:

1.2.1 General Objective

Proposing three simplified models and identifying optimal spatial arrangements to optimize an objective function, in the following ecosystems: a pure culture of microalgae, an anaerobic ecosystem, and an aerobic wastewater treatment plant.

1.2.2 Specifics Objectives

- Reviewing models for each ecosystem, focusing on simple flows of carbon and nitrogen.
- Proposing and/or selecting a simple model for each ecosystem.
- Identifying realistic parameter values associated with each model.
- Defining an objective function to find the optimal configuration of the coupled systems.
- Raising and solving the optimization problem to find the optimal configuration using different objectives functions.

1.3 Scopes and Limitations

As the aim of this work is to give a solution about what is the best arrangement in a simple way. In order to do this, some assumptions are used for the models: the effects of temperature, pH or pressure in the digester are omitted; all the reactors are assumed to be homogeneous.

General mass balance will be used to mathematical modelling of each ecosystem which because of assumptions taken, the permanent fluctuation in the input of many important variables, such as flows and irradiance and composition of nutrients, it will be represented by Ordinary Differential Equations (ODE).

In this work, several parameters of the model were fitted to experimental data. The results of optimal spatial arrangement will not be proved experimentally.

1.4 Methodology

The methodology was defined in order to accomplish the objectives of this work, which has five stages: 1) Designing equations of each ecosystem, using general mass balance and different growth kinetics depending on the microorganism. 2) Model calibration with experimental. 3) Coupling the equations, through the flows of carbon and nitrogen of each model. 4) Raising an objective function to optimize and 5) Optimization itself.

In the next section, it will be explained how all these points were carried out.

1.4.1 Modelling of each ecosystem

As it was mentioned before, it is important to couple different ecosystems to improve the productivity. On the other hand, each ecosystem has their own dynamics, therefore, its own equations which model that behavior.

To couple the ecosystems already described, it is necessary a representation of each ecosystem. So, the following section presents a mathematical framework to support the representation of reality and to be able to predict behavior of the system under different situations.

The mathematical representation of each ecosystem will allow to decide the best connection among them through an optimization problem. Accordingly, the mathematical model of each ecosystem has to be simple, because the aim of this modelling is to insert these equations in a problem of higher level.

1.4.2 Ecosystem: Microalgae Pond (MP)

MP will be based on microalgae culture, which could be either sophisticated culture (photo-bioreactors) or a simple open air pond (see Figure 1.1).

Figure 1.1: Microalgae raceway open ponds[6].

In any case microalgae need nutrients for their growth, such as nitrogen and COD or light to produce its own energy. This kind of metabolism is called mixotrophic metabolism, since if there is no COD, microalgae could continue growing because of the light and vice versa[7]. However, in a pond, not all microorganisms receive the same amount of light, it will depend on the depth they are (z in $[m]$) and the concentration of biomass itself (x in $[mg/L]$).

Assuming that the decay of irradiance of light (I in $[\mu mol/m^2 s]$) is exponential, so I could be modeled as follows[4]:

$$I(z) = I_0 \cdot \exp(-kxz) \tag{1.1}$$

Where I_0 is the irradiance on the surface, which usually oscillates, and k is light attenuation coefficient.

The rate of growth will be modeled as Monod kinetics:

$$\mu\left(I\right) = \mu_m \cdot \frac{I}{I + K_I} \tag{1.2}$$

Where μ_m is the maximum specific growth constant, in $[1/h]$, and K_I the Monod half saturation constant, in $[\mu mol/m^2 s]$. Considering an average of $\mu(I)$, as I is a continuous variable, its average is calculated as:

$$\overline{\mu}\left(I_0\right) = \frac{1}{L}\int_0^L \mu(I(z))dz = \frac{1}{Lkx}\int_{I(L)}^{I_0} \frac{\mu\left(I\right)}{I} dI = \frac{\mu_m}{Lkx} \ln\left(\frac{I_0 + K_I}{I_L + K_I}\right) \tag{1.3}$$

Where $I_L = I(L)$. Note that $\overline{\mu}$ is equal to the average of the yield between the growth rate and the irradiance.

As it was written before, microalgae are mixotrophic microorganisms, it means that its growth could happen because of the consumption of any carbon source, represented by COD, in $[mg/L]$. It is important to highlight that variable COD in the model is the whole organic carbon in the medium, and variable S_0 corresponds to COD without biomass. This behavior will be modeled with a Monod kinetics as well:

$$\mu(S_0) = \mu_{c,\max} \cdot \frac{S_0}{S_0 + K_{S_0}} \tag{1.4}$$

On the other hand, microalgae not only need carbon to survive, moreover, they need a source of nitrogen, which are joined in the variable N, where $N = \alpha NH_4^+ + \beta NO_3^- + \gamma NO_2^-$. If MM is the molecular mass, so:

$$\alpha = \frac{MM(N)}{MM(NH_4^+)} = \frac{14}{14 + 4 \cdot 1}, \quad \beta = \frac{MM(N)}{MM(NO_3^-)}, \quad \gamma = \frac{MM(N)}{MM(NO_2^-)} \tag{1.5}$$

Growth rate due to nitrogen consumptions is also modeled with a Monod kinetics:

$$\mu(N) = \mu_{N,\max} \cdot \frac{N}{N + K_N} \tag{1.6}$$

The last assumption taken is the volume system (V in $[L]$) is constant, that is why V will not appear in the equations. After all of this, it is possible to make a biomass balance, following the form of previous works[5]: Assuming there is no microalgae inflow in the pond and this microorganism has a constant respiration rate (R in $[1/h]$):

$$\frac{dx}{dt} = -Dx + \left(\frac{\mu_m}{Lkx} \ln\left(\frac{I_0 + K_I}{I_L + K_I} \right) + \mu_C f_C(S_0) \right) f_N(N) \cdot x - Rx \tag{1.7}$$

D is the dilution rate (in $[1/h]$) equal to Q_{in}/V, with Q_{in} the input flow, in $[L/h]$, and V is the volume of the pond, in $[L]$. So, the balance of S_0 and N are:

$$\frac{d(S_0)}{dt} = D(S_{0_{in}} - S_0) - k_C \mu_C f_C(S_0) f_N(N) \cdot x \tag{1.8}$$

$$\frac{dN}{dt} = D(N_{in} - N) - f_N(N) \left[k_N \overline{\mu}(I_0) + (k_N - k_{in} k_C) \mu_C f_c(S_0) \right] x \tag{1.9}$$

Where k_{in} is the nitrogen content in the S_0 input (later it will be explained why this term was added), considered as constant and computed from the experimental data:

$$k_{in} = \frac{1}{n}\sum_{k=1}^{n}\left[\frac{(N_{tot})_k - \alpha(NH_4)_k - \beta(NO_3)_k - \gamma(NO_2)_k}{(S_0)_k}\right]_{(in,filt)} \tag{1.10}$$

Finally, if N_{org} (in $[mg\ N/L]$) is the amount of nitrogen associated with S_0, called Organic Nitrogen.

$$\frac{dN_{org}}{dt} = D\left(N_{org,in} - N_{org}\right) - k_{in}k_C\mu_C f_C(S_0) f_N(N)x \tag{1.11}$$

1.4.3 Ecosystem: Aerobic Wastewater Treatment Plant (WWT)

In this case, there are two different microorganisms: microalgae (denoted x_1) and nitrifiers (denoted x_2). It is important to know that nitrifiers oxidize ammonium to nitrate and its source of carbon comes from inorganic carbon, it means, they do not consume S_0, and they produce NO_3^-[8]. The growth of microalgae is represented by the same model as in MP, except for the consumption of nitrogen. Actually, the inorganic nitrogen which is the easiest to uptake for the microalgae is NH_4^+, and the most difficult one is NO_3^-, this after some biochemical steps inside of the microorganism is transformed to NH_4^+. So, NO_3^- is not uptaken until NH_4^+ has a low concentration. So, mass balances are modeled as follows:

$$\frac{dx_1}{dt} = -Dx_1 + \left(\overline{\mu}\left(I_0\right) + \mu_C f_c(S_0)\right)\cdot\left(f_{NH,1} + f_{NO}\right)x_1 - R_1x_1 \tag{1.12}$$

Where

$$f_{NH,1}\left(NH_4^+\right) = \frac{NH_4^+}{NH_4 + K_{NH_4,1}}$$

$$f_{NO}\left(NO, NH_4^+\right) = \frac{NO_3^-}{NO_3^- + K_{NO_3}}\cdot\frac{K_{NH_4,1}}{NH_4^+ + K_{NH_4,1}}$$

$$\frac{dx_2}{dt} = -Dx_2 + \mu_{NH,2}f_{NH,2}x_2 - R_2x_2 \tag{1.13}$$

Where:

$$f_{NH,2}\left(NH_4^+\right) = \frac{NH_4^+}{NH_4^+ + K_{NH_4,2}}$$

S_0 balance (COD without biomass):

$$\frac{d(S_0)}{dt} = D\left(S_{0_{in}} - S_0\right) - k_{C1}\mu_C f_C(S_0)\cdot\left(f_{NH,1} + f_{NO}\right)\cdot x_1 \tag{1.14}$$

NH_4^+ balance: Where $(NH_4^+)_{model} = \alpha NH_4^+$ (in $[mg\ N/mg\ S_0]$):

$$\frac{d(NH_4)}{dt} = D\left(NH_{4,in} - NH_4\right) - f_{NH,1}\left[k_{N1}\overline{\mu}\left(I_0\right) + \left(k_{N1} - k_{in}\right)\mu_C f_c\right]x_1 - k_{NH,2}\mu_{NH,2}f_{NH,2}\cdot x_2 \tag{1.15}$$

NO balance: Where $NO = \beta NO_3^- + \gamma NO_2^-$.

$$\frac{d(NO)}{dt} = D\left(NO_{in} - NO\right) - f_{NO}\left[k_{N1}\overline{\mu}\left(I_0\right) + \left(k_{N1} - k_{in}k_{C1}\right)\mu_C f_c\right]x_1 + k_{NO,2}\mu_{NH,2}f_{NH,2}\cdot x_2 \tag{1.16}$$

And organic nitrogen:

$$\frac{dN_{org}}{dt} = D\left(N_{org,in} - N_{org}\right) - k_{in}k_{C1}\mu_C f_C(S_0)\cdot\left(f_{NH,1} + f_{NO}\right)x_1 \tag{1.17}$$

1.4.4 Ecosystem: Anaerobic Digester (AD)

An AD is a system where microorganisms grow in absence of oxygen. This consortium of bacteria is able to transform a source of COD, such as biomass, to biogas (mainly composed of methane and CO_2), through four reactions: hydrolysis, acidogenesis, acetogenesis and methanogenesis, each of them performed by different microorganisms[9].

The model of this ecosystem will be based on a simplified model designed by Mairet et al., 2011 (see [5]), in order to reduce computational processing time. As assumption, it will be considered only three main bacterial population: x_1 which consume sugar and lipids (S_1 in $[mg\ COD/L]$), x_2 consume proteins (S_2 in $[mg\ COD/L]$), and both produce Volatile Fatty Acids or VFA (S_3 in $[mg\ COD/L]$), and the last bacterial population, x_3, uses as substrate VFA, and produce CO_2 and methane (methanogenesis)[10].

If μ_i is the specific growth rates of x_i, in $[1/h]$, where $i = \{1, 2\}$, in this case μ_i will be modeled as a Contois functions of the corresponding substrates, it means:

$$\mu_i(S_i, x_i) = \mu_{i,\max}\cdot\frac{S_i}{S_i + K_{Si}x_i} \tag{1.18}$$

As specific growth of methanogenic bacteria (μ_3 in $[1/h]$) could suffer inhibition because of

VFA accumulation, it is necessary to model it with an inhibitory term, that is why, Haldane kinetics multiplied by an ammonia inhibition term, is used as follows:

$$\mu_3(S_3, NH_3) = \mu_{3,\max} \cdot \frac{S_3}{S_3 + K_{S3} + \frac{S_3^2}{K_{I3}}} \cdot \frac{K_{I_{NH_3}}}{K_{I_{NH_3}} + NH_3} \tag{1.19}$$

Therefore, the mass balances are:

$$\frac{dS_1}{dt} = D(\beta_1 S_{in} - S_1) - k_1 \mu_1 x_1 \tag{1.20}$$

$$\frac{dx_1}{dt} = -Dx_1 + \mu_1(S_1) \cdot x_1 \tag{1.21}$$

$$\frac{dS_2}{dt} = D(\beta_2 S_{in} - S_2) - k_5 \mu_2 x_2 \tag{1.22}$$

$$\frac{dx_2}{dt} = -Dx_2 + \mu_2(S_2) \cdot x_2 \tag{1.23}$$

$$\frac{dS_3}{dt} = -DS_3 + k_3 \mu_1 x_1 + k_6 \mu_2 x_2 - k_9 \mu_3 x_3 \tag{1.24}$$

$$\frac{dx_3}{dt} = -Dx_3 + \mu_3(S_3) \cdot x_3 \tag{1.25}$$

$$\frac{dN}{dt} = D(N_{in} - N) - k_2 \mu_1 x_1 + k_7 \mu_2 x_2 - k_{10} \mu_3 x_3 \tag{1.26}$$

$$\frac{d(qCH_4)}{dt} = k_{11} \mu_3 x_3 \cdot V \tag{1.27}$$

Where qCH_4 is the flow of methane generated in $[mol]$. This model has already been calibrated and validated on experimental data. A summary of its results could be seen in Appendix E.

1.4.5 Model Calibration

An experiment in a pilot plant of 1.9 $[m^3]$ has been carried out during 39 days with real waste water. The following measurements were made: Total Chemical Oxygen Demand (COD),

NH_4^+, and NO_3^- concentrations as a function of time. Besides the input of the following variables were measured: COD, NH_4^+, NO_3^-, irradiance and flow. Some of these data are represented in Figure 1.2 and 1.3.

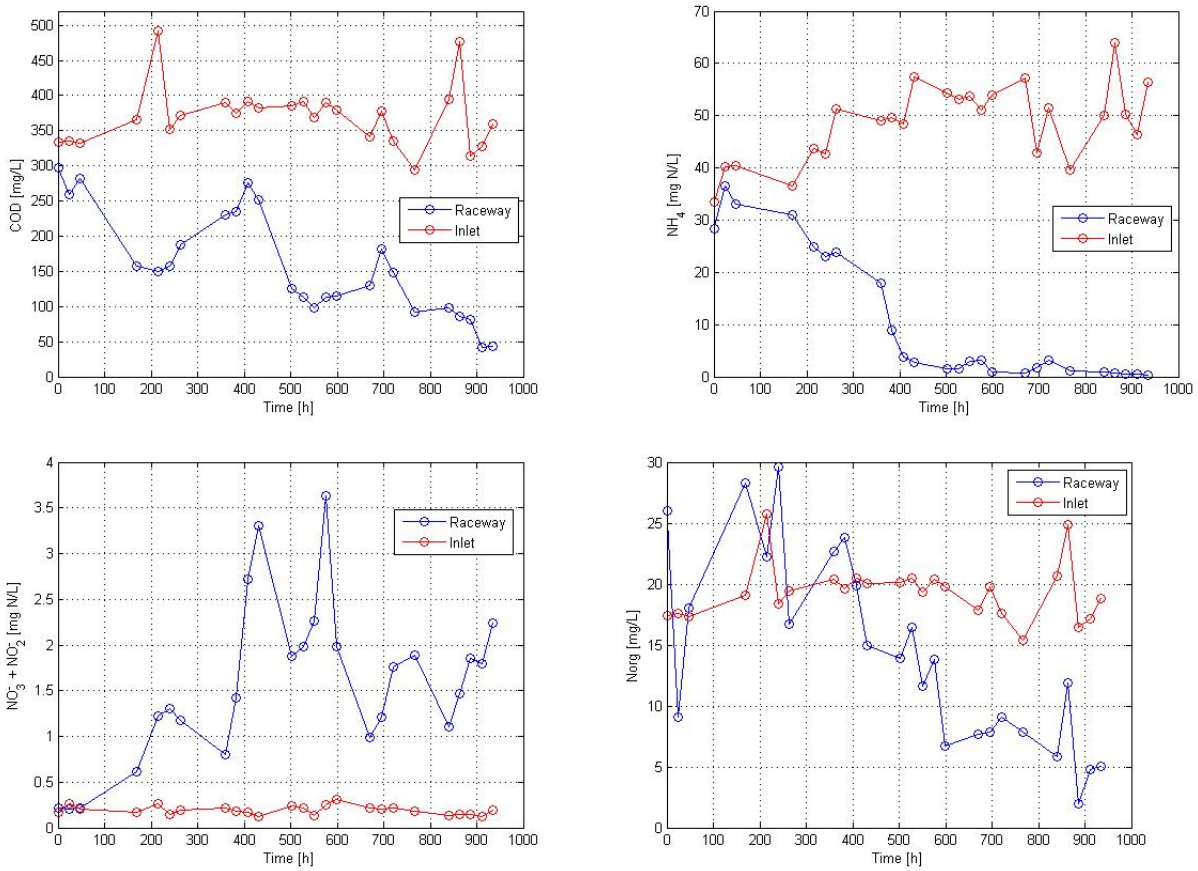

Figure 1.2: Experimental data. Concentrations of the most important variables. Red and blue line, show the concentration in the input flow, and in the bulk of the raceway, respectively.

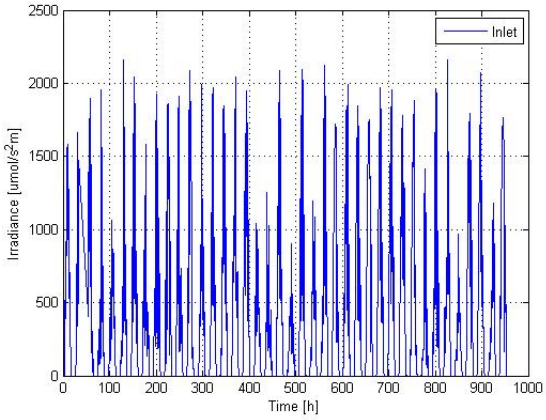

Figure 1.3: Experimental data. Irradiance measured in the raceway. With higher values when there is sunlight and zero at night.

With these experimental data, some of the kinetics parameters (the ones there were not in the literature before and the ones that are most sensitive to the environment), were identified

with a minimization algorithm: the function *fminsearch* in Matlab[11] implementing the Nelder-Mead simplex algorithm[12], which is able to handle nonlinear equations, this is an important feature because, as it was shown before, each growth kinetics does not have a linear behavior.

1.4.6 Coupling Equations

The three ecosystems will be coupled through its flow of nitrogen and carbon, where the effluent of one system will be the influent of the other one. This coupling will be useful to optimize the entire system.

It will be only explained how MP or WWT are connected with AD ecosystem. Because coupling between MP and WWT is direct due to those models have the same inputs and outputs.

If the output of MP or WWT goes to the AD, microalgae will be one of the source of sugar-lipids (S_1) and protein (S_2) (see equations (1.20) and (1.22), respectively), and moreover the output of COD from MP and WWT is also a source of S_1 and S_2 (with a different ratio). Finally, regarding to WWT ecosystem, its flow of NH_4^+ and NO will be connected to be nitrogen input in AD (equation (1.26)). In Figure 1.4 it is possible to see a summary of this coupling (between MP and AD is analogous).

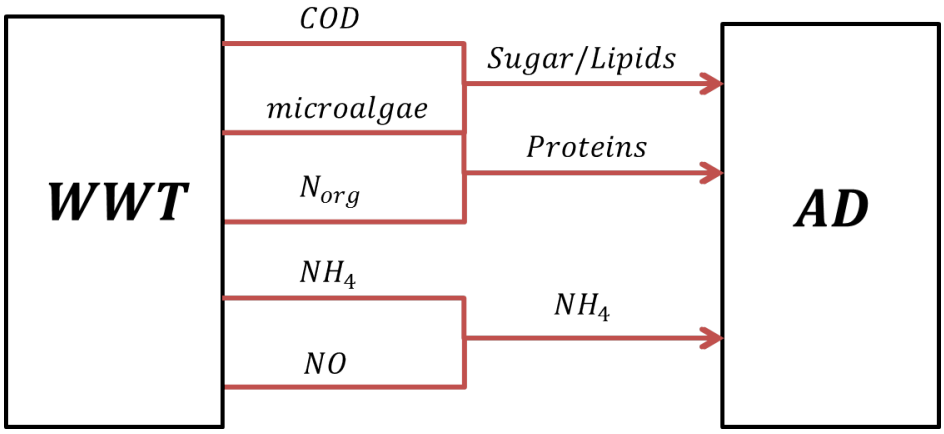

Figure 1.4: Coupling from WWT to AD ecosystems.

On the other way around, if AD feeds MP or WWT, only liquid flows are connected, for instance VFA (S_3, see equation (1.24)) will be source of COD of these ecosystems, and the nitrogen output (equation (1.26)), will be directly the source of nitrogen to MP (equation (1.9)) and in the WWT case, this nitrogen will be considered with a 100% of NH_4^+. In Figure 1.5 it is possible to see a summary of this coupling (between AD and MP is analogous).

It is possible to note that in a coupled system, each process impacts the other ones. Therefore, the manipulation of spatial arrangements is also as a strategy of optimization, the one will be studied in this work.

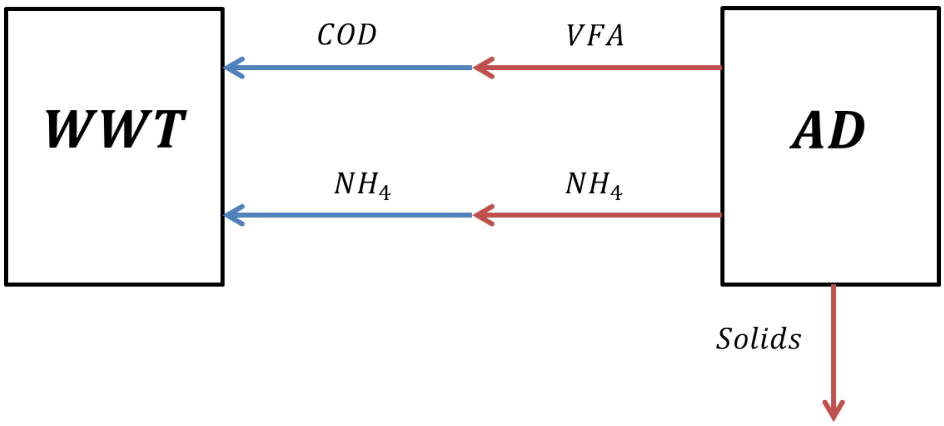

Figure 1.5: Coupling from WWT to MP ecosystems.

In Appendix F it is possible to see the mathematical expression, when the system is coupled.

1.4.7 Objective Function

As it was said, different spatial arrangements could change the characteristics of the entire system. That is why it is necessary to choose an objective function in order to decide the optimal connection among these systems.

Possible objective functions are listed below:

- Maximize methane produced.
- Maximize total energy produced (*i.e.* methane and lipid).
- Maximize profit due to the value of methane and microalgae biomass produced.

1.4.8 Optimization

Optimization will be explicit, not heuristic. The strategy is to evaluate different spatial arrangements. However these arrangements could be either linear or circular. In Figure 1.6, it is possible to see all possible combinations, where F_m is the total output flow from ecosystem m, and α_{mn} represents the F_m fraction from ecosystem m to ecosystem n (α_{0n} means input flow fraction to ecosystem n).

Moreover, V_n is the reactor volume of ecosystem n. So, under certain objective, and after optimization, α_{mn} and V_n should be obtained. In other words, optimization itself will define the sizes of each reactor and the flows between the different ecosystems. Note, for example, that it is possible to get $\alpha_{ki} = \alpha_{ik} = 0$, therefore, ecosystem i and k will not be connected.

Regarding to the constraints, are listed below:

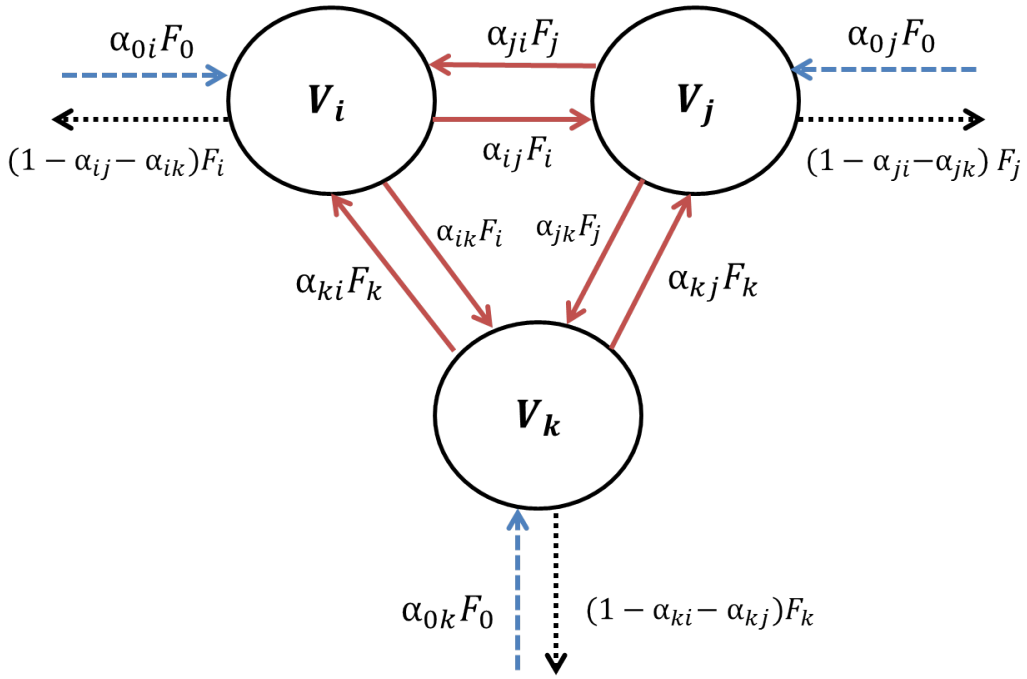

Figure 1.6: All possible arrangements. Blue (dashed line), black (dotted line) and red (continuous line) arrows are input, output and interior flows, respectively.

1. By definition: $\alpha_{mn} \in [0, 1]$ and $V_n > 0$.

2. By definition (all input flow enters to the system): $\alpha_{0i} + \alpha_{0j} + \alpha_{0k} = 1$

3. Non negatives output: $\alpha_{ij} + \alpha_{ik} \leqslant 1$; $\alpha_{ji} + \alpha_{jk} \leqslant 1$ and $\alpha_{ki} + \alpha_{kj} \leqslant 1$

4. The sum of the volume is bounded: $V_i + V_j + V_k \leqslant M$, where $M = 25 \cdot 1880\ [L]$

5. French environmental requirements: $COD_{out} < 33\ [mg/L]$, $N_{out} < 50\ [mg/L]$, and 75% of removal rate of each variable[13].

6. Mass balance in each ecosystem:
   (a) Ecosystem i: $F_i = \alpha_{0i}F_0 + \alpha_{ji}F_j + \alpha_{ki}F_k$
   (b) Ecosystem j: $F_j = \alpha_{0j}F_0 + \alpha_{ij}F_i + \alpha_{kj}F_k$
   (c) Ecosystem k: $F_k = \alpha_{0k}F_0 + \alpha_{ik}F_i + \alpha_{jk}F_j$

Optimization will be carried out with the function *fmincon* in Matlab ®[14] implementing the *interior point* algorithm [15].

Chapter 2

Results and Discussion

In this section, first of all an experimental data analysis will be made, in order to explain why total nitrogen decreases, that is why a new term (k_{in}, see equation (1.10)) and a variable (N_{org}, see equations (1.11) and (1.17)) are added to the MP and WWT models.

After this analysis, results of models designed will be shown and discussed.

2.1 Experimental Data Analysis

In Figure 2.1 it is possible to see that there is a decrease in total nitrogen and total phosphorus between the input and the raceway:

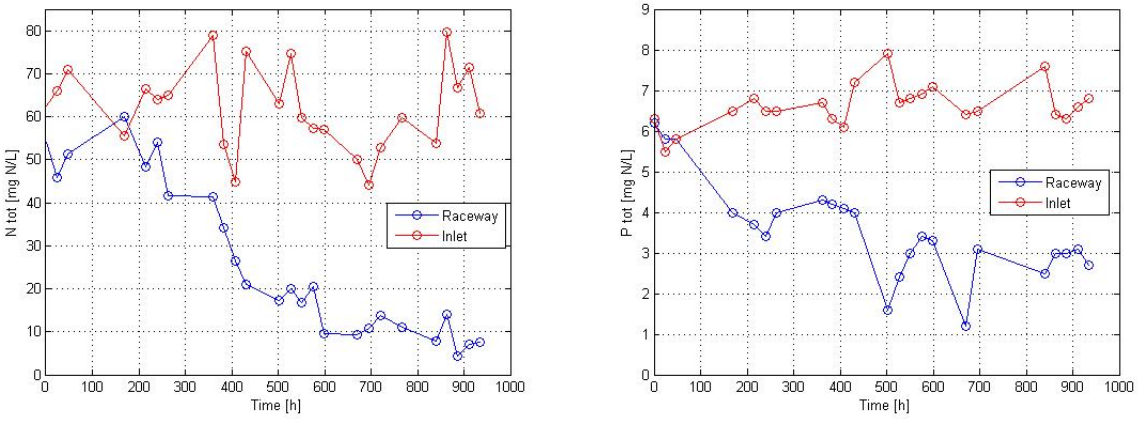

Figure 2.1: Nitrogen and phosphorus in the input and the raceway.

A data analysis was made to explain why there is a lack of total nitrogen and total phosphorus filtered. Variables φ_P and φ_N show the amount of phosphorus and nitrogen lost, respectively. They are defined as follow:

$$\varphi_z = D \cdot (z_{input} - z_{exp})_{tot} - \frac{dz}{dt} \tag{2.1}$$

Where $z = \{N, P\}$ and $D = Q/V$. Note that if there were not lack of variable z, so dz/dt (estimated using spline function of Matlab ®) would be equal to $D \cdot (z_{input} - z_{exp})_{tot}$. But results in the Figure 2.2 show it does not happen.

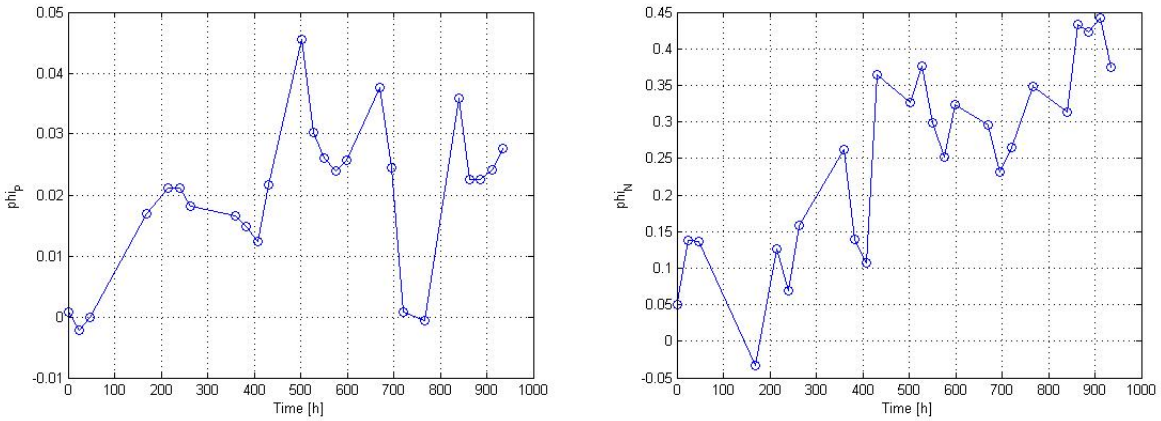

Figure 2.2: On the left φ_P and on the right φ_N

Then the ratio $r = \text{mol } \overline{\varphi_N} : \text{mol } \overline{\varphi_P}$ was calculated, and taking out the outlines, it gives as result $r = 25.5$, this value was compared with the ratio that microalgae has in its organism, and r belongs to characteristics range of mol N : mol P (between 5 and 100, according to [16]).

Hence, the lack of phosphorus and nitrogen in the medium can be due to microalgae uptake. So, it is possible that measurements are not taking into consideration this amount of nitrogen and phosphorous (maybe due to the pretreatment before measurement was not strong enough to hydrolyze the microalgae).

On the other hand, there is a lack of nitrogen in the influent flow, it means: $N_{tot,in} > (NH_4^+ + NO_3^- + NO_2^-)_{in}$, which could be explained because part of the COD is associated with molecules which also contains nitrogen, such as proteins, called before as organic nitrogen (N_{org}, see equations (1.11) and (1.17)). Parameter, k_{in}, was defined as the nitrogen content of the COD fraction, so it modifies each heterotrophic yield, its expression is shown in equation (1.10).

The value of k_{in} is approximately between 3% and 5%.

Finally, studying the concentration of inorganic nitrogen in the input and in the raceway, it is noteworthy (see Figure 2.3) that amount of ammonium decreases meantime nitrate increases (with an input of nitrate fairly constant and low).

So, it is possible to infer that there should be nitrifier microorganisms in the system, because they produce this effect, i.e consuming ammonium to produce nitrate[8].

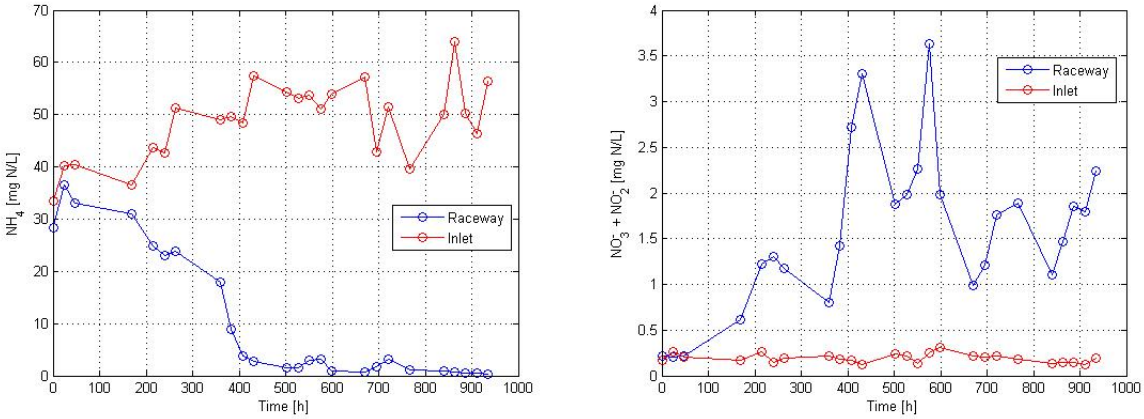

Figure 2.3: Inorganic nitrogen in the input and in the raceway

2.2 Modelling and Calibration

The same experimental data set was used to calibrate MP and WWT. For MP, the effect of bacteria was assumed negligible, considering nitrate and ammonium as a unique variable N.

For the calibration it was taken as objective function the sum of relative error in order to equate each term.

2.2.1 MP Ecosystem

Simulation results after calibration using experimental data are shown in the figures below (in Appendix A it is possible to see MP model codes).

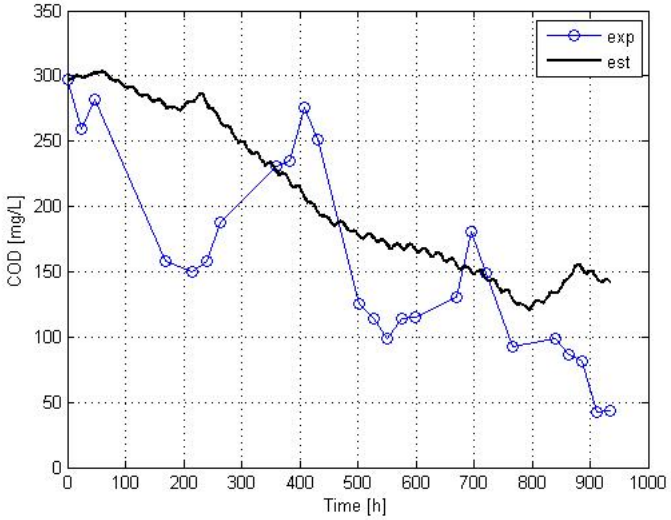

Figure 2.4: COD experimental and estimated (MP model simulation) data.

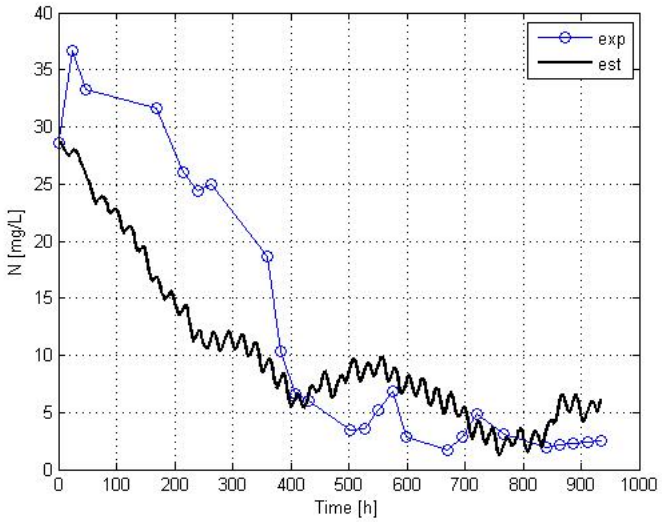

Figure 2.5: N experimental and estimated (MP model simulation) data.

The oscillatory behavior of Figure 2.4 and 2.5, can be explained because of the irradiance daily fluctuation effect on microalgae.

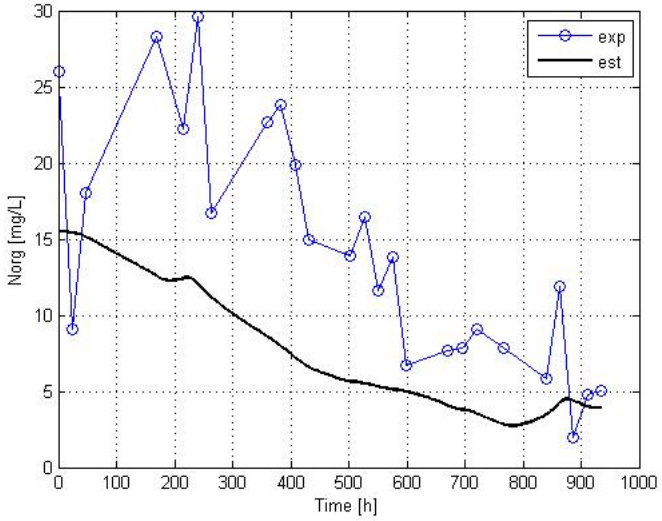

Figure 2.6: N_{org} experimental and estimated (model simulation) data.

For the calibration, it was taken as a objective function:

$$\min \left\{ \frac{1}{2} \sum_{i=1}^{n} \left(\frac{N_{est,i} - N_{exp,i}}{N_{exp,i}} \right)^2 + \frac{1}{2} \sum_{i=1}^{n} \left(\frac{(S_0 + x)_{est,i} - COD_{exp,i}}{COD_{exp,i}} \right)^2 \right\} \tag{2.2}$$

It is possible to see that N_{org} equations were not considered in the objective function of the adjustment (see equation (2.2)), because its percentage is small in comparison with all COD (around 5%), that could explain why in Figure 2.6, the estimated curve does not fit as well as the others ones.

A 22.5% of error was obtained, which is reasonable since the used simplified models are not considering some factors such as temperature, pH, alkalinity, etc., besides two curves were adjusted at the same time, so this 22.5% is the sum of two terms (see equation (2.2)). On the other hand, as usual, there could be error in the experimental data because of the instrumentation. But an important point, it is the initial condition of microalgae is unknown, and it was obtained thanks to the calibration.

It is possible to see that the model follows the same behavior of the experimental data. The model shows that the nitrogen decrease is due to microalgae consumption, and there is nitrogen linked to COD in the system.

The obtained parameters are similar to the ones found in literature. They are shown in Table 2.1. On the other hand, the experimental initial condition of microorganism biomass are unknown and it was also obtained through the minimization procedure, the result is 3.25% of experimental COD in the pond (297 $[mg/L]$ at $t = 0$) for microalgae, it means $x_0 = 3.25\% \cdot 297 = 9.65\ [mg/L]$.

Parameter	Value Adjusted	Value Bibliography	Unit	Reference
μ_m	0.0352	0.0708	$[1/h]$	[4]
K_I	130.4245	100	$[\mu mol/m^2 s]$	[4]
k	200	~ 120	$[m^2/kg]$	[17]
μ_C	0.0109	0.0887	$[1/h]$	[18]
K_{S_0}	17.0062	20	$[mg\ /L]$	[18]
R	0.0082	0.0020	$[1/h]$	[4]
K_N	0.0162	0.0100	$[mg\ /L]$	[18]
k_N	0.3935	0.0800	$[mg\ N/mg\ COD]$	[19]
k_C	3.1654	1.5000	$[mg\ COD/mg\ COD]$	[20]

Table 2.1: Parameters adjusted to the MP Ecosystem model

Finally, a sensitivity analysis of each parameter was carried out (see details in Appendix B), the most sensitive parameters are μ_m, μ_c, k_c and k_N, and the least ones are K_N and K_{S_0}, which makes sense because the first ones multiplies directly each non linear terms and the second ones are inside of them (see equations (1.8) and (1.9)).

2.2.2 WWT Ecosystem

Simulation results after calibration using experimental data are shown in the figures below (in Appendix C it is possible to see WWT model codes).

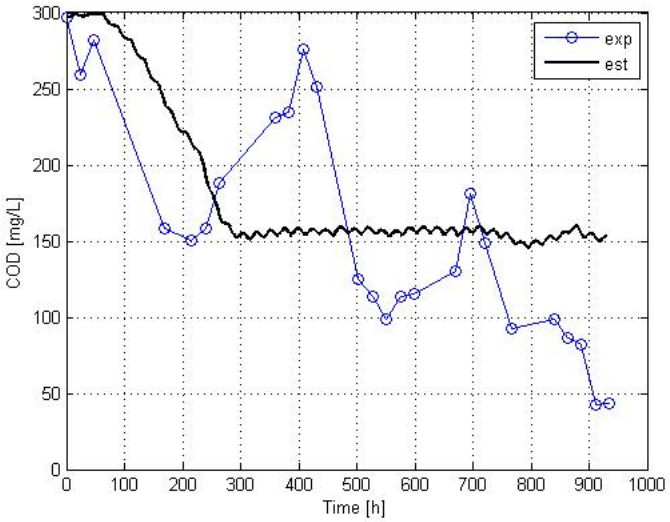

Figure 2.7: COD experimental and estimated (WWT model simulation) data.

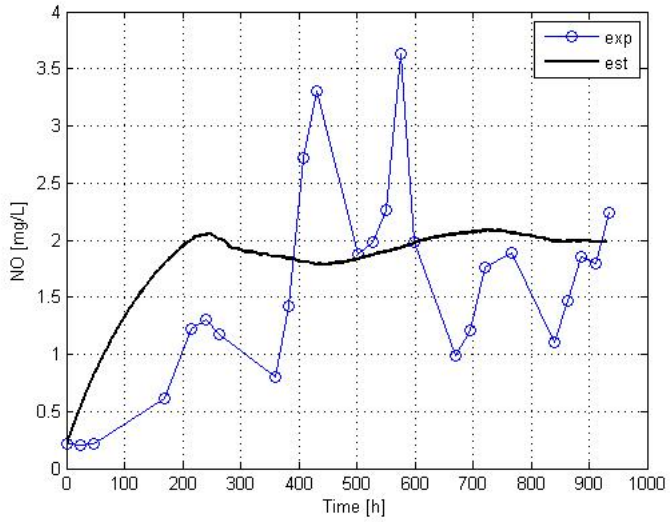

Figure 2.8: *NO* experimental and estimated (WWT model simulation) data.

In Figure 2.8, it is possible to note that this model reproduces an increase in nitrates. This feature was not possible with the MP model, because it includes only nitrate consumption and no nitrate production terms (see equation (1.9)). In fact, estimated behavior is similar to the experimental data, following in a good way the general tendency. So, according to WWT model there should be nitrifiers in the system.

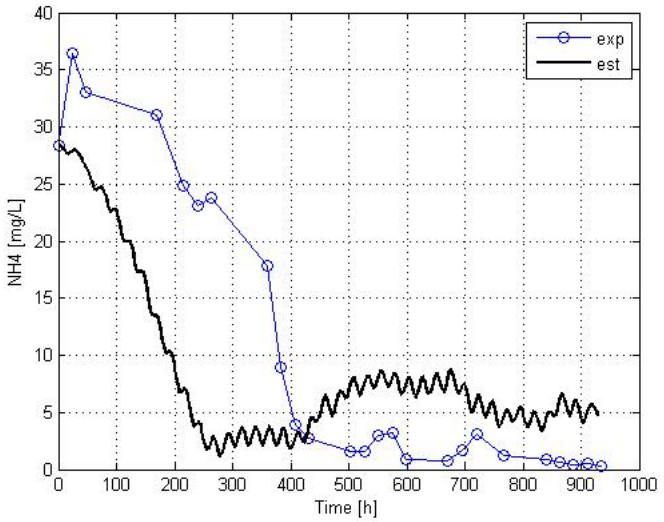

Figure 2.9: NH_4^+ experimental and estimated (WWT model simulation) data.

In Figure 2.7 and 2.9 once again it is possible to see the oscillatory behavior, which can be explained, like in MP model, because of daily fluctuation in the irradiance.

For the calibration, it was taken as objective function:

$$\min \left\{ \frac{1}{3} \sum_{i=1}^{n} \left(\frac{NH_{4_{est,i}} - NH_{4_{exp,i}}}{NH_{4_{exp,i}}} \right)^2 + \frac{1}{3} \sum_{i=1}^{n} \left(\frac{NO_{est,i} - NO_{exp,i}}{NO_{exp,i}} \right)^2 \right\} + \\ + \left\{ \frac{1}{3} \sum_{i=1}^{n} \left(\frac{(S_0 + x_1 + x_2)_{est,i} - COD_{exp,i}}{COD_{exp,i}} \right)^2 \right\}$$

An error of 38.5% was obtained which could be explained because three curves were adjusted and moreover (analogous discussion of MP ecosystem), and it is very important to say, some parameters were constrained in a range, in order to obtain a realistic model. With any restriction error was even lower than MP model, but there were non realistic parameters, that is why, a trade-off between small error and model reproducibility was considered. That is why following constrains were taken, using the function *fmincon* in Matlab ®[14]: $\mu_m > 0.02$; $k \in [120, 200]$; $k_{NH,2} > 2.0$; $k_{NH,2} - k_{NO} = 0.1$ and $k_{C1} < 5.0$.

Obtained parameters are realistic due to being similar to ones found in previous literature, they are shown in Table 2.2.

Initial condition of microorganism were also obtained through this calibration, results are 3.25% and 0.25% of experimental COD in the pound (297 $[mg/L]$) for microalgae and nitrifiers, respectively.

Finally, a sensitivity analysis of each parameter was made, the most sensitive parameters are μ_m, μ_c, k_c and k_{N1}, and the least ones are $K_{NH_4,1}$, $K_{NH_4,2}$, and $K_{NO_3,1}$ which makes sense because the first ones multiplies directly each non linear terms and the second ones are

Parameter	Value Adjusted	Value Bibliography	Unit	Reference
μ_m	0.0204	0.0708	$[1/h]$	[4]
K_I	44.4093	100	$[\mu mol/m^2 s]$	[4]
k	181.1	~ 120	$[m^2/kg]$	[17]
μ_C	0.0115	0.0887	$[1/h]$	[18]
K_{S_0}	21.3288	20	$[mg\ /L]$	[18]
R_1	0.001	0.0020	$[1/h]$	[4]
$K_{NH_4,1}$	0.0127	~ 0.01	$[mg\ /L]$	[18]
$K_{NO_3,1}$	0.0047	~ 0.01	$[mg\ /L]$	[18]
$\mu_{NH,2}$	0.009	0.0307	$[1/h]$	[21]
$K_{NH_4,2}$	1.0461	1.0000	$[mg\ /L]$	[21]
R_2	0.0006	0.0083	$[1/h]$	[21]
k_{NO}	2.2723	4.1600	$[mg\ N/mg\ COD]$	[21]
$k_{NH,2}$	2.3723	4.2460	$[mg\ N/mg\ COD]$	[21]
k_{N1}	0.3871	0.0800	$[mg\ N/mg\ COD]$	[21]
k_{C1}	3.6537	2.3300	$[mg\ COD/mg\ COD]$	[20]

Table 2.2: Parameters adjusted to the WWT Ecosystem model

inside of them (see equations (1.14), (1.15) and (1.16)). In Appendix D it is possible to see all these figures.

2.3 Arrangement Optimization

This section will be divided in three subsections, each of them describes what objective function was taken into in consideration with its results and discussions. The sections are:

- Maximization of methane produced.
- Maximization of total energy produced (*i.e.* methane and lipid).
- Maximization of profit due to the value of methane and microalgae biomass produced.

Let i, j, and k be the ecosystem indicators of MP, AD, and WWT ecosystems, respectively. Applying mass balance in each node of the graph (see Figure 1.6), the following equations system is obtained:

$$F_i = \alpha_{0i}F_0 + \alpha_{ji}F_j + \alpha_{ki}F_k \tag{2.3}$$

$$F_j = \alpha_{0j}F_0 + \alpha_{ij}F_i + \alpha_{kj}F_k \tag{2.4}$$

$$F_k = \alpha_{0k}F_0 + \alpha_{jk}F_j + \alpha_{ik}F_i \tag{2.5}$$

Where, F_n with $n = \{i, j, k\}$, is the output flow from ecosystem n in $[L/h]$, and α_{mn} represents the F_m fraction from ecosystem m to ecosystem n, so $\alpha_{mn}F_m$ is the volumetric flow from ecosystem m to n. As it is possible to see, there are 3 equations and 4 variables,

but the input flow, F_0, is known, so this equation system has a solution, where each flow will be function of F_0, and α_{mn}, as follow:

$$F_i = \varepsilon \cdot F_0, \quad F_j = \lambda \cdot F_0, \quad F_k = \gamma \cdot F_0 \tag{2.6}$$

Where

$$\gamma = \frac{(\alpha_{0j} + \alpha_{ij}\alpha_{0i})(\alpha_{jk} + \alpha_{ik}\alpha_{ji}) + (\alpha_{0k} + \alpha_{ik}\alpha_{0i})(1 - \alpha_{ij}\alpha_{ji})}{(1 - \alpha_{ik}\alpha_{ki})(1 - \alpha_{ij}\alpha_{ji}) - (\alpha_{ij}\alpha_{ki} + \alpha_{kj})(\alpha_{jk} + \alpha_{ik}\alpha_{ji})} \tag{2.7}$$

$$\varepsilon = \frac{(\alpha_{0i} + \alpha_{ki} \cdot \gamma) + (\alpha_{0j} + \alpha_{ij} \cdot \gamma)\alpha_{ji}}{1 - \alpha_{ij}\alpha_{ji}} \tag{2.8}$$

$$\lambda = \alpha_{ij} \cdot \varepsilon + \alpha_{0j} + \alpha_{kj} \cdot \gamma \tag{2.9}$$

The detailed computation to get these expressions are presented in Appendix F.

Another important definition is F_{mn}, the volumetric flow from ecosystem m to n. These values will be computed using formula (2.6) to take F_m, so $F_{mn} = \alpha_{mn}F_m$.

To limit the risk of local minima, each optimization process was initialized three times, starting from different initials conditions. For instance, if the objective function is maximization of methane produced, so one initial condition will be run, the value of the objective function will be computed, then a second initial condition will be run and so on. Finally the best obtained value is kept as the final result.

Initials conditions are shown in Table 2.3, where it is possible to see there are three different values for V_j.

Parameter	Value	Unit
α_{0i}	0.33	$[-]$
α_{ij}	0.33	$[-]$
α_{ik}	0.33	$[-]$
α_{0j}	0.34	$[-]$
α_{ji}	0.33	$[-]$
α_{jk}	0.33	$[-]$
α_{0k}	0.33	$[-]$
α_{ki}	0.33	$[-]$
α_{kj}	0.33	$[-]$
V_i	2	$[m^3]$
V_j	4, 8 and 11	$[m^3]$
V_k	2	$[m^3]$

Table 2.3: Initial condition for each optimization process.

The objective function will be computed when all the variables have reached a quasi steady-state, it means when $t_{ss} = 3000$ $[h]$ approximately (simulations were carried out until $t_f = 5000$ $[h]$).

In order to account for the daily light fluctuation, the dynamics of the system was taken into account, it means steady-state variables are not considered. Indeed, this light/dark dynamics is crucial for outdoor microalgae open ponds, and it is one of the originality of our approach. Neglecting this dynamics (considering algebraic equation instead of differential equations) would have lead to biased results.

In order to decrease processing time, the problem was slightly simplified. 1) The inputs are constant: F_0, COD_{in}, NO_{in} and $NH_{4_{in}}$. 2) I_0 it was approximated with the function: $(\max\{0; a \sin(b \cdot t + c)\})^2$, with the aim of representing daily fluctuation in the irradiance. After a parameter adjustment, this function corresponds to equation (2.10) and it is shown in Figure 2.10.

$$I_0(t) = (\max\{0; 39.8202 \cdot \sin(0.2666 \cdot t - 1.7771)\})^2 \tag{2.10}$$

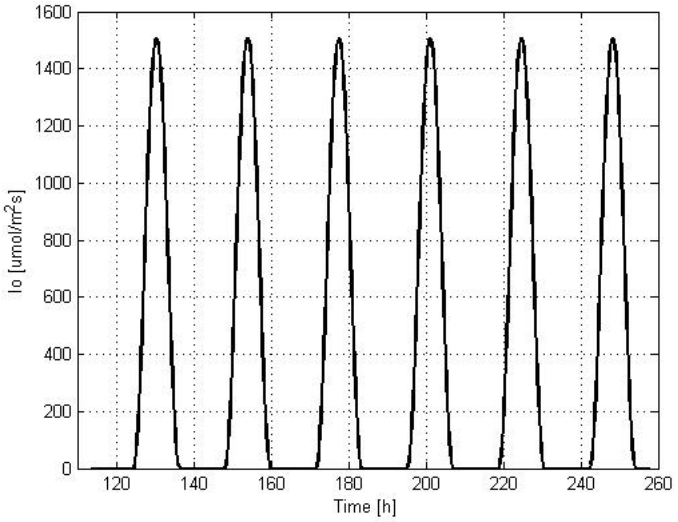

Figure 2.10: I_0 approximation. This function represents the daily fluctuation in the irradiance.

2.3.1 Maximization of Methane Produced

This objective function computes all the methane generated between $t_{ss} = 3000$ and $t_f = 5000$ $[h]$ (see equation 1.27), because methane generated is considered when quasi steady state is reached, it means since t_{ss}.

$$\max\{\text{Energy in } CH_4\} \tag{2.11}$$

Where:

$$\text{Energy in } CH_4 = [qCH_4\,(t_f) - qCH_4\,(t_{ss})] \cdot K_1 \tag{2.12}$$

As it was aforementioned, optimization will be carried out with the function *fmincon* in Matlab ®[14] implementing the *interior point* algorithm [15].

K_1 is the conversion factor to change the units of qCH_4 from a molar basis (mol) to energy (kWh). Since 1 $[ft^3]$ of natural gas contains about 1.027 $[BTU]$ [22], it means 1 $[mol\ CH_4]$ is equal to 0.254 $[kWh]$. Therefore:

$$K_1 = 0.254 \left[\frac{kWh}{mol\ CH_4}\right] \tag{2.13}$$

Results are summarized in Table 2.4. The final value of the objective function is: 25.39 $[kWh]$ produced during $t_f - t_{ss} = 2000$ $[h]$, with $V_j = 11$ $[m^3]$ in the initial condition.

Parameter	Value	Unit
α_{0i}	0.2436	$[-]$
α_{ij}	0.3601	$[-]$
α_{ik}	0.3100	$[-]$
α_{0j}	0.4323	$[-]$
α_{ji}	0.3514	$[-]$
α_{jk}	0.3572	$[-]$
α_{0k}	0.3241	$[-]$
α_{ki}	0.2898	$[-]$
α_{kj}	0.3300	$[-]$
V_i	2.0000	$[m^3]$
V_j	9.3203	$[m^3]$
V_k	3.1001	$[m^3]$

Flow	Value $[L/h]$
F_{0i}	3.1644
F_{ij}	4.2872
F_{ik}	3.6907
$F_{i,out}$	3.9276
F_{0j}	5.6156
F_{ji}	4.9834
F_{jk}	5.0657
$F_{j,out}$	4.1325
F_{0k}	4.2101
F_{ki}	3.7577
F_{kj}	4.2789
$F_{k,out}$	4.9298

Table 2.4: Optimization results. Maximization of methane in AN ecosystem

The associated arrangement is represented in Figure 2.11 as a flowsheet and in Figure 2.12 as a graph, where it is possible to note all ecosystems are connected together and fed with the input flow. This result determined that the spatial arrangement cannot be summarized by a chain of processes since the flowsheet obtained is a circular one. Which makes sense because all flows go to AD ecosystem to feed COD to generate methane, and this ecosystem feeds the other ones because these can decrease the amount of COD and nitrogen in the whole system, in order to follow the environmental constraints.

On the other hand, if $(qCH_4)_{\max}$ denotes the maximum methane mol from COD, it can be calculated as follows, with the theoretical methane yield under standard conditions: $Y = 350$ $[mL\ CH_4/g\ COD] = 1.435 \cdot 10^{-5}$ $[mol\ CH_4/mg\ COD]$ [23].

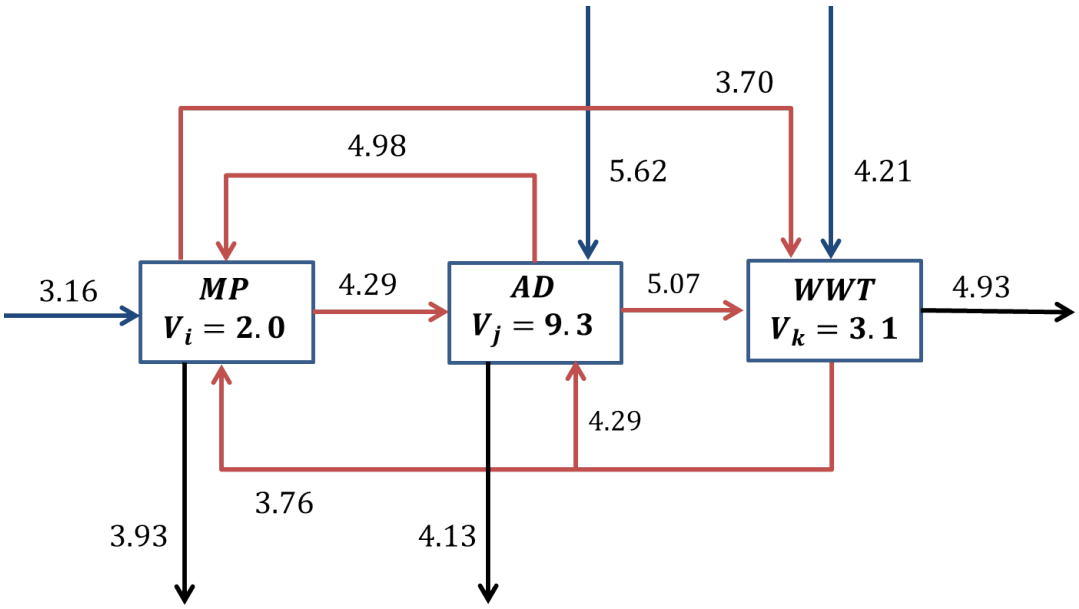

Figure 2.11: Flowsheet obtained after maximization of methane produced. Volumes are in $[m^3]$, and number above/below each arrow is value of the flow in $[L/h]$.

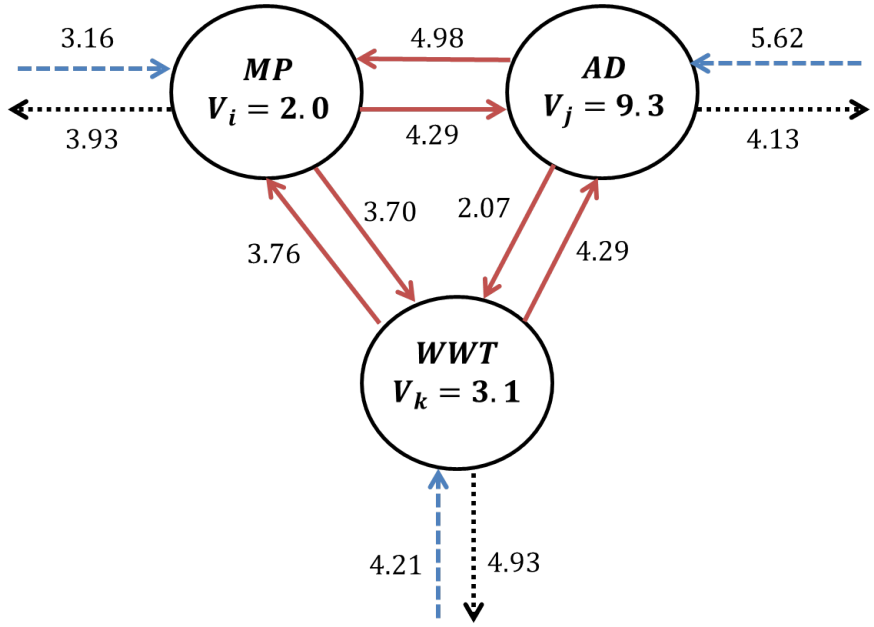

Figure 2.12: Graph obtained after maximization of methane produced. Volumes are in $[m^3]$, and number above/below each arch is value of the flow in $[L/h]$.

$$
\begin{aligned}
(qCH_4)_{\max} &= F_0 \cdot COD_{in} \cdot Y \\
&= 12.99 \left[\frac{L}{h}\right] \cdot 370.32 \left[\frac{mg\ COD}{L}\right] \cdot 1.435 \cdot 10^{-5} \left[\frac{mol\ CH_4}{mg\ COD}\right] \\
&= 0.069 \left[\frac{mol\ CH_4}{h}\right]
\end{aligned}
$$

So, in this case, it is produced 72.4% of $(qCH_4)_{\max}$. Moreover Figure 2.11 shows that the highest flow is F_{0j}, which makes sense if the goal is to produce more methane.

However it is noteworthy that F_{0i} and F_{ik} are the smallest flows, which also makes sense, because if the aim is to generate methane there is no need to feed MP ecosystem and that MP feeds WWT, because they do not have the function of producing CH_4. It was evaluated how much change the objective function is one imposes $F_{0i} = F_{ik} = 0$, it means $\alpha_{0i} = \alpha_{ik} = 0$, results are shown in Figure 2.13.

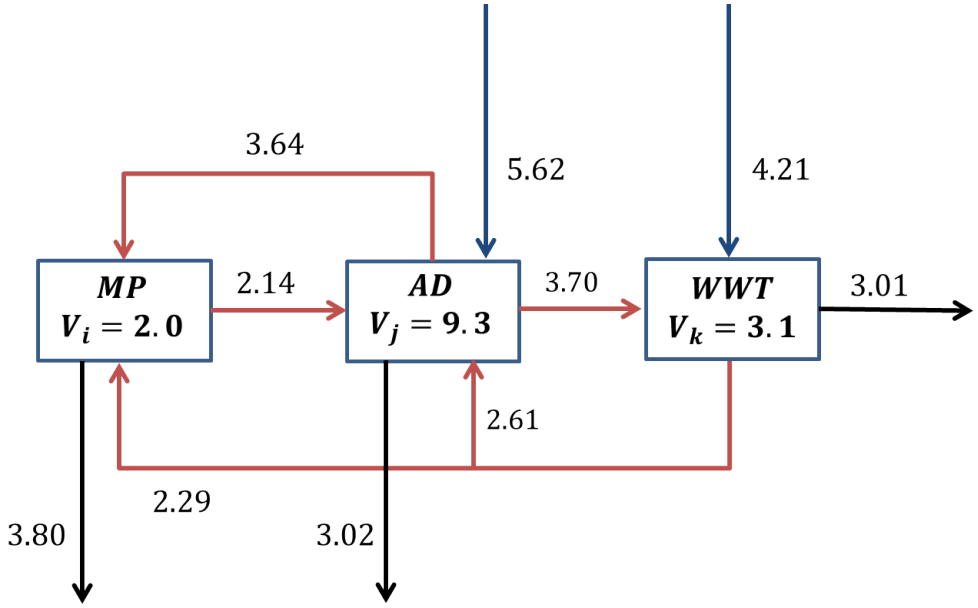

Figure 2.13: Flowsheet obtained after taking out the smallest flows in maximization of methane produced. Volumes are in $[m^3]$, and number above/below each arrow is value of the flow in $[L/h]$.

In that case, the objective function is 24.15 $[kWh]$, corresponding to decrease of 4.9%. As this decreasing is small and in order to keep a simplified system, a simplified configuration should rather deploy (Figure 2.13).

Finally, it is possible to get more energy from methane, if the COD in the input is higher, because there will be more nutrients to consume by anaerobic microorganism. It will be studied in the section: Sensitivity Analysis.

2.3.2 Maximization of Total Energy Produced

Under the objective function:

$$\max \{\text{Energy in Biomass} + \text{Energy in } CH_4\} \tag{2.14}$$

Where

$$\text{Energy in Biomass} = K_2 \cdot \sum_{t=t_{ss}}^{t_f} \left(Biomass_i \cdot F_{out,i} + Biomass_k \cdot F_{out,k}\right)_t \tag{2.15}$$

Where K_2 is a conversion factor from $[mg]$ of biomass to $[kWh]$. Energy in Biomass is calculated, assuming an oil content in microalgae of 30% by weight of dry biomass (a realistic fraction according experimental data [24]), and as 1 $[kg]$ of lipid is possible to transform in 10.5 $[kWh]$ [25], so the conversion is:

$$K_2 = 3.15 \cdot 10^{-6} \left[\frac{kWh}{mg\ microalgae}\right] \tag{2.16}$$

And this biomass is obtained from MP and WWT ecosystem. And energy from methane was already explained in equation (2.12).

Results are summarized in Table 2.5. The final value of the objective function is: 115.5 $[kWh]$, using as initial condition data shown in Table 2.3, but with $V_j = 10.5$ $[m^3]$, because with $V_j = 11$ $[m^3]$ constraints were not satisfied.

In this case, the amount of energy coming from biomass corresponds to 93.5%, so methane does not have a big influence, in fact, only 21.32% of $(qCH_4)_{\max}$ is produced. In Figure 2.14, the biggest input flow feeds to WWT ecosystem, which make sense, because this ecosystem supplies 82.2% of the energy produce.

Parameter	Value	Unit
α_{0i}	0.0012	$[-]$
α_{ij}	0.0018	$[-]$
α_{ik}	0.0029	$[-]$
α_{0j}	0.1219	$[-]$
α_{ji}	0.7959	$[-]$
α_{jk}	0.2034	$[-]$
α_{0k}	0.8769	$[-]$
α_{ki}	0.0001	$[-]$
α_{kj}	0.1373	$[-]$
V_i	1.5740	$[m^3]$
V_j	6.8290	$[m^3]$
V_k	3.9237	$[m^3]$

Flow	Value $[L/h]$
F_{0i}	0.0156
F_{ij}	0.0047
F_{ik}	0.0075
$F_{i,out}$	2.5863
F_{0j}	1.5835
F_{ji}	2.5817
F_{jk}	0.6598
$F_{j,out}$	0.0023
F_{0k}	11.3909
F_{ki}	0.0012
F_{kj}	1.6556
$F_{k,out}$	10.4014

Table 2.5: Optimization results. Maximization of total energy produced

Once again, it was evaluated what happens if one cuts the smallest flows, it means, imposing that $F_{0i} = F_{ij} = F_{ik} = F_{ki} = 0$. Results are shown in Figure 2.15 as a flowsheet and in Figure 2.16 as a graph, the objective function is equal to 115.53 $[kWh]$, it means there is a small increasing, around 0.03%, due to F_{0i} was added to F_{0k}, like this WWT ecosystem has a higher feed. Because of this and in order to keep a simplified system, it should be adviced to deploy this last arrangement.

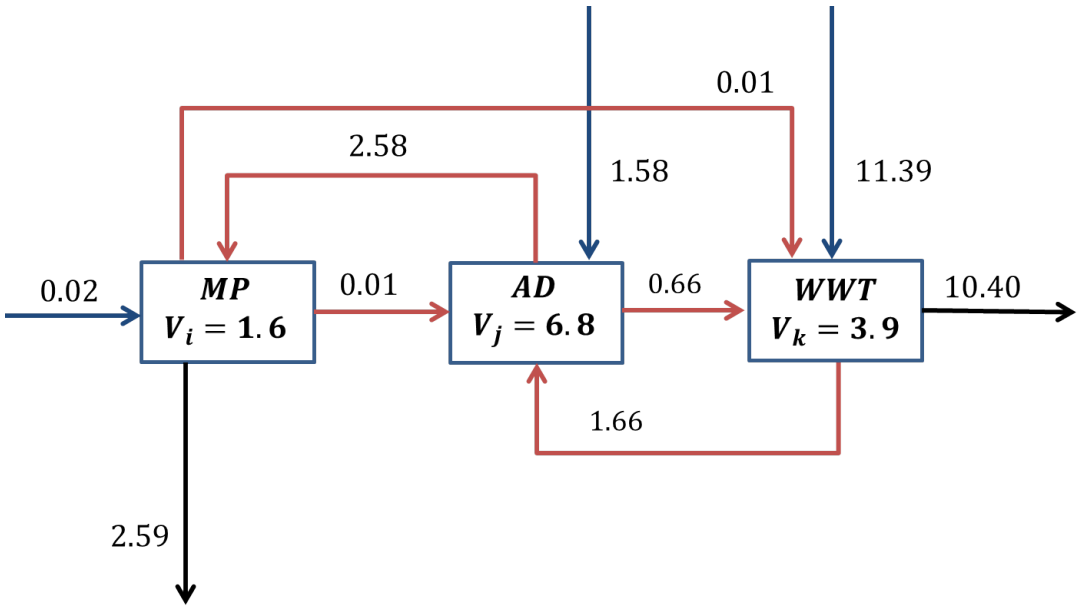

Figure 2.14: Flowsheet obtained after maximization of total energy produced. Volumes are in $[m^3]$, and number above/below each arrow is value of the flow in $[L/h]$.

In Figure 2.16 it is noteworthy only AD ecosystem and WWT ecosystem are completely connected and they are fed with the input flow, so it is possible to note the order of the ecosystems as WWT, AD, and finally MP. As a result of this and maximization of methane generated the general objective of this work was accomplished.

On the other hand, the amount of biomass that comes from autotrophic metabolism was computed, which corresponds to 54.5% of the total biomass. This is something important, because the higher is this percentage the less amount COD is necessary to grow microalgae, because in the autotrophic metabolism light is enough.

With this arrangement maybe it is possible to expect that either MP reactor or WWT reactor should be the biggest one, but it was not like that because the AD has the highest hydraulic retention time, so the size of this one has to be the largest one. It is noteworthy that in this case the AD reactor is smaller than the one obtained in methane optimization (see Figure 2.11), which also makes sense because this optimization strategy does not want to produce more CH_4 but biomass.

2.3.3 Maximization of Profit from Produced Methane and Microalgae Biomass

The following objective function will compute a trade-off between the profit due to methane and biomass. As it is shown in (2.17):

$$\max \{\text{Money from Microalgae Biomass} + \text{Money from } CH_4\} \tag{2.17}$$

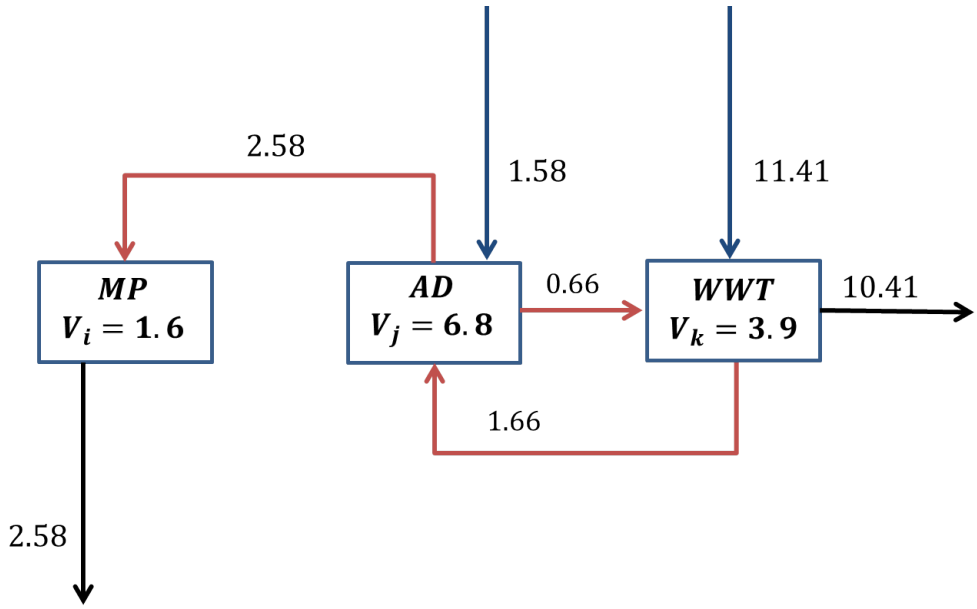

Figure 2.15: Flowsheet obtained after taking out the smallest flows in maximization of total energy produced. Volumes are in $[m^3]$, and number above/below each arrow is value of the flow in $[L/h]$.

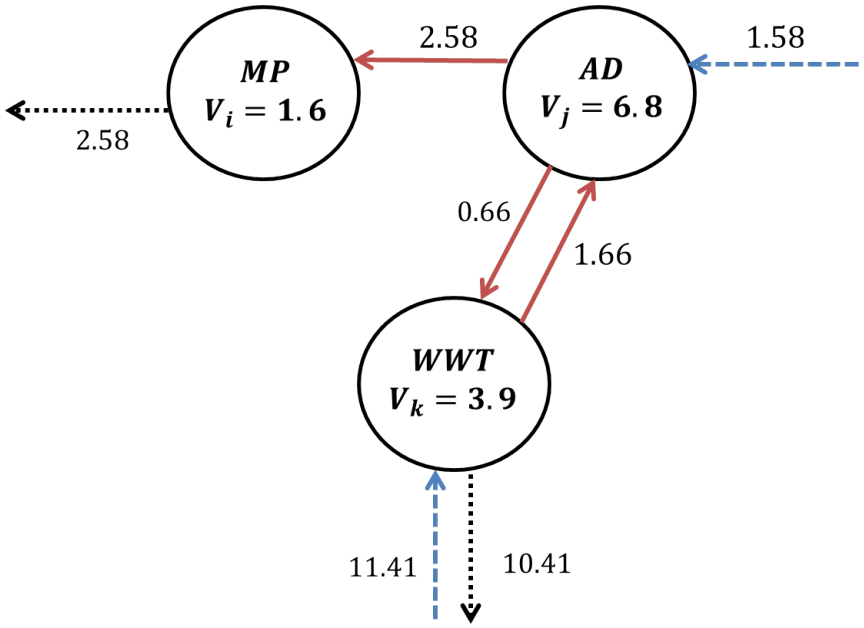

Figure 2.16: Graph obtained after taking out the smallest flows in maximization of total energy produced. Volumes are in $[m^3]$, and number above/below each arch is value of the flow in $[L/h]$.

It means:

$$\max \left\{ \left[\sum_{t=t_{ss}}^{t_f} \left(Biomass_{i,k} \cdot F_{out,i,k} \right)_t \right] \cdot K_3 + \left[qCH_4 \left(t_f \right) - qCH_4 \left(t_{ss} \right) \right] \cdot K_4 \right\} \tag{2.18}$$

Where K_3 is the conversion factor from microalgae biomass to US$[26] (value for human nutritional products):

$$K_3 = 1 \cdot 10^{-5} \left[\frac{US\$}{mg\ microalgae} \right] \tag{2.19}$$

And K_4 is the conversion factor from methane to US$[27]:

$$K_4 = 2.18 \cdot 10^{-2} \left[\frac{US\$}{mol\ CH_4} \right] \tag{2.20}$$

The value of the objective function was 343.6 US$, but the results are the same (α_{mn} and V_n) as energy optimization. To explain this fact, first of all it is necessary to understand how important is the initial condition in the final results. As the problem has several variables to optimize the space where solutions exist has too many local minima, moreover some constraints among these variables are non-linear, for instance the mass balance in ecosystem i is: $F_i = \alpha_{0i}F_0 + \alpha_{ji}F_j + \alpha_{ki}F_k$, where each F_m is a non-linear function of F_0 and α_{mn} (see equations (2.7), (2.8) and (2.9)).

Most likely all these results do not correspond to the global optimum, that is why for each objective function, different initial conditions were tried.

In this case the results of optimization from energy and methane produced were used as a initial condition (values of Table 2.3 are taken into consideration as well). The first one lead to the best results, even in each iteration α_{mn} and V_n did not change, it could mean under this objective function and with this initial condition, the arrangement optimization algorithm (see Appendix G to see the code) started to be executed from a local minima and that is why there were not changes in the variables.

Another explanation could be that objective function is similar to the optimization of total energy, only conversion factor are different. If we write the objective function (OF) explicitly as it shows in equation (2.21).

$$OF = \max \left\{ \left[\sum_{t=t_{ss}}^{t_f} \left(Biomass_{i,k} \cdot F_{out,i,k} \right)_t \right] \cdot K_m + \left[qCH_4\left(t_f\right) - qCH_4\left(t_{ss}\right) \right] \cdot K_n \right\} \tag{2.21}$$

If OF is optimize energy, the factors are (see equations (2.13) and (2.16)) shown in (2.22). On the other hand, if OF is optimize profit, the factors are (see equations (2.20) and (2.19)) shown in (2.23).

$$K_m = 3.15 \cdot 10^{-6} \left[\frac{kWh}{mg\ microalgae} \right], \quad K_n = 0.254 \left[\frac{kWh}{mol\ CH_4} \right] \tag{2.22}$$

$$K_m = 1 \cdot 10^{-5} \left[\frac{US\$}{mg\ microalgae} \right], \quad K_n = 0.0218 \left[\frac{US\$}{mol\ CH_4} \right] \tag{2.23}$$

As it is possible to see, the objective functions have the same form, with the only difference that K_m and K_n are not equals, however the product $K_m \cdot K_n$ is similar. So, these two reasons could explain why both OF have identical local minima.

2.4 Sensitivity Analysis

This section aims to study the total energy produced by the whole system, using the arrangement of Figure 2.15, when some parameters have variations.

2.4.1 Growth rates for all microorganisms (μ)

A sensitivity analysis of these parameters will be carried out, because they are sensitive to several variables, such as temperature [28]. Microalgae ponds are open to the atmosphere, so it is common to register changes in temperature, therefore there are changes in μ.

In this case, we will analyze the effect of changing all microorganism rates in a range of 30% around the calibrated values (see Table 2.2). Results are shown in Figure 2.17. We can see something expected: the higher μ, the higher energy production. Indeed, if microorganisms grow faster, more methane and biomass can be produced in less time.

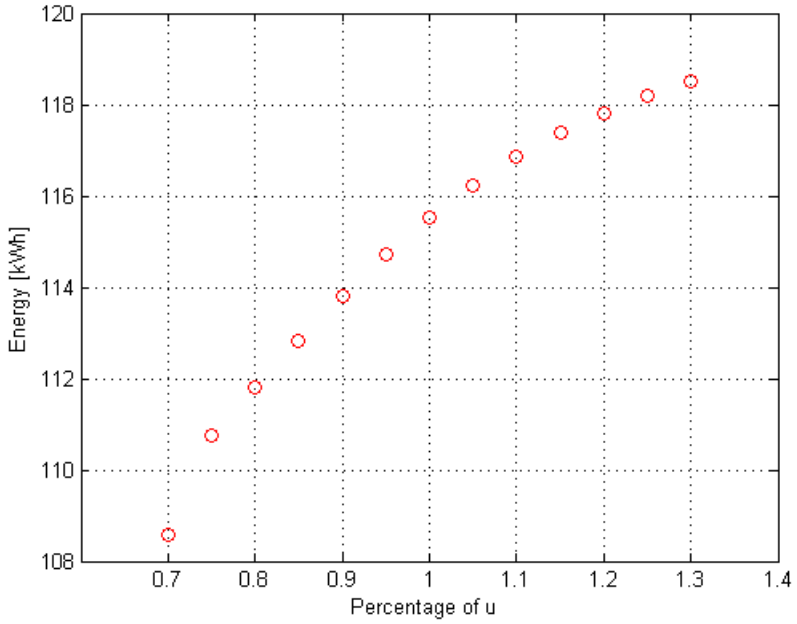

Figure 2.17: Sensitivity Analysis. Growth rates for all microorganisms.

2.4.2 μ from MP and WWT ecosystems

A sensitivity analysis of all μ parameters from MP and WWT model will be carried out (denoted as μ_{MP} and μ_{WWT}, respectively), because normally AD ecosystem are highly controlled, it means, changes in temperature, pH, pressure, etc. most of them are small, and therefore variation on the growth rate of anaerobic microorganism would be negligible.

In this case, the range taken was 50% around the calibrated values (see Table 2.2). Results are shown in Figure 2.18. We can see, once again: the higher μ in MP and WWT ecosystem, the higher energy production, but it seems that a maximum is reached at 120 $[kWh]$, when the variation is close to a 30% greater than the original value.

This effect could be explained because the higher growth rates the faster nutrients consumption, so after a while there is not enough nutrients to keep growing (growth in MP and WWT ecosystem are modelled as Monod Kinetics, see equations (1.4) and (1.6)).

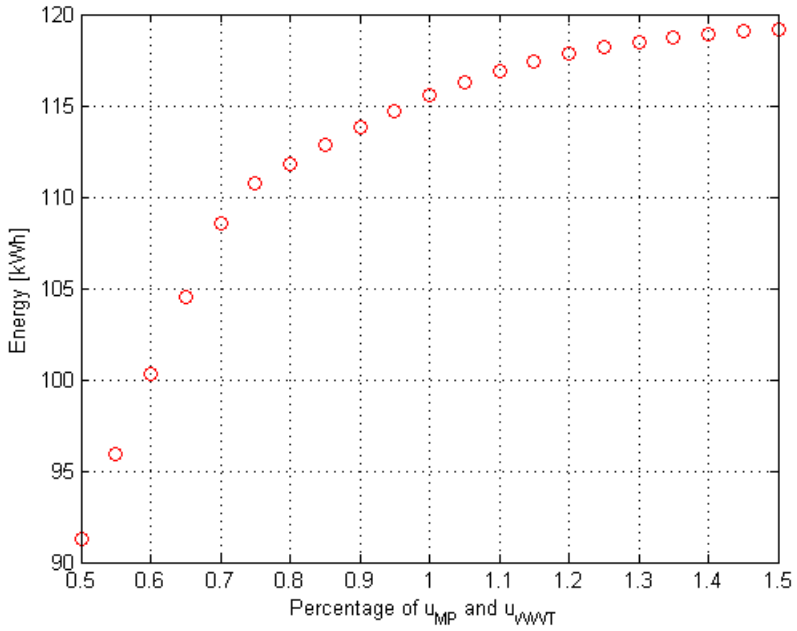

Figure 2.18: Sensitivity Analysis. Only μ from MP and WWT ecosystems

2.4.3 COD_{in} concentration

The sensitivity analysis of COD input concentration will be carried out because of the aforementioned relation between COD_{in} and the total energy produced. The upper bound for this variation is 500% higher than the original COD_{in}. Results are shown in Figure 2.19.

According to the Figure 2.19, the higher COD_{in} concentration, the higher is energy produced, as it was expected. Moreover this increase in the efficiency is due to energy from methane, because this curve grows almost parallel in comparison with the main energy curve.

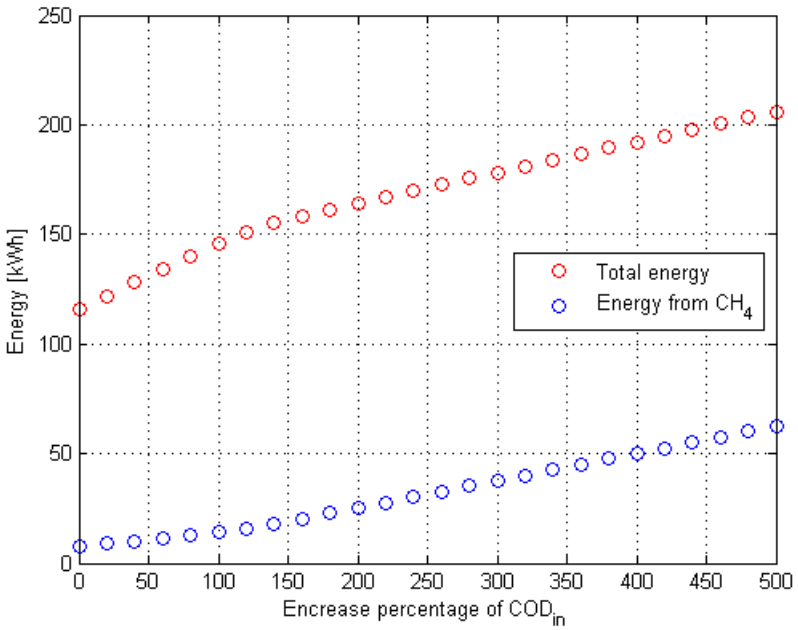

Figure 2.19: Sensitivity Analysis. COD_{in} concentration.

In Figure 2.20, it is possible to see two constraints that are not satisfied if COD_{in} increases significantly. As it was said, according to the French law: 1) It is forbidden COD output concentrations over 125 $[mg/L]$. 2) The nitrogen removal rate must be higher than 75%[13].

If COD_{in} increases over 300%, the point 1) is not respected, and when is over 260% the point 2) neither. Therefore, if COD_{in} increases over 260%, the arrangement used (see Figure 2.15) will not be able to follow with the French law. If the aim is this arrangement tolerates higher COD_{in} concentration, so it is necessary to run a new optimization.

2.5 General Discussion

Thus far three models have been utilized, two that were designed and one that was customized specifically for this problem. After this customization, the models were coupled based on their nitrogen and carbon flows. As a final step arrangement optimization was carried out, yielding a circular or linear arrangement depending on the objective function.

While it is noteworthy that the methodology used was enough to reach the stated objectives, there some unanswered questions could emerge. For instance, there could be a different way to solve this problem, but the main steps would most likely be similar to the ones presented in this thesis. These steps include designing models and then coupling them to finally carry out the arrangement optimization. Thus, the real question is related to which steps can be modified.

Moreover, the models could have been more complex to the extent that more variables, such as pH, could be used. However, that was not the aim of this work. If the arrangements

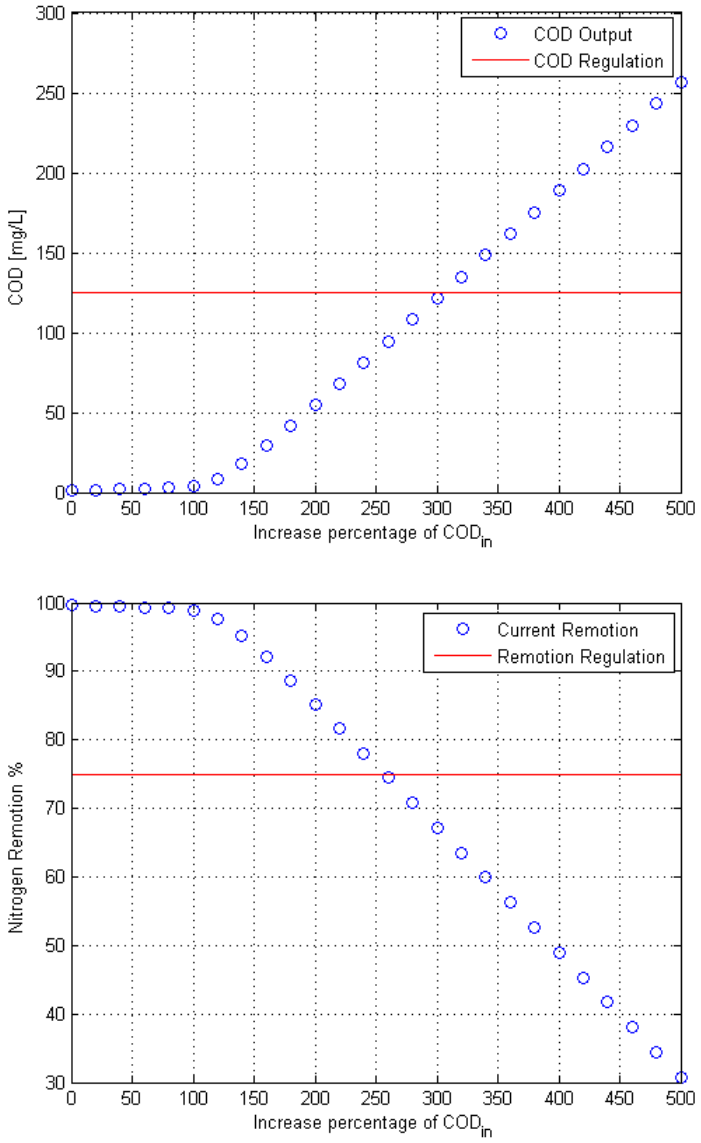

Figure 2.20: Sensitivity Analysis. *COD* and Nitrogen regulations.

obtained were deployed in the reality, in that case models would consider all those phenomena.

A strength of the methodology is that the coupling of models was carried out in a general way, thus it is possible to scale the system and add another ecosystem (or more than one), the ones could be completely different to the MP, WWT or AD, it is enough to follow the steps showed in Appendix F to accomplish it, so this work could be useful in diverse fields.

Another strength is the results of the arrangements optimization because they are consistent with the intuition under a qualitative point of view and moreover they give a quantitative solution of the problem.

A weakness of the methodology is the assumptions taken in the coupling ecosystem, even though each has a justification. It is possible to reduce the number of assumptions and design models with the same inputs and outputs. A more pertinent weakness is that there is no

guarantee that the solution found is the universal optimum. Other possible solutions will be explained below in the section Projections.

Chapter 3

Conclusions

3.1 Conclusions

One of the specific objectives of this work was to propose and/or select a simple model for microalgae pond, anaerobic digester and waste water treatment plant. This objective was accomplished and models have a good behavior with parameters comparable with literature and produced minimal errors when compared with experimental data.

According to the experimental data, it is possible to conclude that there are nitrifiers in the pond. Another conclusion is that total nitrogen decreases and is consumed by microorganisms, revealing a problem with the measurement methodology made by the kit.

Three different objective functions were defined. When methane produced is maximized, it obtained 99.96 $[mol]$. The anaerobic reactor has the biggest size, it received the highest input flow, and CH_4 generated is the 72.4% of maximum theoretical.

If the aim is to maximize the methane produced, it is possible to conclude that the spatial arrangement cannot be summarized by a chain of processes since the flowsheet obtained is a circular one, in order to feed COD to the anaerobic digester and to follow environmental requirements.

On the other hand, when profit and total energy are maximized, it obtained 343.6 US$ and 115.53 $[kWh]$, respectively. Waste water ecosystem received the biggest input flow, which produced the largest amount of revenues and energy. In this case methane produced is 21% of maximum theoretical.

If the aim is to maximize profit and total energy produced, it is possible to conclude that the first step is WWT ecosystem, then AD ecosystem and finally MP ecosystem. As a result of this finding the general objective of this work was accomplished.

A sensitivity analysis was conducted that led to the conclusion that the growth rate is positively correlated to the energy produced, but also has a maximum level of efficiency, which

is reached at 120 [kWh]. Moreover, the higher COD concentration (COD_{in}) in the input, the higher energy is produced, this is due to an increasing in the energy from methane. COD_{in} can not increase more than 260%, otherwise French environmental requirements would not be followed.

As a final point, while the methodology has weaknesses, its strengths are enough to reach the objectives of this work, even it is possible to scale the system adding more than one ecosystem or using this methodology in other fields.

3.2 Projections

In order to design a more realistic model, variables such as temperature, pH and phosphorus should be added. Moreover, initial condition of the different microorganism should be measured and should not be calibrated. Finally a validation of the model should be done.

To improve the results of the optimization more initial conditions should be taken into considerations to be closer to the global optimum, and others objectives functions, such as to minimize operational costs or to maximize autotrophic metabolism in microalgae.

While this optimization strategy reduces the uncertainty about finding an optimal solution, it can not guarantee that is the universal optima. In order to further the search other software programs, such as GAMS/BARON[29], could be used.

In order to decrease the amount of assumptions in the ecosystem coupling, models should have the same input and output variables, and neither should be considered input constants (F_0, COD_{in}, NO_{in} and $NH_{4_{in}}$) nor I_0 approximation, but experimental data should be used.

Bibliography

[1] Chisti, Y. Biodiesel from microalgae beats bioethanol. Trends in Biotechnology, 26(3): 126–131, 2008.

[2] Larrain Rencoret Urzua. New incentives on Non-Conventional Renewable Energy, august 2015.

[3] Microbiología Industrial. Principios y microbiología del tratamiento de aguas residuales, October 2015.

[4] Bernard, O. Hurdles and challenges for modelling and control of microalgae for CO 2 mitigation and biofuel production. Journal of Process Control, 21(10): 1378–1389, 2011.

[5] Mairet, F., Bernard, O., Cameron, E., Ras, M., Lardon, L., Steyer, J.P., Chachuat, B. Three-reaction model for the anaerobic digestion of microalgae. Biotechnology and Bioengineering, 109(2): 415–425, 2012.

[6] Algae Industry Magazine. Innovations, august 2015.

[7] Yang, C., Hua, Q., Shimizu, K. Energetics and carbon metabolism during growth of microalgalcells under photoautotrophic, mixotrophic and cycliclight-autotrophic/dark-heterotrophic conditions. Biochemical engineering journal, 6: 87–102, 2000.

[8] Wang, Z., Zhang, X.X., Lu, X., Liu, B., Li, Y., Long, C., Li, A. Abundance and Diversity of Bacterial Nitrifiers and Denitrifiers and Their Functional Genes in Tannery Wastewater Treatment Plants Revealed by High-Throughput Sequencing. PLoS ONE, 9(11): e113603, 2014.

[9] Waste to Energy. Anaerobic Digestion Process, august 2015.

[10] Bernard, O., Hadj-Sadok, Z., Dochain, D., Genovesi, a., Steyer, J.P. Dynamical model development and parameter identification for an anaerobic wastewater treatment process. Biotechnology and bioengineering, 75(4): 424–438, 2001.

[11] Mathworks. fminsearch, 2015.

[12] Nelder J, M.R. A simplex method for function minimization. 4: 308 – 313., 1965.

[13] Minister de l ecologie, de l energie, du developpement durable et de la mer. Les textes techniques relatifs a l assainissement collectifs des eaux usees domestiques, 2013.

[14] Mathworks. fmincon, 2015.

[15] Karmarkar, N. A new polynomial-time algorithm for linear programming. Combinatorica, 4(4): 373–395, 1984.

[16] Geider, R., La Roche, J. Redfield revisited: variability of C:N:P in marine microalgae and its biochemical basis. European Journal of Phycology, 37(1): 1–17, 2002.

[17] Béchet, Q., Chambonnière, P., Shilton, A., Guizard, G., Guieysse, B. Algal productivity modeling: A step toward accurate assessments of full-scale algal cultivation. Biotechnology and bioengineering, 112(5): 987–96, 2015.

[18] Sophia-antipolis, A.s.E.P.I.C.I. Modelling the Coupling Between an Anaerobic Digester (AD) and Microalgal Pond (MP). 1–13, 2004.

[19] J. Alex, H. Pastagiya, N.H. First results of the development of a combined high rate biomass-algal model for wastewater treatment applications. 2(March): 273–281, 2010.

[20] Maroneze, M.M., Barin, J.S., Menezes, C.R.D., Queiroz, M.I., Queiroz, L., Jacob-lopes, E. Treatment of cattle-slaughterhouse wastewater and the reuse of sludge for biodiesel production by microalgal heterotrophic bioreactors. 4(December): 521–524, 2014.

[21] Jeppsson, U. A General Description of the Activated Sludge Model No . 1 (ASM1). Components, 1(1): 1–16, 1996.

[22] NaturalGas. Overview of Natural Gas, September 2013.

[23] Passos, F., Gutiérrez, R., Brockmann, D., Steyer, J.P., García, J., Ferrer, I. Microalgae production in wastewater treatment systems, anaerobic digestion and modelling using ADM1. Algal Research, 10: 55–63, 2015.

[24] Chisti, Y. Biodiesel from microalgae beats bioethanol. Trends in Biotechnology, 26(3): 126–131, 2008.

[25] Coons, J.E., Kalb, D.M., Dale, T., Marrone, B.L. Getting to low-cost algal biofuels: A monograph on conventional and cutting-edge harvesting and extraction technologies. Algal Research, 6(PB): 250–270, 2014.

[26] Benemann, J. Microalgae for Biofuels and Animal Feeds. Energies, 6(11): 5869–5886, 2013.

[27] CNG Europe. Map of Natural Gas Vehicle (NVG) Compressed natural gas (CNG) filling stations in Europe, November 2015.

[28] Bernard, O., Rémond, B. Validation of a simple model accounting for light and temperature effect on microalgal growth. Bioresource Technology, 123: 520–527, 2012.

[29] GAMS. BARON, 2015.

Appendices

Appendix A

MP Model Codes

```
%% Clear variables
clc;
close all
clear all
tic

%% Time of program
tin   = 0;
tfin  = 934;
tspan = tin:1:tfin;

%% Initials Conditiones
Ci    = zeros(5,1);

CODexp = 297;
kin    = 0.0523;

Ci(1) = 0.0325*CODexp;              % Biomass: Something important: Initial biomass plus initial COD should be equal to 297
Ci(2) = (1-0.0325)*CODexp;          % COD: According to experimental data "LHRA: DCO filtré"
Ci(3) = 28.6;                       % Ntot: According to experimental data, before filter
Ci(4) = 0;                          % M: Micraalgae dead
Ci(5) = kin*CODexp;                 % Norg = Ntot,filt - NH4 - NO2 - NO3


%% Experimental Data
experimental = xlsread('data','Experimental','A2:F26'); % Data filtered: LHRA


%% Inlets
influent = xlsread('data','Inlets','A2:I26');  % Data filtered: Eau Brute. Between 11/05 and 19/06
I_0 = xlsread('data','I_0','A2:B3765');        % Irradiance. Between 11/05 and 19/06 at 8:45

%% Parameters
```

```
%p = [um          KI          ucod       Kcod       R          k_N        Kn         k_C
    ]
% Literature:
%p = [0.0708     100        0.041      20.000  0.0081/24     0.08     0.01    1.5]';
p = [0.0352   130.4245     0.0109    17.0062     0.0082     0.3935     0.0162
    3.1654]'; %22.5


%% Solver
[time y]=ode45(@ODEs_raceway,tspan,Ci,[],influent,I_0,p);
toc

%% COD and N total estimated (ye)
CODe = y(:,1)+y(:,2);


%% Graphs

%%%%%%%%%%%%%% Biomass and COD %%%%%%%%%%
figure
subplot(2,2,1)
plot(time,y(:,1),'b')
xlabel('Time [h]');
ylabel('Biomass [mg/L]');
grid on

subplot(2,2,2)
plot(time,y(:,2),'b')
xlabel('Time [h]');
ylabel('COD [mg/L]');
grid on

subplot(2,2,3)
plot(time,CODe,'r')
xlabel('Time [h]');
ylabel('Estim. Tot COD [mg/L]');
grid on

subplot(2,2,4)
plot(experimental(:,1),experimental(:,2),'r')
xlabel('Time [h]');
ylabel('Exper. Tot COD [mg/L]');
grid on

%%%%% Importants ones %%%%%%%
figure
plot(experimental(:,1),experimental(:,2),'-o','MarkerSize',7)
xlabel('Time [h]');
ylabel('COD [mg/L]');
hold on
plot(time,CODe,'-black','LineWidth',1.5)
legend('exp','est')
grid on

figure
plot(experimental(:,1),experimental(:,5),'-o','MarkerSize',7)
```

```
xlabel('Time [h]');
ylabel('N [mg/L]');
hold on
plot(time,y(:,3),'-black','LineWidth',1.5)
legend('exp','est')
grid on

figure
plot(experimental(:,1),experimental(:,6),'-o','MarkerSize',7)
xlabel('Time [h]');
ylabel('Norg [mg/L]');
hold on
plot(time,y(:,5),'-black','LineWidth',1.5)
legend('exp','est')
grid on
```

```
function dy = ODEs_raceway(t,y,influent,I_0,p)
dy = zeros(5,1);  % Initialization of variables


%% Parameters

um    = p(1);      % umax of u(I)
KI    = p(2);      % KI of u(I)
k     = 0.2;      % I=I0*exp(-kxz)
ucod  = p(3);
Kcod  = p(4);
R     = p(5);      % Respiration
k_N   = p(6);      % Kind of yield
Kn    = p(7);      % Half saturation constant
k_C   = p(8);
L     = 0.4;       % Deep of the pond
V     = 1880;      % Volume pond
kin   = 0.0523;


%% Inlets
CODint = interp1(influent(:,1),influent(:,2),t);
Nint   = interp1(influent(:,1),influent(:,8),t);
Norint = kin*CODint;
Qint   = interp1(influent(:,1),influent(:,5),t);
I0     = interp1(I_0(:,1),I_0(:,2),t);


%% Equations
% Auxiliar variables to make easier to write equations
f_I   = (1/(L*k*y(1)))*log((I0+KI)/(I0*exp(-k*L*y(1))+KI));
f_C   = (y(2)/(y(2)+Kcod));
f_N   = (y(3)/(y(3)+Kn));

% ODEs
dy(1) = -(Qint/V)*y(1)+(um*f_I+ucod*f_C)*f_N*y(1)-R*y(1);      % Microalgae
dy(2) = (Qint/V)*(CODint-y(2))-k_C*ucod*f_C*f_N*y(1);                                  %
    COD
dy(3) = (Qint/V)*(Nint-y(3))-f_N*(k_N*um*f_I+(k_N-kin*k_C)*ucod*f_C)*y(1); %
```

```
    Nitrogen
dy(4) = -(Qint/V)*y(4)+R*y(1);                                    % Microalgae dead
dy(5) = (Qint/V)*(Norint-y(5))-kin*k_C*ucod*f_C*f_N*y(1); % Nitrogen organic
```

Appendix B

Sensitivity analysis of MP model

The sensitivity analysis was made only with the most importants curves, it means COD and nitrogen concentration. The range taken was 50% around the calibrated values.

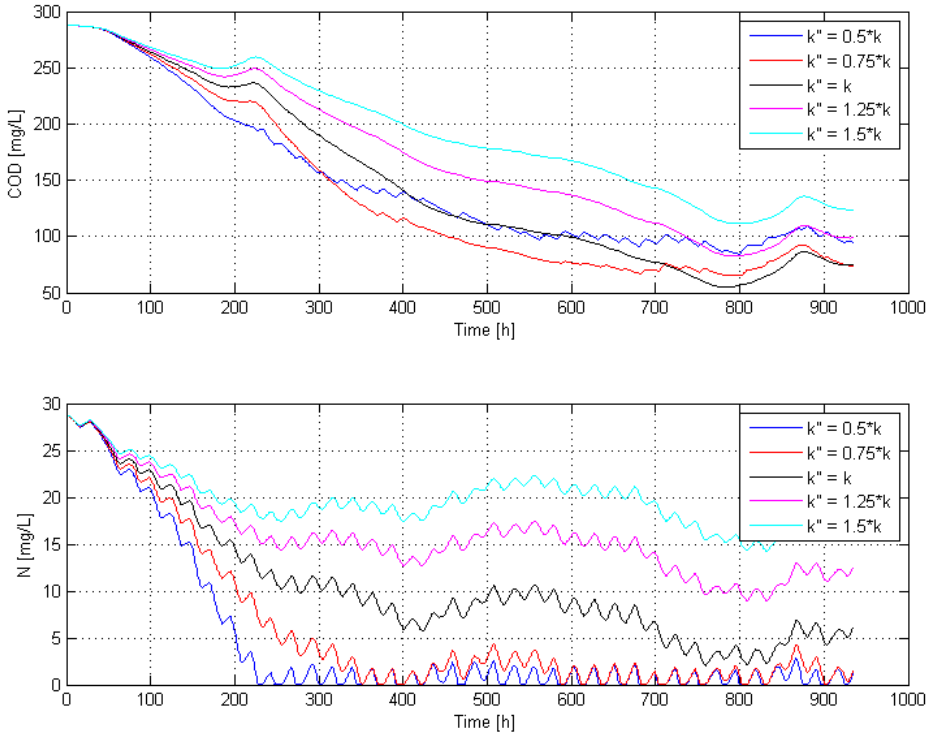

Figure B.1: Sensitivity analysis of parameter k in MP model.

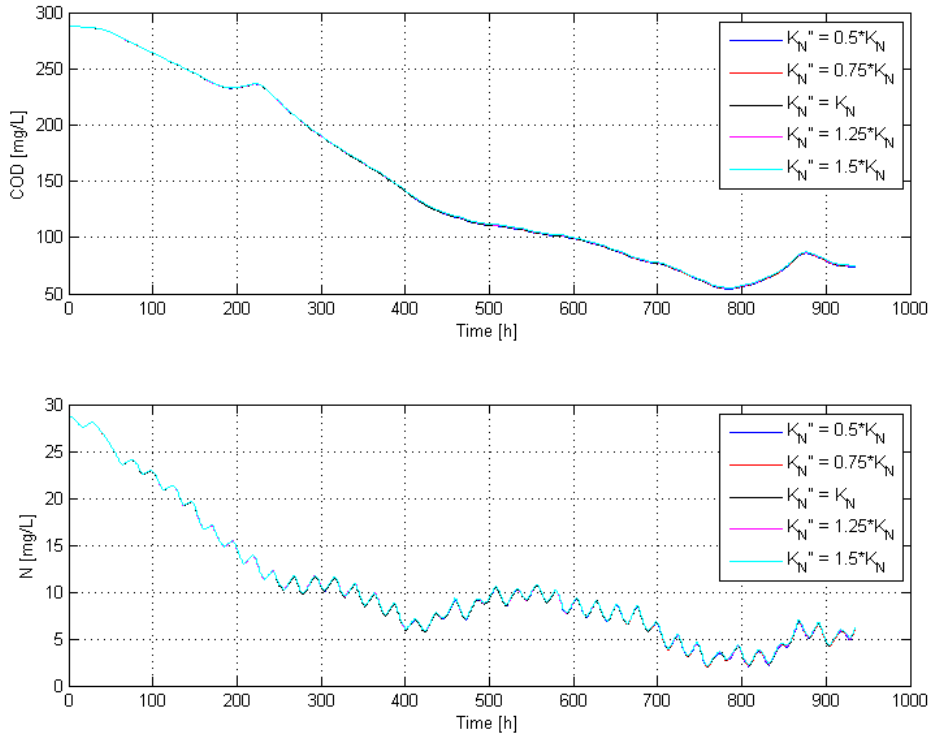

Figure B.2: Sensitivity analysis of parameter K_N in MP model.

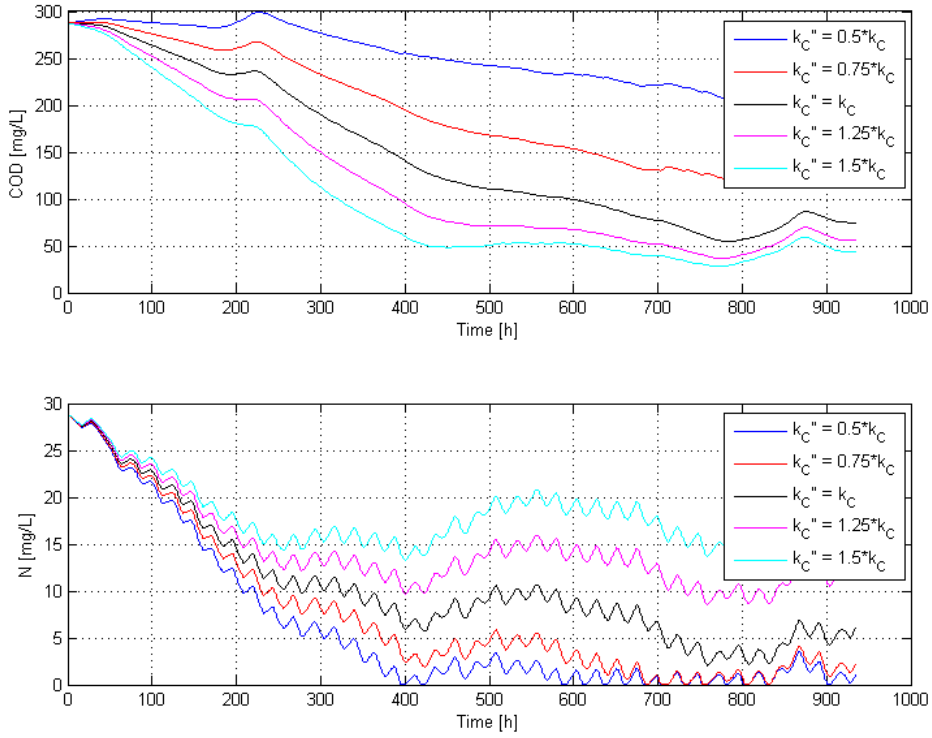

Figure B.3: Sensitivity analysis of parameter k_c in MP model.

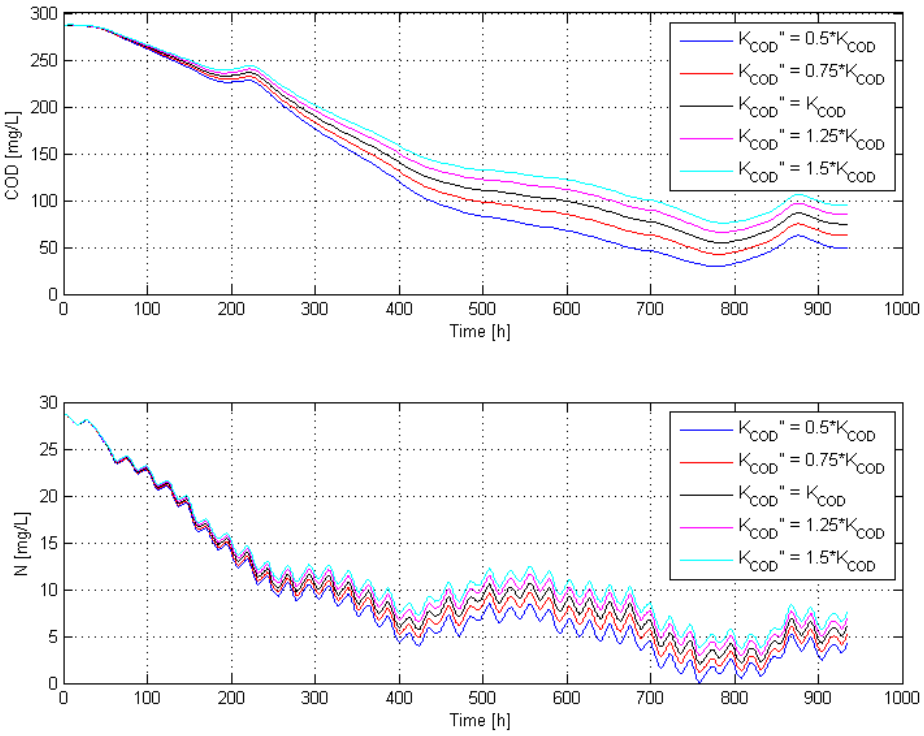

Figure B.4: Sensitivity analysis of parameter K_{COD} in MP model.

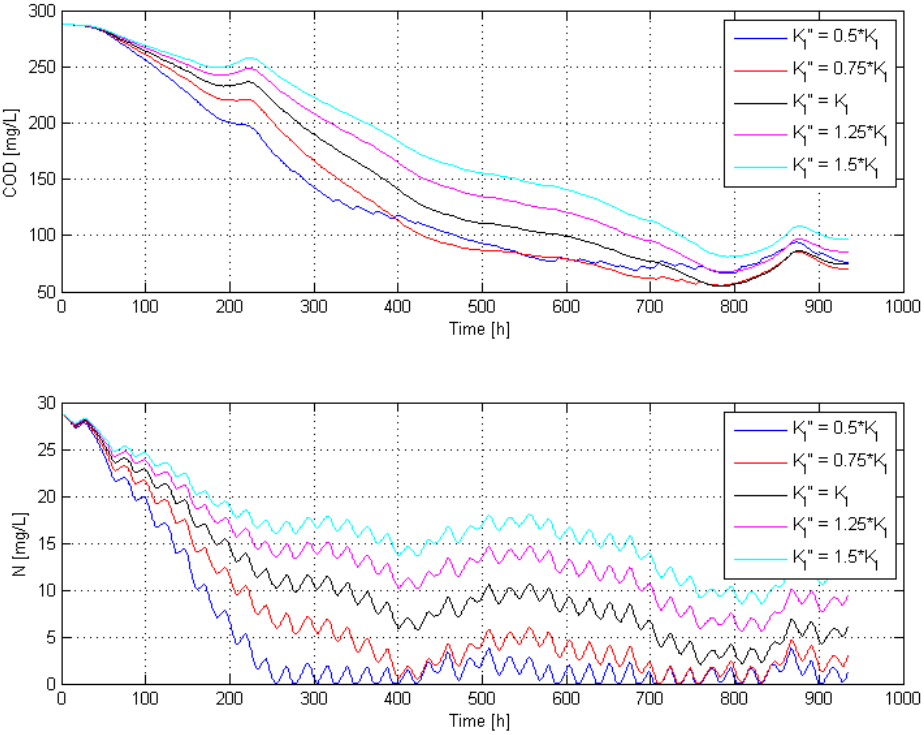

Figure B.5: Sensitivity analysis of parameter K_I in MP model.

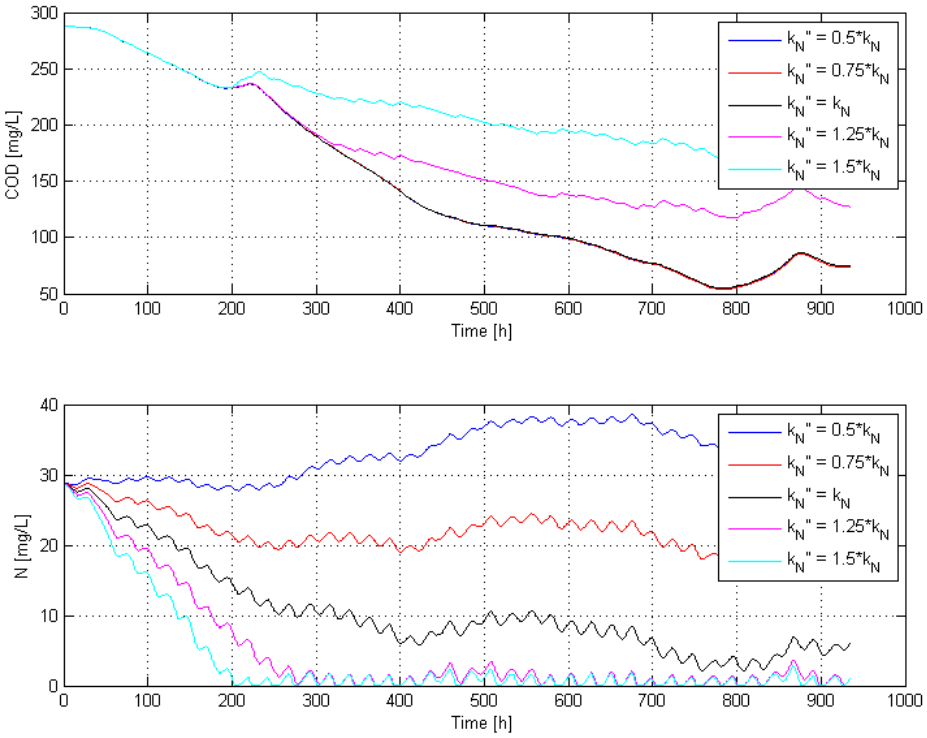

Figure B.6: Sensitivity analysis of parameter k_N in MP model.

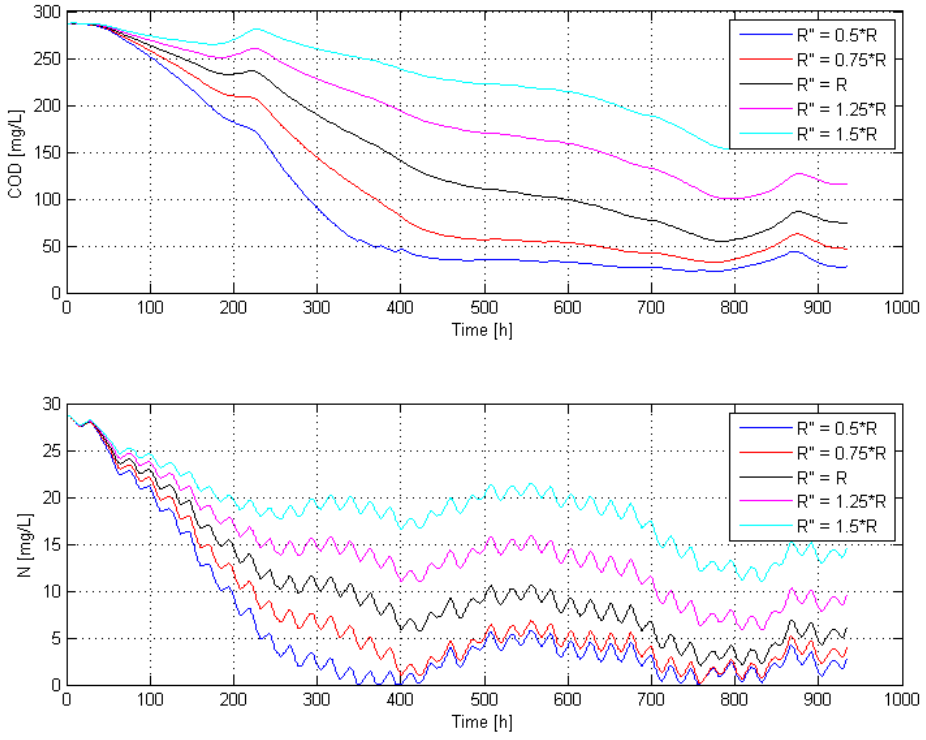

Figure B.7: Sensitivity analysis of parameter R in MP model.

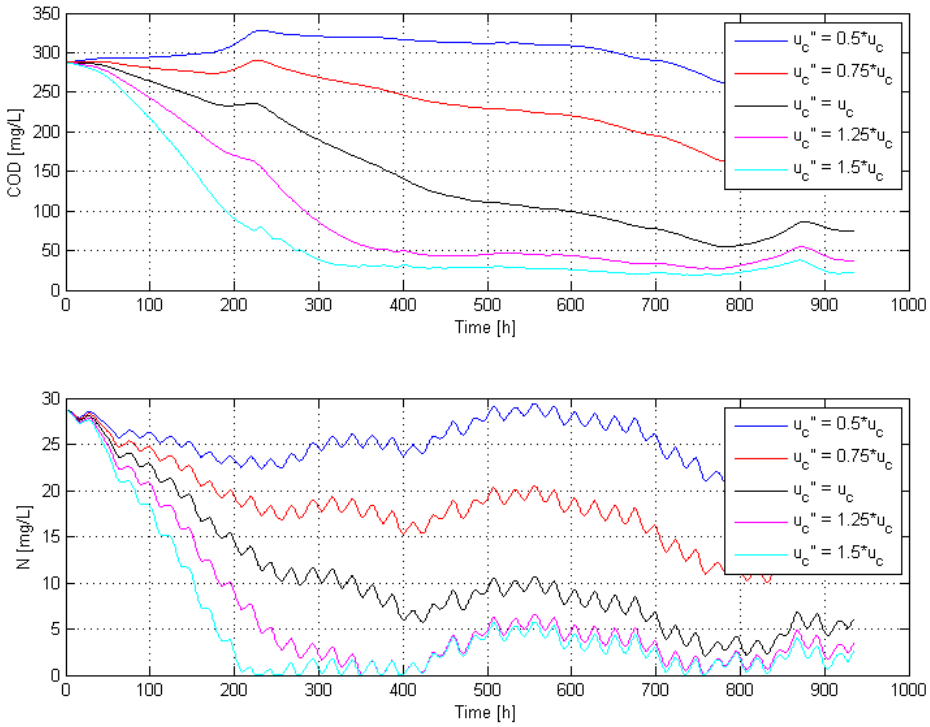

Figure B.8: Sensitivity analysis of parameter μ_C in MP model.

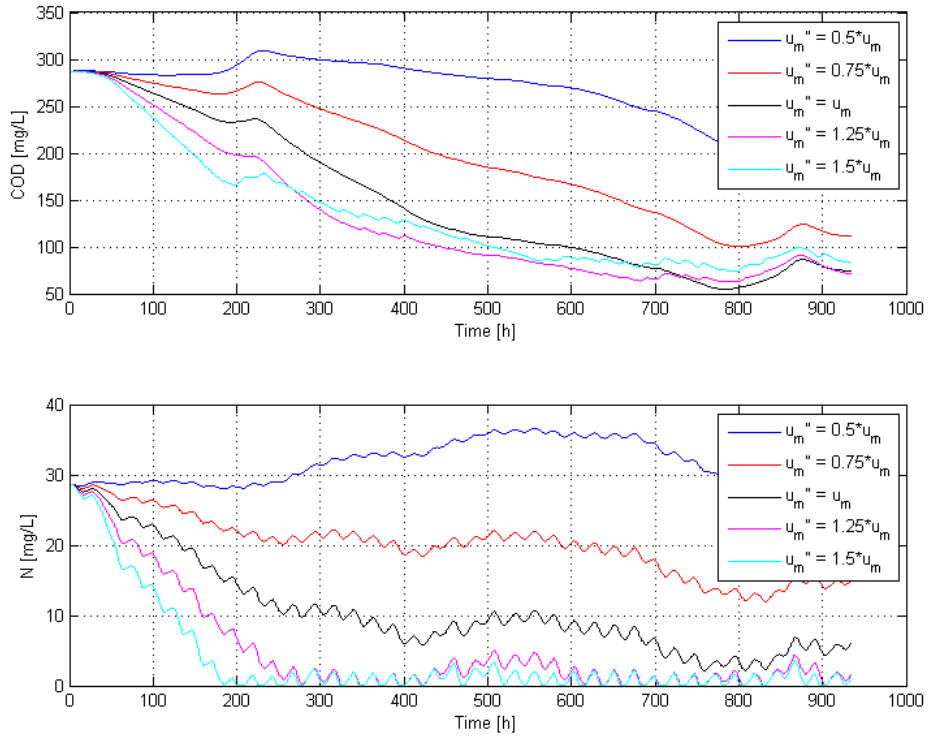

Figure B.9: Sensitivity analysis of parameter μ_m in MP model.

Appendix C

WWT Model Codes

```
%% Clear variables
clc;
clear all
%close all
tic
%global I_0


%% Time of program
tin   = 0;
tfin  = 935;
tspan = tin:1:tfin;

%% Parameters
%p = [(1)k     (2)k_NH2    (3)k_C]
p = [0.1811     2.3723     3.6537]; %38,5

%% Initials Conditiones
Ci    = zeros(7,1);

CODexp = 297;
kin    = 0.0523;

Ci(1) = 0.0325*CODexp;            % Microalgae: Something important: Initial biomass plus initial COD should be equal to 297
Ci(2) = 0.0025*CODexp;            % Nitrifiers: Something important: Initial biomass plus initial COD should be equal to 297
Ci(3) = (1-0.0325-0.0025)*CODexp;          % COD: According to experimental data "LHRA: DCO filtré"
Ci(4) = 28.39;                    % N/NH4: According to experimental data
Ci(5) = 0.216;                    % N/(NO3+NO2): According to experimental data
Ci(6) = 0;            % M: Biomass dead
Ci(7) = kin*CODexp;               % Norg = Ntot,filt - NH4 - NO2 - NO3


%% Experimental Data
experimental = xlsread('data','Experimental','A2:F26'); % Data filtered: LHRA

```

```
%% Inlets
influent = xlsread('data','Inlets','A2:J26');  % Data filtered: Eau Brute.
    Between 11/05 and 19/06
I_0 = xlsread('data','I_0','A2:B3765');        % Irradiance. Between 11/05 and
     19/06 at 8:45


%% Solver
%options = odeset('MaxStep',0.08,'AbsTol',1e-9,'RelTol',1e-6);
%[time y]=ode45(@ODEs_raceway_N,tspan,Ci,options,influent,I_0,p);
[time y]=ode45(@ODEs_raceway_N,tspan,Ci,[],influent,I_0,p);
toc

%% COD total estimated (CODe), total biomass (Be)and total Nitrogen (Ne)
CODe = y(:,1)+y(:,2)+y(:,3);
Be   = y(:,1)+y(:,2)+y(:,6);


%% Graphs

%%%%% Importants ones %%%%%%%
figure
plot(experimental(:,1),experimental(:,2),'-o','MarkerSize',7)
xlabel('Time [h]');
ylabel('COD [mg/L]');
hold on
plot(time,CODe,'-black','LineWidth',1.5)
legend('Experimental','Estimated')
grid on

figure
plot(experimental(:,1),experimental(:,3),'-o','MarkerSize',7)
xlabel('Time [h]');
ylabel('NH4 [mg/L]');
hold on
plot(time,y(:,4),'-black','LineWidth',1.5)
legend('Experimental','Estimated')
grid on

figure
plot(experimental(:,1),experimental(:,4),'-o','MarkerSize',7)
xlabel('Time [h]');
ylabel('NO [mg/L]');
hold on
plot(time,y(:,5),'-black','LineWidth',1.5)
legend('Experimental','Estimated')
%set(gca,'YTickLabel',num2str(get(gca,'YTick').'))
grid on

figure
plot(experimental(:,1),experimental(:,6),'-o','MarkerSize',7)
xlabel('Time [h]');
ylabel('Norg [mg/L]');
hold on
plot(time,y(:,7),'-black','LineWidth',1.5)
legend('Experimental','Estimated')
```

```
grid on


%%%%%%%%%%%%% Biomass %%%%%%%%%%
figure
subplot(3,1,1)
plot(time,y(:,1),'b')
xlabel('Time [h]');
ylabel('Microalgae [mg/L]');
grid on

subplot(3,1,2)
plot(time,y(:,2),'b')
xlabel('Time [h]');
ylabel('Nitrifiers [mg/L]');
grid on

subplot(3,1,3)
plot(time,Be,'b')
xlabel('Time [h]');
ylabel('Total Biomass [mg/L]');
grid on


%%%%%%%%%% Total Biomass and COD %%%%%%%%%%

figure
subplot(2,3,1)
plot(time,y(:,1),'b')
xlabel('Time [h]');
ylabel('Microalgae [mg/L]');
grid on

subplot(2,3,2)
plot(time,y(:,2),'b')
xlabel('Time [h]');
ylabel('Nitrifiers [mg/L]');
grid on

subplot(2,3,3)
plot(time,y(:,6),'b')
xlabel('Time [h]');
ylabel('Biomass died [mg/L]');
grid on

subplot(2,3,4)
plot(time,Be,'r')
xlabel('Time [h]');
ylabel('Total Biomass [mg/L]');
grid on

subplot(2,3,5)
plot(time,y(:,3),'r')
xlabel('Time [h]');
ylabel('COD [mg/L]');
grid on
```

```
147
subplot(2,3,6)
149 plot(time,CODe,'r')
xlabel('Time [h]');
151 ylabel('Total COD [mg/L]');
grid on
153

155 %%%%%%%%%%%%% NH4 and NO3 %%%%%%%%%
figure
157 subplot(2,2,1)
plot(time,y(:,4),'g')
159 xlabel('Time [h]');
ylabel('Estim. NH4 [mg/L]');
161 %axis ([0 1000 0 50]);
grid on
163
subplot(2,2,2)
165 plot(experimental(:,1),experimental(:,3),'r')
xlabel('Time [h]');
167 ylabel('Exper. NH4 [mg/L]');
grid on
169
subplot(2,2,3)
171 plot(time,y(:,5),'g')
xlabel('Time [h]');
173 ylabel('Estim. NO3 [mg/L]');
grid on
175
subplot(2,2,4)
177 plot(experimental(:,1),experimental(:,4),'r')
xlabel('Time [h]');
179 ylabel('Exper. NO3 [mg/L]');
grid on
```

```
function dy = ODEs_raceway_N(t,y,influent,I_0,p)
2 %global I_0
dy = zeros(7,1);  % Initialization of variables
4 % I_0
% influent
6
%% Parameters
8 % Parameters from the literature
%p = [(1)um1    (2)KI      (3)k       (4)ucod    (5)Kcod    (6)R1     (7)Knh1
    (8)Kno31   (9)unh42   (10)Knh2   (11)R2    (12)k_NO    (13)k_NH    (14)k_N1   (15)
    k_C]
10 %p = [0.0708   100.000     0.9979       0.041     20.000     0.0020     0.01
    0.01     0.033        1.0000     0.0083    4.16       4.246      0.08       2.33
    kin];
L     = 0.4;       % Deep of the pond
12 V     = 1880;      % Volume pond


14
% Microalgae
16 um1    = 0.0204;      % umax of u(I)
KI     = 44.4093;      % KI of u(I)
```

```
k       = p(1);      % I=I0*exp(-kxz)
ucod    = 0.0115;      % umax of u(COD)
Kcod    = 21.3288 ;      % Kcod of u(COD)
R1      = 0.001;      % Respiration
Knh41   = 0.0127;      % Half saturation constant
Kno31   = 0.0047;      % Half saturation constant


% Nitrifiers
unh42   = 0.009;
Knh42   = 1.0461;
R2      = 0.0006;
k_NO2   = p(2)-0.1;
k_NH2   = p(2);
k_N1    = 0.3871;
k_C     = p(3);
kin     = 0.0523;

%% Inlets
CODint = interp1(influent(:,1),influent(:,2),t);
NH4int = interp1(influent(:,1),influent(:,3),t);
NOint  = interp1(influent(:,1),influent(:,4),t);
Qint   = interp1(influent(:,1),influent(:,5),t);
%Norint   = interp1(influent(:,1),influent(:,9),t);
Norint = kin*CODint;
I0     = interp1(I_0(:,1),I_0(:,2),t);


%% Equations

% Auxiliar variables to make easier to write equations
f_I    = (1/(L*k*y(1)))*log((I0+KI)/(I0*exp(-k*L*y(1))+KI));
%f_I   = (1/(L*k*y(1)))*log((I0+KI)/(I0*exp(-k*L*(y(1)+y(2)))+KI));
f_C    = (y(3)/(y(3)+Kcod));
f_NH1  = (y(4)/(y(4)+Knh41));
f_NO1  = (y(5)/(y(5)+Kno31))*(Knh41/(y(4)+Knh41));
f_NH2  = (y(4)/(y(4)+Knh42));



% ODEs
dy(1) = -(Qint/V)*y(1)+(um1*f_I+ucod*f_C)*(f_NH1+f_NO1)*y(1)-R1*y(1);
                                                    % 1. Microalgae Balance
%dy(2) = 0;
dy(2) = -(Qint/V)*y(2)+unh42*f_NH2*y(2)-R2*y(2);    % 2. Nitrifiers balance
dy(3) = (Qint/V)*(CODint-y(3))-k_C*ucod*f_C*(f_NH1+f_NO1)*y(1);
                                                    % 3. COD Balance
dy(4) = (Qint/V)*(NH4int-y(4))-f_NH1*(k_N1*um1*f_I+(k_N1-kin*k_C)*ucod*f_C)*y
    (1)-k_NH2*unh42*f_NH2*y(2);                     % 4. NH4 Balance
dy(5) = (Qint/V)*(NOint-y(5))-f_NO1*(k_N1*um1*f_I+(k_N1-kin*k_C)*ucod*f_C)*y
    (1)+k_NO2*unh42*f_NH2*y(2);                     % 5. NO Balance
%dy(5) = (Qint/V)*(NOint-y(5))-f_NO1*(k_N1*um1*f_I)*y(1)+k_NO2*unh42*f_NH2*y
    (2);                % 5. NO Balance
dy(6) = -(Qint/V)*y(6)+R1*y(1)+R2*y(2);

   % 6. Microalgae died Balance
```

```
dy(7) = (Qint/V)*(Norint-y(7))-kin*k_C*ucod*f_C*(f_NH1+f_NO1)*y(1);
```

Appendix D

Sensitivity analysis of WWT model

The sensitivity analysis was made only with the most important curves, it means COD and nitrogen concentration. The range taken was 50% around the calibrated values.

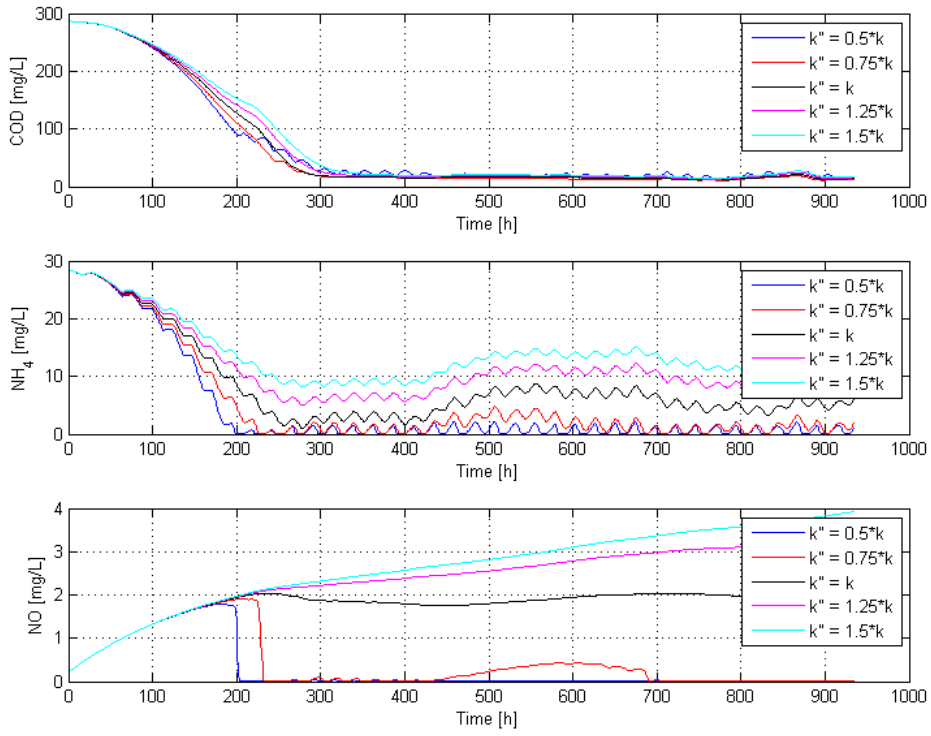

Figure D.1: Sensitivity analysis of parameter k in WWT model.

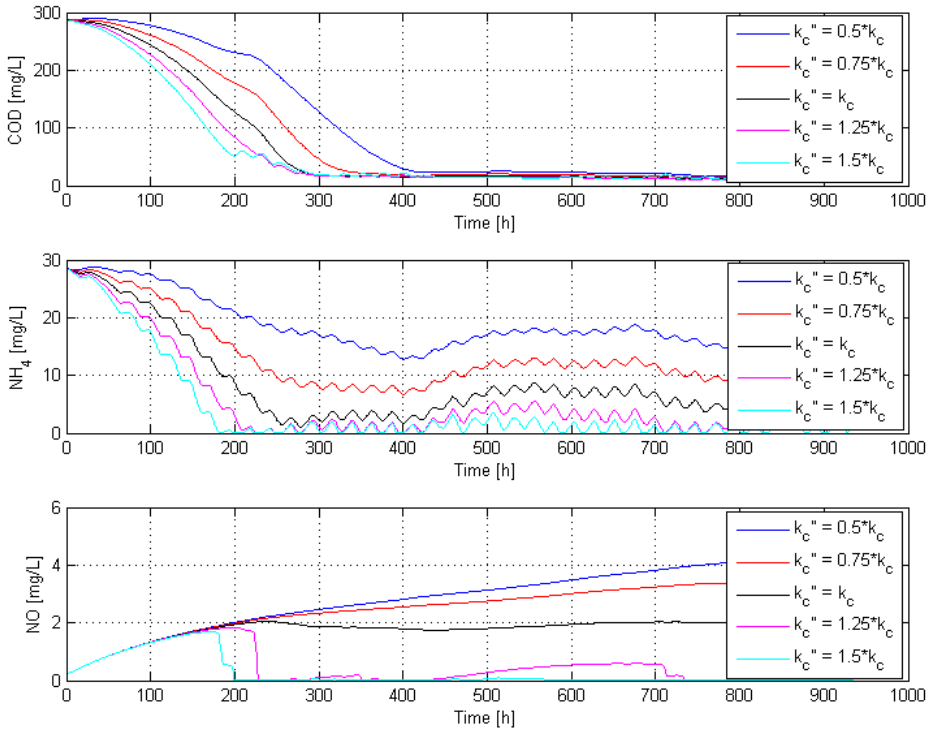

Figure D.2: Sensitivity analysis of parameter k_{C1} in WWT model.

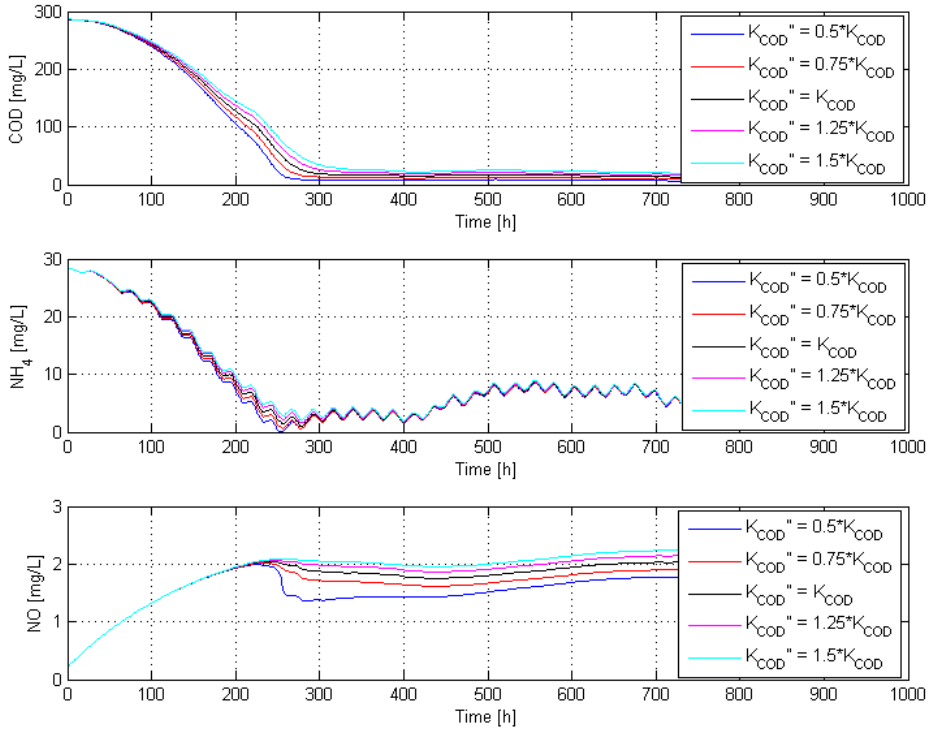

Figure D.3: Sensitivity analysis of parameter K_{COD} in WWT model.

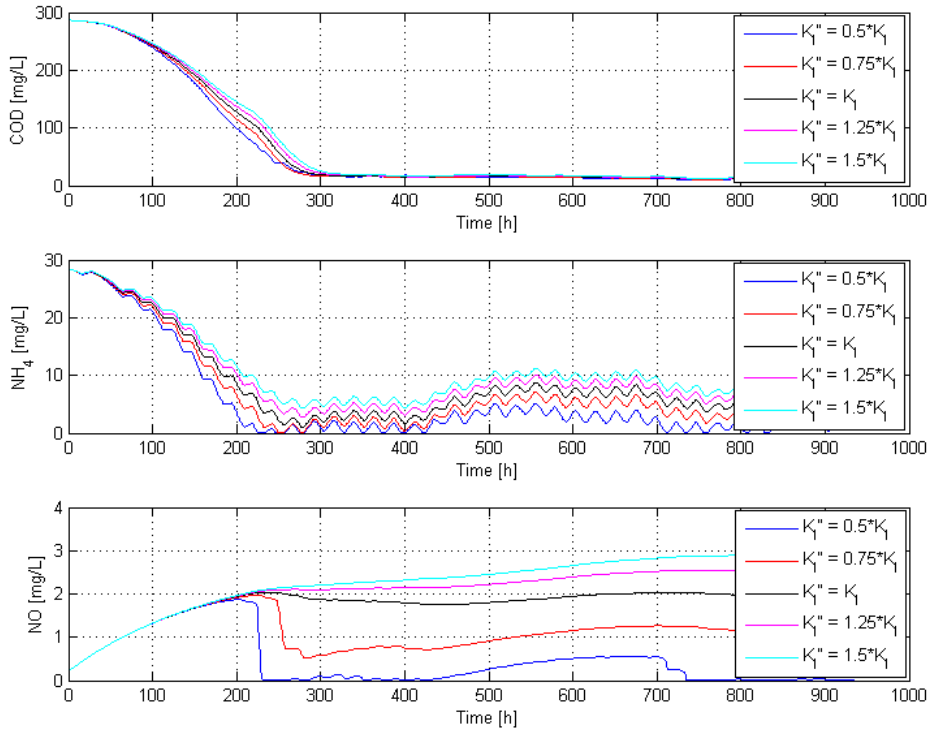

Figure D.4: Sensitivity analysis of parameter K_I in WWT model.

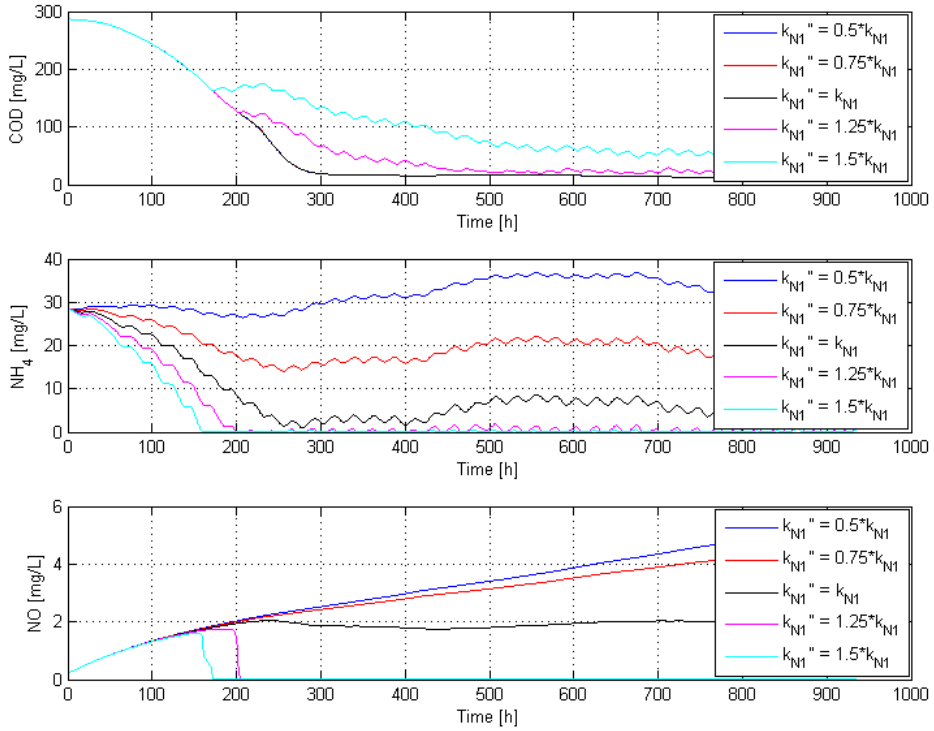

Figure D.5: Sensitivity analysis of parameter k_{N1} in WWT model.

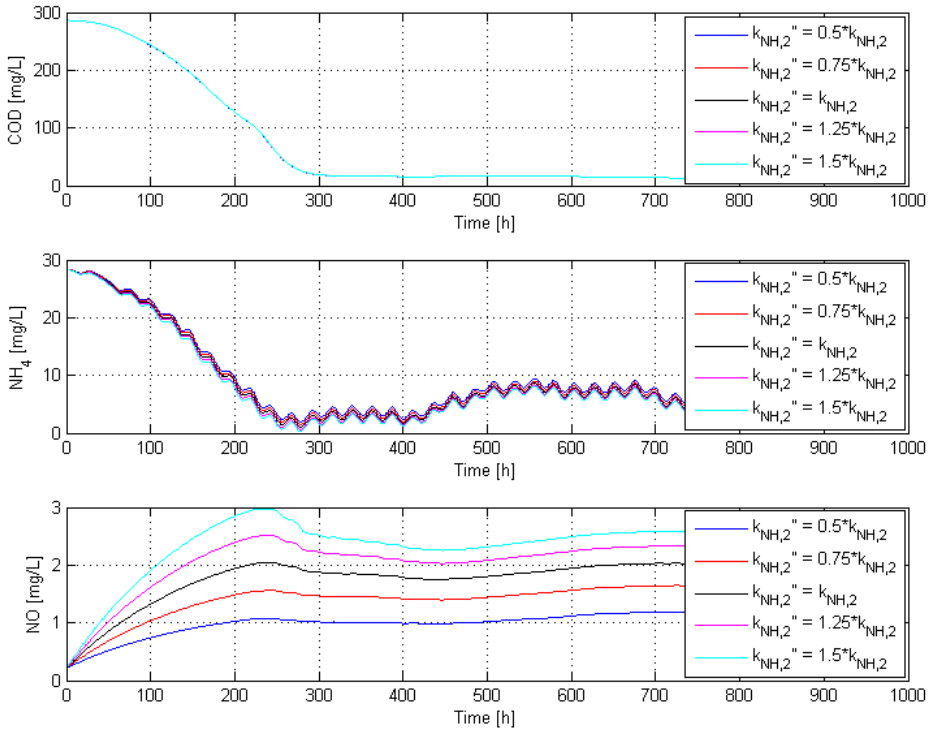

Figure D.6: Sensitivity analysis of parameter $k_{NH,2}$ in WWT model.

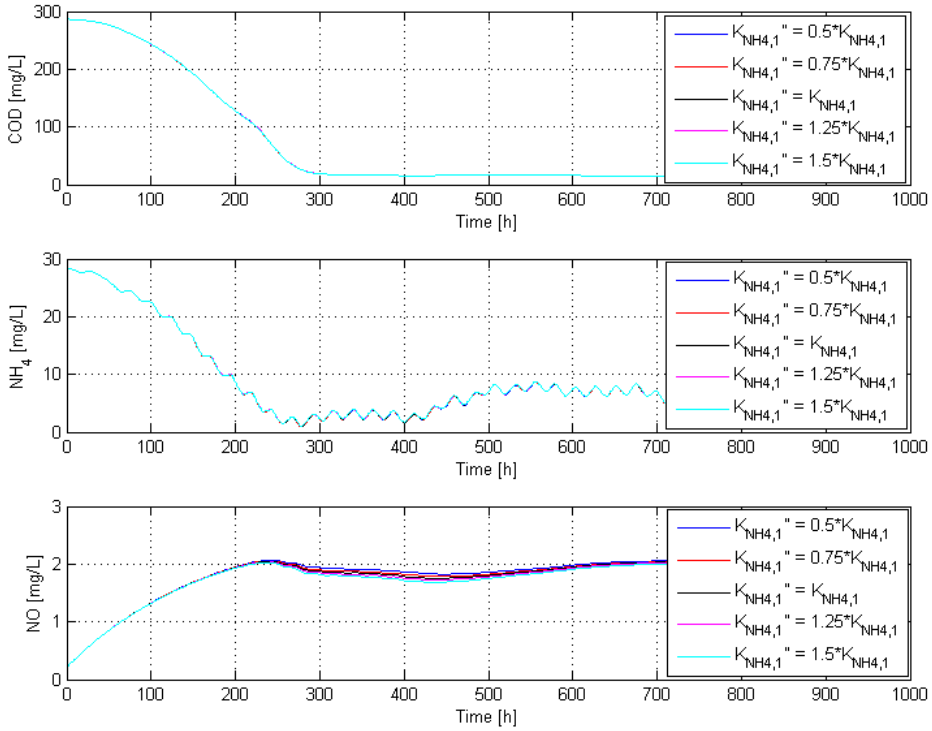

Figure D.7: Sensitivity analysis of parameter $K_{NH_4,1}$ in WWT model.

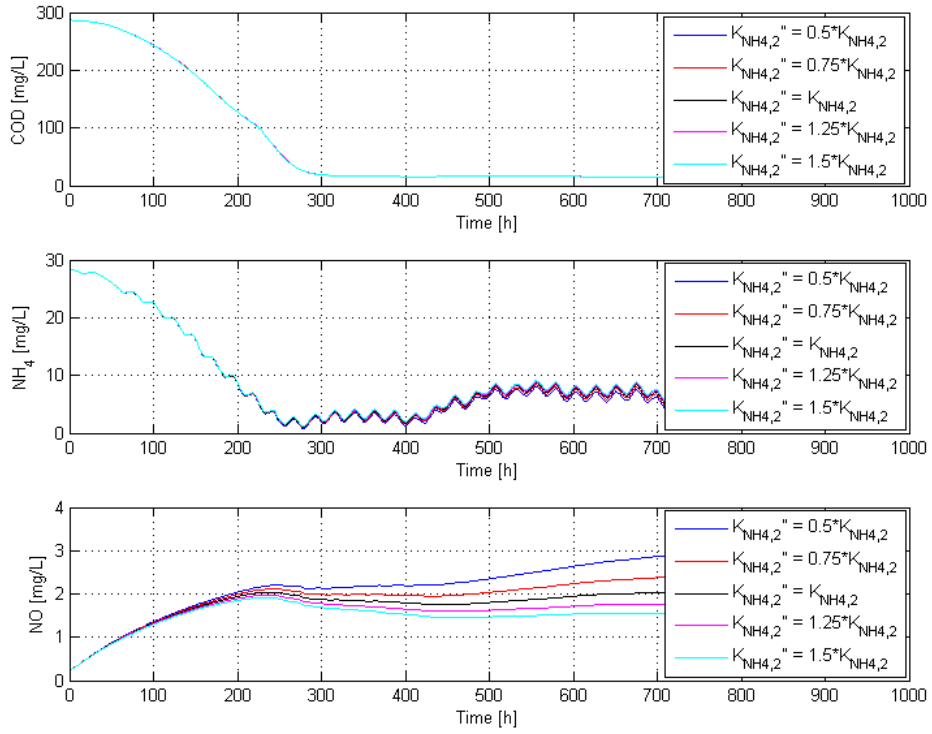

Figure D.8: Sensitivity analysis of parameter $K_{NH_4,2}$ in WWT model.

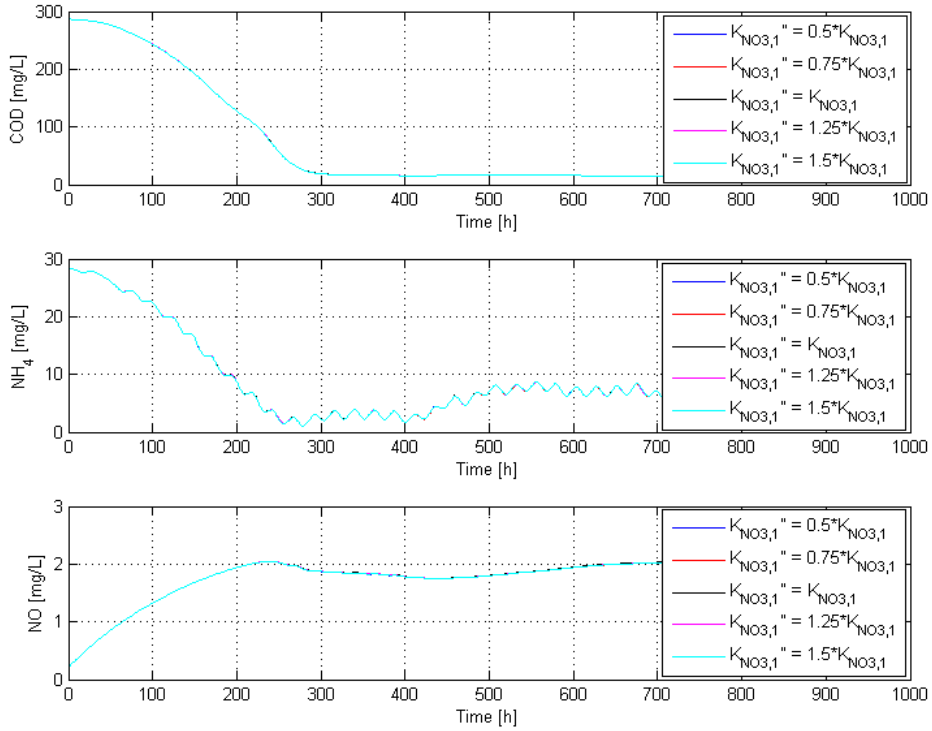

Figure D.9: Sensitivity analysis of parameter $K_{NO_3,1}$ in WWT model.

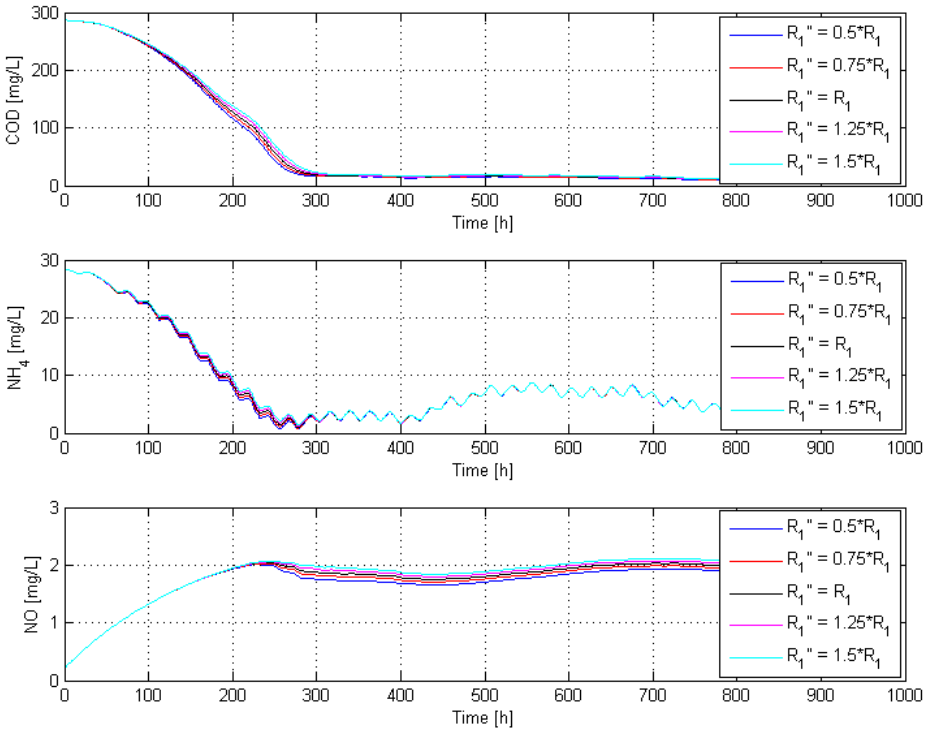

Figure D.10: Sensitivity analysis of parameter R_1 in WWT model.

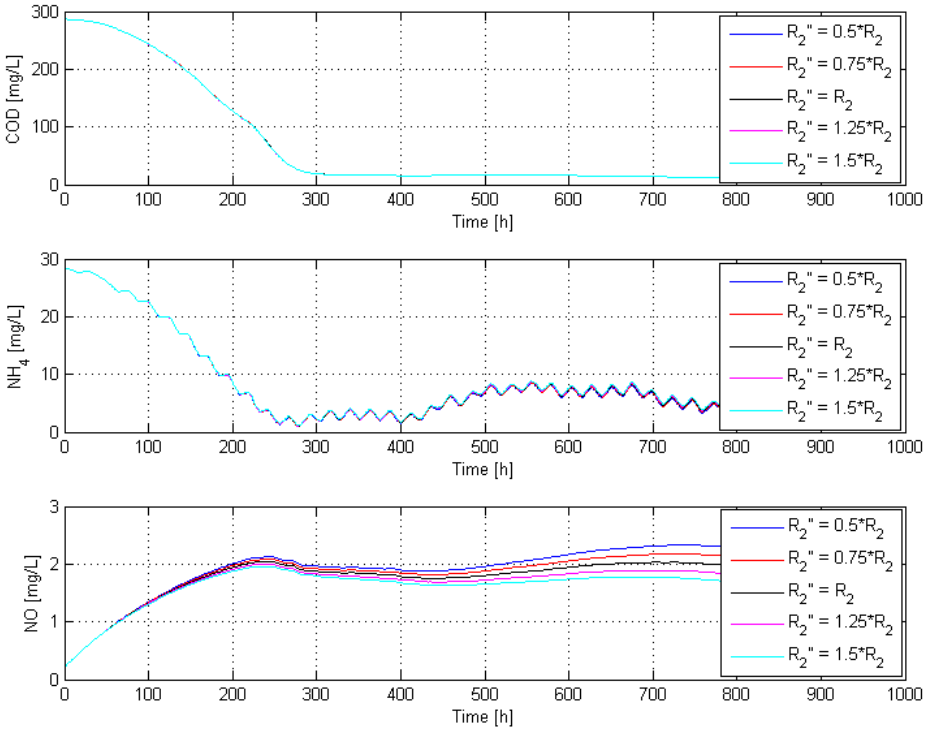

Figure D.11: Sensitivity analysis of parameter R_2 in WWT model.

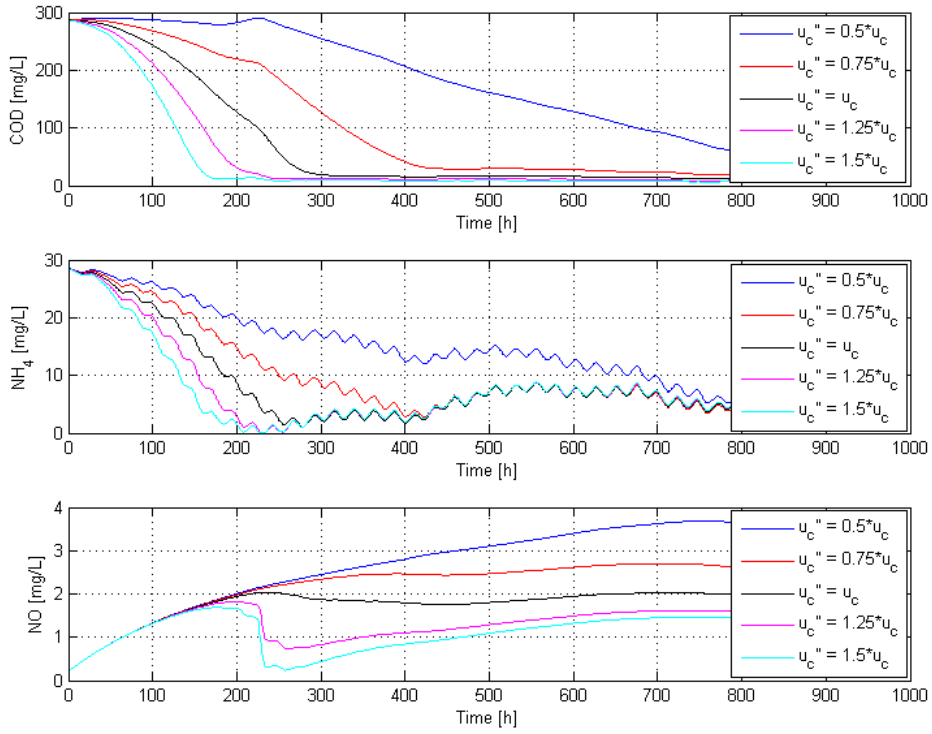

Figure D.12: Sensitivity analysis of parameter μ_C in WWT model.

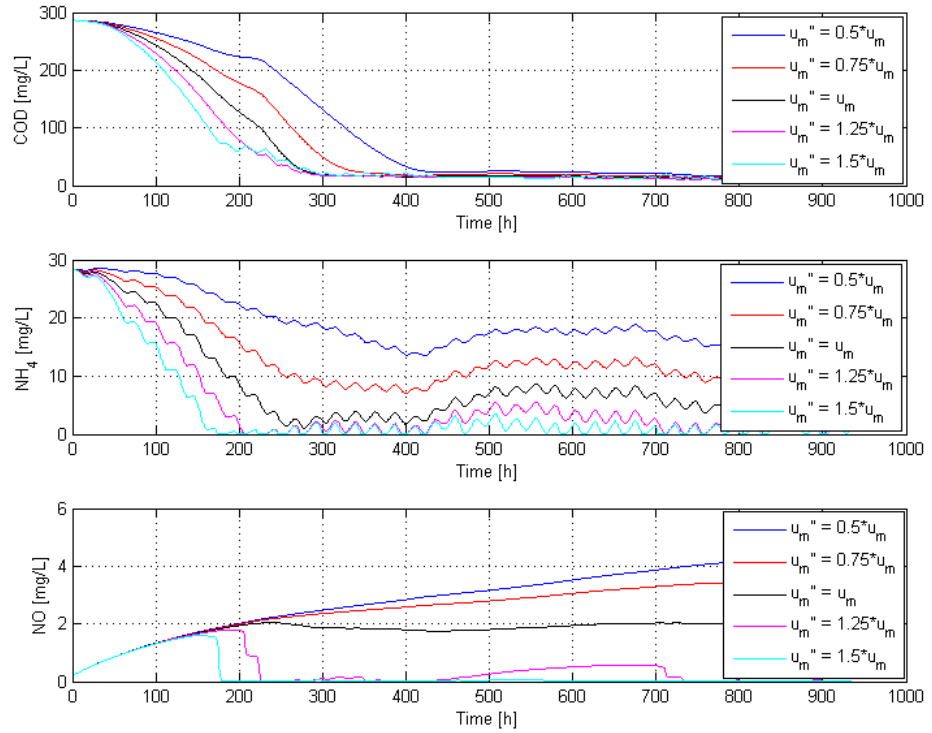

Figure D.13: Sensitivity analysis of parameter μ_m in WWT model.

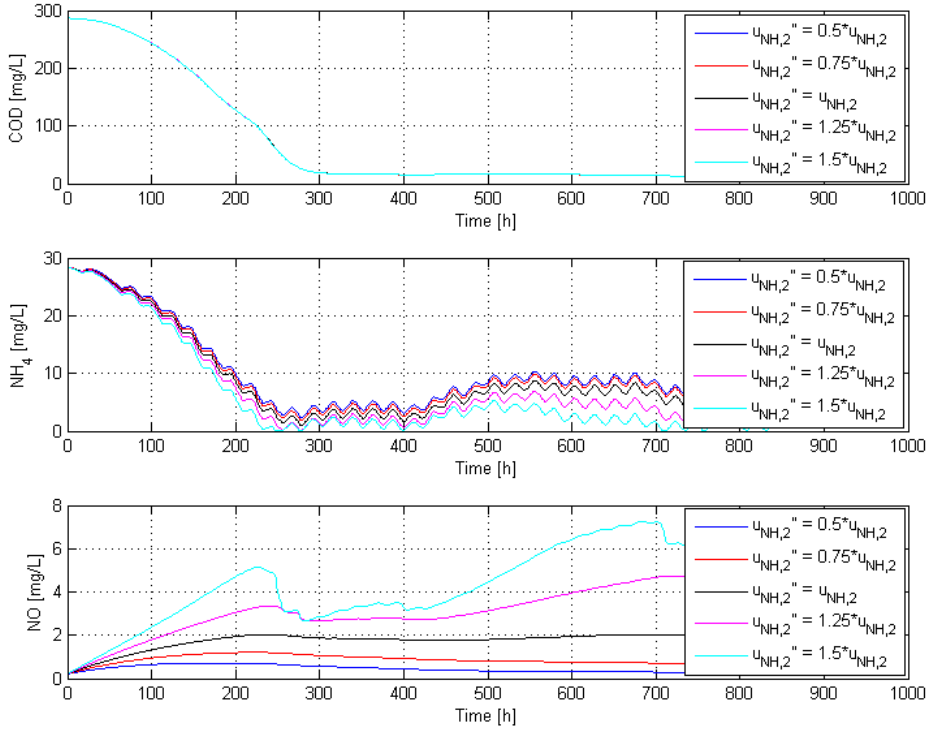

Figure D.14: Sensitivity analysis of parameter $\mu_{NH,2}$ in WWT model.

Appendix E

Results AD Model

As it was said, in this work a simplification of model designed by Mairet et al., 2011 (see [5]), in particular it was assumed pH constant, and neither inorganic carbon nor inert charge imbalance were not taken into in consideration, that is why just parameters showed in Table E.1 were used in simulations.

Parameter	Value	Unit
β_1	0.3	$[g\ COD/g\ COD]$
β_2	0.4	$[g\ COD/g\ COD]$
k_1	12.5	$[g\ COD/g\ COD]$
k_2	0.0062	$[mol/g\ COD]$
k_3	11.5	$[g\ COD/g\ COD]$
k_5	9.1	$[g\ COD/g\ COD]$
k_6	8.1	$[g\ COD/g\ COD]$
k_7	0.054	$[g\ COD/g\ COD]$
k_9	20	$[g\ COD/g\ COD]$
k_{10}	0.0062	$[mol/g\ COD]$
k_{11}	0.30	$[mol/g\ COD]$
$\overline{\mu_1}$	0.3	$[1/day]$
$\overline{\mu_2}$	0.053	$[1/day]$
$\overline{\mu_3}$	0.14	$[1/day]$
K_{S1}	2.11	$[g\ COD/g\ COD]$
K_{S2}	0.056	$[g\ COD/g\ COD]$
K_{S3}	0.02	$[g\ COD/L]$
K_{I3}	16.4	$[g\ COD/L]$
$K_{I_{NH_3}}$	0.0018	$[M]$

Table E.1: Parameters AN ecosystem.

Simulations under these assumptions are showed in Figure E.1.

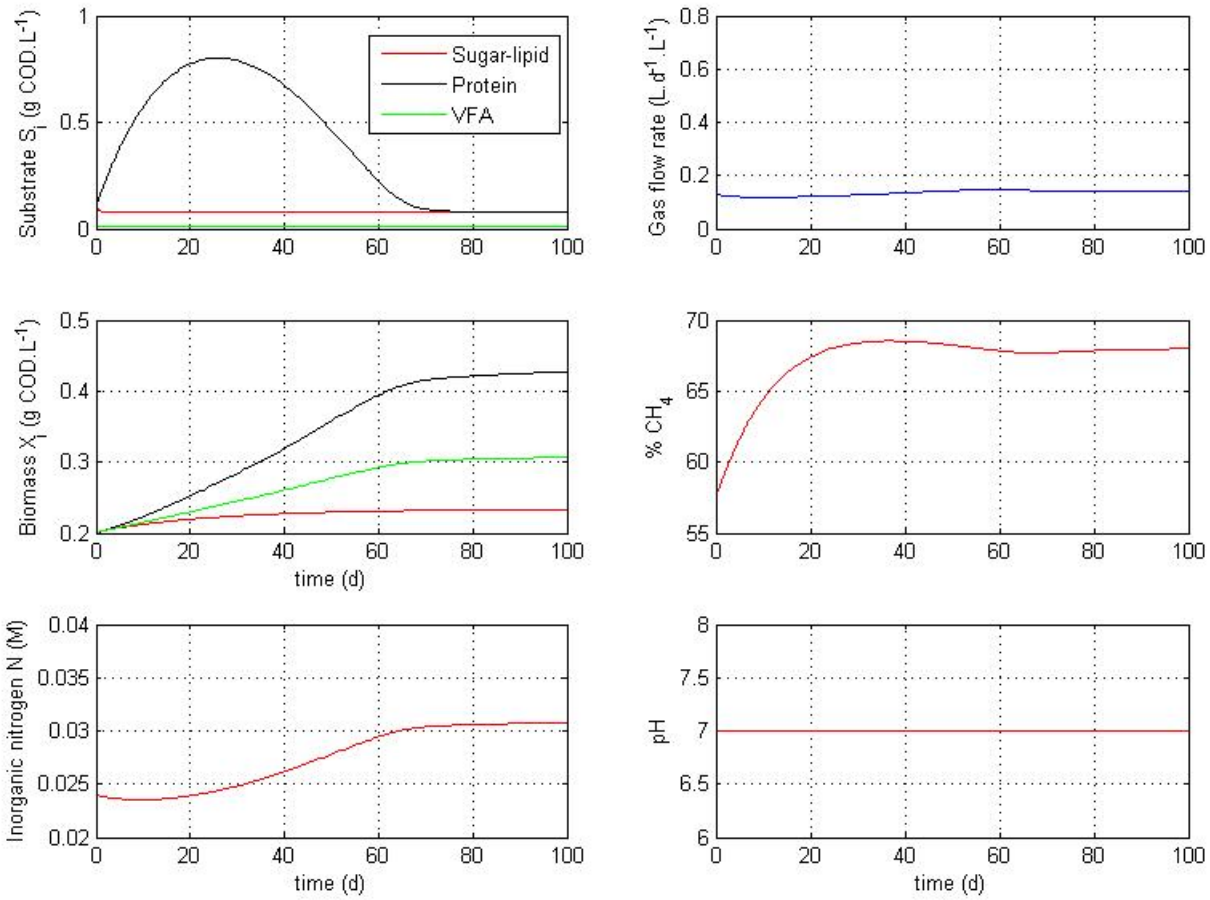

Figure E.1: Graphs AN model.

Appendix F

Deduction General Arrangement

In Figure F.1 it is possible to see all possibles configurations.

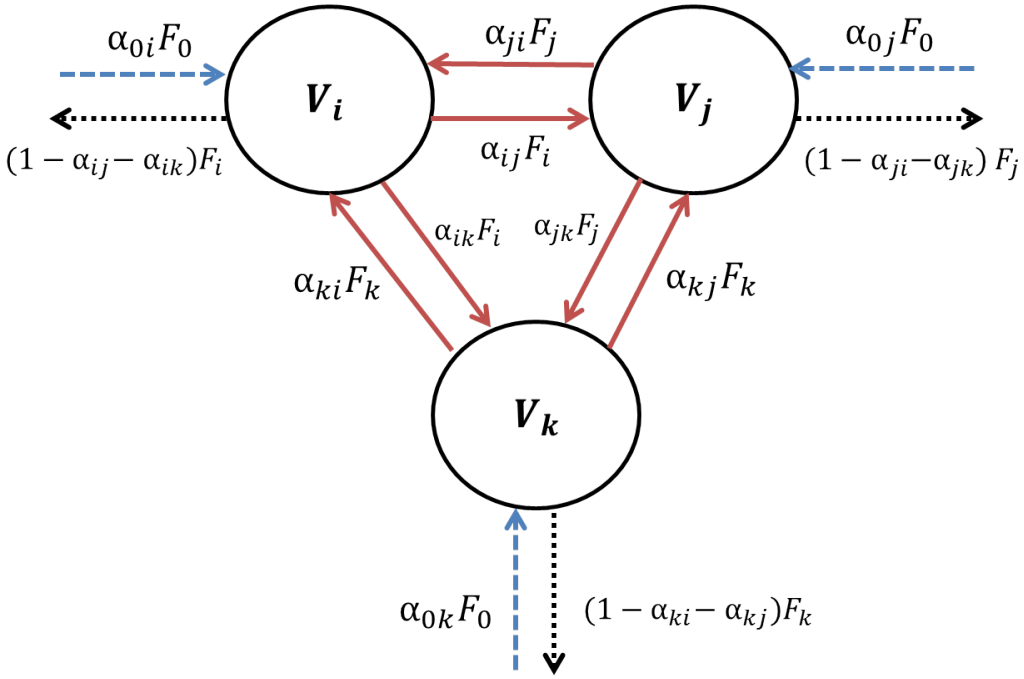

Figure F.1: All possible arrangements. Blue, black and red arrows are input, output and interior flows, respectively.

So, the mass balance for each ecosystem are showed in the following equation system:

$$F_i = \alpha_{0i}F_0 + \alpha_{ji}F_j + \alpha_{ki}F_k \tag{F.1}$$
$$F_j = \alpha_{0j}F_0 + \alpha_{ij}F_i + \alpha_{kj}F_k \tag{F.2}$$
$$F_k = \alpha_{0k}F_0 + \alpha_{jk}F_j + \alpha_{ik}F_i \tag{F.3}$$

Replacing equation (F.1) in (F.2), we will have:

$$F_j = \alpha_{0j}F_0 + \alpha_{ij}\left(\alpha_{0i}F_0 + \alpha_{ji}F_j + \alpha_{ki}F_k\right) + \alpha_{kj}F_k$$

Ordering terms:

$$\left(1 - \alpha_{ij}\alpha_{ji}\right)F_j - \left(\alpha_{ij}\alpha_{ki} + \alpha_{kj}\right)F_k = \left(\alpha_{0j} + \alpha_{ij}\alpha_{0i}\right)F_0 \tag{F.4}$$

Repeating the idea: replacing equation (F.1) in (F.3), we will have, after ordering terms:

$$-\left(\alpha_{kj} + \alpha_{ik}\alpha_{ji}\right)F_j + \left(1 - \alpha_{ik}\alpha_{ki}\right)F_k = \left(\alpha_{0k} + \alpha_{ik}\alpha_{0i}\right)F_0 \tag{F.5}$$

So, if we multiply (F.4) by $\frac{\alpha_{kj}+\alpha_{ik}\alpha_{ji}}{1-\alpha_{ij}\alpha_{ji}}$ plus (F.5), result is:

$$\left[\left(\alpha_{kj} + \alpha_{ik}\alpha_{ji}\right)\cdot\frac{\alpha_{kj} + \alpha_{ik}\alpha_{ji}}{1 - \alpha_{ij}\alpha_{ji}} - \left(1 - \alpha_{ik}\alpha_{ki}\right)\right]F_k = \left[\left(\alpha_{0j} + \alpha_{ij}\alpha_{0i}\right)\cdot\frac{\alpha_{kj} + \alpha_{ik}\alpha_{ji}}{1 - \alpha_{ij}\alpha_{ji}} + \left(\alpha_{0k} + \alpha_{ik}\alpha_{0i}\right)\right]F_0$$

$$\iff F_k = \left[\frac{(\alpha_{0j} + \alpha_{ij}\alpha_{0i})(\alpha_{jk} + \alpha_{ik}\alpha_{ji}) + (\alpha_{0k} + \alpha_{ik}\alpha_{0i})(1 - \alpha_{ij}\alpha_{ji})}{(1 - \alpha_{ik}\alpha_{ki})(1 - \alpha_{ij}\alpha_{ji}) - (\alpha_{ij}\alpha_{ki} + \alpha_{kj})(\alpha_{jk} + \alpha_{ik}\alpha_{ji})}\right]F_0$$

Therefore, we found an expression of F_k in function of F_0, which could be rewriteen as follow:

$$F_k = \gamma \cdot F_0 \tag{F.6}$$

Where:

$$\gamma = \frac{(\alpha_{0j} + \alpha_{ij}\alpha_{0i})(\alpha_{jk} + \alpha_{ik}\alpha_{ji}) + (\alpha_{0k} + \alpha_{ik}\alpha_{0i})(1 - \alpha_{ij}\alpha_{ji})}{(1 - \alpha_{ik}\alpha_{ki})(1 - \alpha_{ij}\alpha_{ji}) - (\alpha_{ij}\alpha_{ki} + \alpha_{kj})(\alpha_{jk} + \alpha_{ik}\alpha_{ji})}$$

Now, using (F.6) in (F.1) and in (F.2), respectively:

$$F_i - \alpha_{ji}F_j = \left(\alpha_{0i} + \alpha_{ki}\gamma\right)F_0 \tag{F.7}$$

$$-\alpha_{ij}F_i + F_j = \left(\alpha_{0j} + \alpha_{kj}\gamma\right)F_0 \tag{F.8}$$

If we multiply (F.8) by α_{ji} plus (F.7), result is:

$$F_i - \alpha_{ij}\alpha_{ji}Q_i = \left[\left(\alpha_{0i} + \alpha_{ki}\gamma\right) + \left(\alpha_{0j} + \alpha_{kj}\gamma\right)\alpha_{ji}\right]F_0$$

$$\iff F_i = \left[\frac{(\alpha_{0i} + \alpha_{ki} \cdot \gamma) + (\alpha_{0j} + \alpha_{ij} \cdot \gamma)\alpha_{ji}}{1 - \alpha_{ij}\alpha_{ji}}\right] F_0$$

So, F_i in function of F_0 could be rewritten as follow:

$$F_i = \varepsilon \cdot F_0 \tag{F.9}$$

Where:

$$\varepsilon = \frac{(\alpha_{0i} + \alpha_{ki} \cdot \gamma) + (\alpha_{0j} + \alpha_{ij} \cdot \gamma)\alpha_{ji}}{1 - \alpha_{ij}\alpha_{ji}}$$

Finally, (F.9) in (F.8)

$$F_j = \lambda \cdot F_0 \tag{F.10}$$

Where:

$$\lambda = \alpha_{ij} \cdot \varepsilon + \alpha_{0j} + \alpha_{kj} \cdot \gamma$$

These expression will be used to arise the mass balance of nitrogen and carbon. As it is known, the general mass balance is:

$$\text{Accumulation} = \text{Input} - \text{Output} + \text{Generation} - \text{Consumption} \tag{F.11}$$

If $Y_n = \{COD, N, microalgae\}$, with $n = \{i, j, k\}$. So in ecosystem i, we will have:

$$\frac{dY_i}{dt} = \frac{\alpha_{0i}F_iY_0}{V_i} + \frac{\alpha_{ji}F_jY_j}{V_i} + \frac{\alpha_{ki}F_kY_k}{V_i} - \frac{F_iY_i}{V_i} \pm f_i \tag{F.12}$$

Where f_i could be generation or consumption term. If we define $D_n = F_0/V_n$ for all $n = \{i, j, k\}$, so (F.12) could be rewritten in the following way, using (F.9),(F.10) and (F.6):

$$\frac{dY_i}{dt} = D_i \left(\alpha_{0i}Y_0 + \alpha_{ji} \cdot \lambda \cdot Y_j + \alpha_{ki} \cdot \gamma \cdot Y_k - \varepsilon \cdot Y_i\right) \pm f_i \tag{F.13}$$

Similarly with the others ecosystems:

$$\frac{dY_j}{dt} = D_j \left(\alpha_{0j}Y_0 + \alpha_{ij} \cdot \varepsilon \cdot Y_i + \alpha_{kj} \cdot \gamma \cdot Y_k - \gamma \cdot Y_j\right) \pm f_j \tag{F.14}$$

$$\frac{dY_k}{dt} = D_k \left(\alpha_{0k} Y_0 + \alpha_{ik} \cdot \varepsilon \cdot Y_i + \alpha_{jk} \cdot \lambda \cdot Y_j - \gamma \cdot Y_k \right) \pm f_k \tag{F.15}$$

Appendix G

Optimization codes

```
clear all
close all
clc
tic

%% Optimization
%p =  [(1)alp_0i (2)alp_ij (3)alp_ik (4)alp_0j (5)alp_ji (6)alp_jk (7)alp_0k
    (8)alp_ki (9)alp_kj (10)Vi (11)Vj (12)Vk]
%p0  = [0.33 0.33 0.33 0.34 0.33 0.33 0.33
         0.33 0.33 2 10.5 2];
p0  = [0.0012 0.0018 0.0029 0.1219 0.7959 0.2034 0.8769
    0.0001 0.1373 1.5740 6.8290 3.9237];



% Restriction: A*p <= b; alp_ij+alp_ik<=1; alp_jk+alp_ji<=1;alp_ki+alp_kj<=1;
    Vi+Vj+Vk<=25*1880
% (Output of each ecosystem can not be negative, where output is for example
    :1-alp_ij-alp_ik)
M = 1.88*25; % M = 47 [m^3]
A = [0 1 1 0 0 0 0 0 0 0 0 0; 0 0 0 0 1 1 0 0 0 0 0 0; 0 0 0 0 0 0 0 1 1 0 0
    0; 0 0 0 0 0 0 0 0 0 1 1 1];
b = [1 1 1 M];

% Restriction: Aeq*p = beq;  alp_0i+alp_0j+alp_0k=1 (all input enters to the
    system)
Aeq = [1 0 0 1 0 0 1 0 0 0 0 0];
beq = 1;

% Restriction: alp_mn in [0,1] and Vn>=1 [L].
lb = [0 0 0 0 0 0 0 0 0 1 1 1];
ub = [1 1 1 1 1 1 1 1 1 M M M];

options = optimoptions(@fmincon,'Algorithm','interior-point');
[p,OF] = fmincon(@objective_function,p0,A,b,Aeq,beq,lb,ub,@rest,options)
toc

%% %%%%%%%%% Anaerobic %%%%%%%%%%%%%%%
% Initial conditions
```

```
Xi(1) = 0.1;     % S1
Xi(2) = 0.2;     % X1
Xi(3) = 0.1;     % S2
Xi(4) = 0.2;     % X2
Xi(5) = 0.02;    % S3
Xi(6) = 0.024;   % N
Xi(7) = 0.2;     % SI
Xi(8) = 0.2;     % X3
Xi(9) = 0.65;   % PCH4


%% %%%%%%%%%%%%%%%%%% MP %%%%%%%%%%%%%%%%%%%%%%
% Initials Conditiones

CODexp = 297;
kin    = 0.0523;

Xi(10) = 0.0325*CODexp;           % Biomass: Something important: Initial
    biomass plus initial COD should be equal to 297
Xi(11) = (1-0.0325)*CODexp;       % COD: According to experimental data "LHRA:
    DCO filtré"
Xi(12) = 28.39;                     % N/NH4: According to experimental data
Xi(13) = 0.216;
Xi(14) = kin*CODexp;              % Norg = Ntot,filt - NH4 - NO2 - NO3



%% %%%%%%%%%%%%%%%%%%%% WWT %%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%

Xi(15) = 0.0325*CODexp;             % Microalgae: Something important: Initial
    biomass plus initial COD should be equal to 297
Xi(16) = 0.0025*CODexp;             % Nitrifiers: Something important: Initial
    biomass plus initial COD should be equal to 297
Xi(17) = (1-0.0325-0.0025)*CODexp; % COD: According to experimental data "LHRA
    : DCO filtré"
Xi(18) = 28.39;                     % N/NH4: According to experimental data
Xi(19) = 0.216;                     % N/(NO3+NO2): According to experimental
    data
Xi(20) = kin*CODexp;                % Norg = Ntot,filt - NH4 - NO2 - NO3



%% Solver
tin    = 0;
tfin   = 5000;
inter = 0.1;
tspan = [tin:inter:tfin];
[time1 X]=ode45(@ODEs_W_A_N_p,tspan,Xi,[],p);

time = time1/24; % From hours to days

%% Graphs

% %%%%%%%%%%%%%% MP %%%%%%%%%%
figure
subplot(3,2,1)
```

```
plot(time,X(:,11),'r')
ylabel('COD MP [mg COD/L]');
axis([0 time(end)+2 0 max(X(:,11))+10])
grid on

subplot(3,2,2)
plot(time,X(:,12)+X(:,13),'b')
axis([0 time(end)+2 0 max(X(:,12)+X(:,13))+10])
ylabel('Inorg. nitrogen MP [mg N/L]');
grid on

subplot(3,2,3)
plot(time,(X(:,5))*1e3,'r')
ylabel('COD AN [mg COD/L]')
axis([0 time(end)+2 0 max((X(:,5))*1e3)+10])
grid on

subplot(3,2,4)
plot(time,X(:,6)*(14e3),'b')
ylabel('Inorg. nitrogen AN [mg N/L]')
axis([0 time(end)+2 0 max(X(:,6))*(14e3)+10])
grid on

%%%%%%%%% WWT %%%%%%%%%%%%%%%%%%
subplot(3,2,5)
plot(time,X(:,17),'r')
xlabel('Time [h]');
ylabel('COD WWT [mg COD/L]');
axis([0 time(end)+2 0 max(X(:,17))+10])
grid on

subplot(3,2,6)
plot(time,X(:,18)+X(:,19),'b')
xlabel('Time [days]');
ylabel('Inorg. nitrogen WWT [mg N/L]');
axis([0 time(end)+2 0 max(X(:,18)+X(:,19))+10])
grid on


%%%% Methane %%%%%%%%%%
figure
plot(time,X(:,9),'r')
ylabel('Methane Generated [mol]')
%legend(sprintf('Recicle= %0.5g.',maximum))
axis([0 time(end)+2 0 max(X(:,9))+10])
xlabel('Time [days]');
grid on


%%%%%%%% Microorganism %%%%%%%%
figure
subplot(3,1,1)
plot(time,X(:,10),'b')
ylabel('Microalgae MP [mg COD/L]');
axis([0 time(end)+2 0 max(X(:,10))+10])
grid on
```

```
139
subplot(3,1,2)
141 plot(time,X(:,2)*1e3,'r',time,X(:,4)*1e3,'k',time,X(:,8)*1e3,'g')
ylabel('Biomass X_i AN [mg COD/L]')
143 axis([0 time(end)+2 0 210])
legend('X_1','X_2','X_3')
145 grid on

147 subplot(3,1,3)
plot(time,X(:,15),'b',time,X(:,16),'r')
149 legend('Microalgae','Nitrifiers')
xlabel('Time [days]');
151 ylabel('Biomass WWT [mg COD/L]');
axis([0 time(end)+2 0 max(X(:,15))+10])
153 grid on

155 %%%% Anaerobic Information %%%%%%%%
figure
157 plot(time,X(:,1)*1e3,'r',time,X(:,3)*1e3,'k',time,X(:,5)*1e3,'g')
ylabel('Substrate S_i [mg COD/L]')
159 xlabel('Time [days]')
axis([0 time(end)+2 0 60])
161 legend('Sugar-lipid','Protein','VFA')
grid on
163

%% Print Information
165

NonNegativeOutput = A*p'
167 SumOne = Aeq*p'

169 % Connections/Recycle
% i
171 alp_0i  = p(1);      % Percentage recycle
alp_ij  = p(2);
173 alp_ik  = p(3);

175 % j
alp_0j  = p(4);      % Percentage recycle
177 alp_ji  = p(5);
alp_jk  = p(6);
179
% k
181 alp_0k  = p(7);      % Percentage recycle
alp_ki  = p(8);
183 alp_kj  = p(9);


185
% Factors
187 Q0 = 12.99

189 gam = ((alp_0j+alp_ij*alp_0i)*(alp_jk+alp_ik*alp_ji)+(alp_0k+alp_ik*alp_0i)
    *(1-alp_ij*alp_ji))/((1-alp_ik*alp_ki)*(1-alp_ij*alp_ji)-(alp_ij*alp_ki+
    alp_kj)*(alp_jk+alp_ik*alp_ji));
Qk = gam*Q0
191

eps = ((alp_0i+alp_ki*gam)+(alp_0j+alp_kj*gam)*alp_ji)/(1-alp_ij*alp_ji);
```

```
193 Qi = eps*Q0

195 lam = alp_ij*eps+alp_0j+alp_kj*gam;
    Qj = lam*Q0
197
    Fout = [(1-alp_ij-alp_ik)*Qi  (1-alp_ji-alp_jk)*Qj (1-alp_ki-alp_kj)*Qk]'
199
    %% Restrictions
201 %tss = 3000;
    tss_index = 3000/inter; % tss = 3000 [h], but tss will be an index in the
        following vectors, that is why is divided by inter
203
    Fout = [(1-alp_ij-alp_ik)*Qi  (1-alp_ji-alp_jk)*Qj (1-alp_ki-alp_kj)*Qk]';
205
    % Nitrogen Flows
207 Ni = Fout(1)*(X(tss_index:end,12)+X(tss_index:end,13));
    Nj = Fout(2)*(X(tss_index:end,6))*(14*1e3);
209 Nk = Fout(3)*(X(tss_index:end,18)+X(tss_index:end,19));
    %N = (Ni+Nj+Nk)/(Qi+Qj+Qk);
211
    Ci = Fout(1)*(X(tss_index:end,11));
213 Cj = Fout(2)*(X(tss_index:end,5))*1e3;
    Ck = Fout(1)*(X(tss_index:end,17));
215
    % Inputs
217 kin   = 0.0523;
    NH40  = 48.67; % NH4
219 NO0   = 0.19;
    C0    = 370.32;
221 Norg0 = kin*C0;

223 % Output nitrogen requirements
    Nitrogen = mean((Ni+Nj+Nk)/(Fout(1)+Fout(2)+Fout(3)))-15
                                                    % N_out < 15
225 COD = mean((Ci+Cj+Ck)/(Fout(1)+Fout(2)+Fout(3)))-125
                                                        % COD_out < 125
    Nitrogen_remotion = (C0-mean((Ci+Cj+Ck)/(Fout(1)+Fout(2)+Fout(3))))*100/C0
                                    % Remotion > 75%
227 COD_remotion  = ((NH40+NO0+Norg0)-mean((Ni+Nj+Nk)/(Fout(1)+Fout(2)+Fout(3))))
        *100/(NH40+NO0+Norg0)     % Remotion > 75%

229 % Mass balance
    EcosystemFlowBalance = [alp_0i*Q0+alp_ji*Qj+alp_ki*Qk-Qi alp_0j*Q0+alp_ij*Qi+
        alp_kj*Qk-Qj alp_0k*Q0+alp_jk*Qj+alp_ik*Qi-Qk]
231 GlobalFlowBalance = Fout(1)+Fout(2)+Fout(3)-Q0
```

```
    function OF = objective_function(p)
2   Xi    = zeros(20,1);
    %% %%%%%%%%% Anaerobic %%%%%%%%%%%%%%%%
4   % Initial conditions
    Xi(1) = 0.1;    % S1
6   Xi(2) = 0.2;    % X1
    Xi(3) = 0.1;    % S2
8   Xi(4) = 0.2;    % X2
    Xi(5) = 0.02;   % S3
10  Xi(6) = 0.024;  % N
```

```
Xi(7) = 0.2;     % SI
Xi(8) = 0.2;     % X3
Xi(9) = 0.65;  % PCH4


%% %%%%%%%%%%%%%%%%%% MP %%%%%%%%%%%%%%%%%%%%%%
% Initials Conditiones

CODexp = 297;
kin    = 0.0523;

Xi(10) = 0.0325*CODexp;           % Biomass: Something important: Initial
    biomass plus initial COD should be equal to 297
Xi(11) = (1-0.0325)*CODexp;       % COD: According to experimental data "LHRA:
    DCO filtré"
Xi(12) = 28.39;                     % N/NH4: According to experimental data
Xi(13) = 0.216;
Xi(14) = kin*CODexp;              % Norg = Ntot,filt - NH4 - NO2 - NO3



%% %%%%%%%%%%%%%%%%%%%% WWT %%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%

Xi(15) = 0.0325*CODexp;             % Microalgae: Something important: Initial
    biomass plus initial COD should be equal to 297
Xi(16) = 0.0025*CODexp;             % Nitrifiers: Something important: Initial
    biomass plus initial COD should be equal to 297
Xi(17) = (1-0.0325-0.0025)*CODexp; % COD: According to experimental data "LHRA
    : DCO filtré"
Xi(18) = 28.39;                     % N/NH4: According to experimental data
Xi(19) = 0.216;                     % N/(NO3+NO2): According to experimental
    data
Xi(20) = kin*CODexp;                % Norg = Ntot,filt - NH4 - NO2 - NO3


%% Solver
tin    = 0;
tfin   = 5000;
inter = 0.1;
tspan = [tin:inter:tfin];
%tspan = [tin tfin];
%[time X]=ode45(@ODEs_W_A_N_p,tspan,Xi,[],p,influent,I_0);
[time X]=ode45(@ODEs_W_A_N_p,tspan,Xi,[],p);

%% Connections/Recycle
% i
alp_0i  = p(1);       % Percentage recycle
alp_ij  = p(2);
alp_ik  = p(3);

% j
alp_0j  = p(4);       % Percentage recycle
alp_ji  = p(5);
alp_jk  = p(6);

% k
```

```
alp_0k  = p(7);        % Percentage recycle
alp_ki  = p(8);
alp_kj  = p(9);


%% Factors
Q0 = 12.99;

gam = ((alp_0j+alp_ij*alp_0i)*(alp_jk+alp_ik*alp_ji)+(alp_0k+alp_ik*alp_0i)
    *(1-alp_ij*alp_ji))/((1-alp_ik*alp_ki)*(1-alp_ij*alp_ji)-(alp_ij*alp_ki+
    alp_kj)*(alp_jk+alp_ik*alp_ji));
Qk = gam*Q0;

eps = ((alp_0i+alp_ki*gam)+(alp_0j+alp_kj*gam)*alp_ji)/(1-alp_ij*alp_ji);
Qi = eps*Q0;

lam = alp_ij*eps+alp_0j+alp_kj*gam;
Qj = lam*Q0;

Fout = [(1-alp_ij-alp_ik)*Qi  (1-alp_ji-alp_jk)*Qj (1-alp_ki-alp_kj)*Qk]';

%% %%%%%%%%%% Objective Function %%%%%%%%%%%%%%%%%%
tss = 3000;
tss_index = 3000/inter; % tss = 3000 [h], but tss will be an index in the
    following vectors, that is why is divided by inter

%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%
%%%%%%%%%%%%%%%%%%%%%  Methane %%%%%%%%%%%%%%%%%%%%%%%%%%%%
%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%
%
% OF = -(X(end,9)-X(tss_index,9))*0.254

%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%
%%%%%%%%%%%%%%%%%%%%% Money %%%%%%%%%%%%%%%%%%%%%%%%%%%%
%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%


 Methane_in_dolar = (X(end,9)-X(tss_index,9))*2.18e-2;
                                                % 2.18e-2 [dolar/mol CH4]
 Biomass_in_dolar = sum(X(tss_index:end,10)*Fout(1)+X(tss_index:end,15)*Fout(3)
     )*1e-5;      % 1e-5  [dolar/mg Biomass]
OF  = -(Methane_in_dolar+Biomass_in_dolar)

%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%
%%%%%%%%%%%%%%%%%%%%%  Energy %%%%%%%%%%%%%%%%%%%%%%%%%%%%%
%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%

% Methane_energy = (X(end,9)-X(tss_index,9))*0.254; % 1 mol methane -> 0.254
    kWh
% lipid_content = 0.3;
% Biomass_energy = sum(X(tss_index:end,10)*Fout(1)+X(tss_index:end,15)*Fout(3)
     )*lipid_content*1.05e-5 % 1 mg lipid -> 1.05e-5 kWh
% OF = -(Methane_energy+Biomass_energy)

%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%
```

```
p

end
```

```
function [c,ceq] = rest(p)
Xi    = zeros(20,1);

%% %%%%%%%%% Anaerobic %%%%%%%%%%%%%%%%
% Initial conditions
Xi(1) = 0.1;     % S1
Xi(2) = 0.2;     % X1
Xi(3) = 0.1;     % S2
Xi(4) = 0.2;     % X2
Xi(5) = 0.02;    % S3
Xi(6) = 0.024;   % N
Xi(7) = 0.2;     % SI
Xi(8) = 0.2;     % X3
Xi(9) = 0.65;   % PCH4


%% %%%%%%%%%%%%%%%% MP %%%%%%%%%%%%%%%%%%%%%%
% Initials Conditiones

CODexp = 297;
kin    = 0.0523;

Xi(10) = 0.0325*CODexp;          % Biomass: Something important: Initial biomass plus initial COD should be equal to 297
Xi(11) = (1-0.0325)*CODexp;      % COD: According to experimental data "LHRA: DCO filtré"
Xi(12) = 28.39;                  % N/NH4: According to experimental data
Xi(13) = 0.216;
Xi(14) = kin*CODexp;             % Norg = Ntot,filt - NH4 - NO2 - NO3



%% %%%%%%%%%%%%%%%%%%%% WWT %%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%

Xi(15) = 0.0325*CODexp;          % Microalgae: Something important: Initial biomass plus initial COD should be equal to 297
Xi(16) = 0.0025*CODexp;          % Nitrifiers: Something important: Initial biomass plus initial COD should be equal to 297
Xi(17) = (1-0.0325-0.0025)*CODexp; % COD: According to experimental data "LHRA: DCO filtré"
Xi(18) = 28.39;                  % N/NH4: According to experimental data
Xi(19) = 0.216;                  % N/(NO3+NO2): According to experimental data
Xi(20) = kin*CODexp;             % Norg = Ntot,filt - NH4 - NO2 - NO3

%% %%%%%%%%%%%%%%%%%%%% Nitrifiers %%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%

Xi(14) = 0.0325*CODexp;          % Microalgae: Something important: Initial biomass plus initial COD should be equal to 297
Xi(15) = 0.0025*CODexp;          % Nitrifiers: Something important: Initial biomass plus initial COD should be equal to 297
Xi(16) = (1-0.0325-0.0025)*CODexp;          % COD: According to experimental
```

```
    data "LHRA: DCO filtré"
Xi(17) = 28.39;                    % N/NH4: According to experimental data
Xi(18) = 0.216;                    % N/(NO3+NO2): According to experimental data
Xi(19) = kin*CODexp;               % Norg = Ntot,filt - NH4 - NO2 - NO3


%% Solver
tin    = 0;
tfin   = 5000;
inter = 0.1;
tspan = [tin:inter:tfin];
[time X]=ode45(@ODEs_W_A_N_p,tspan,Xi,[],p);

%% Connections/Recycle
% i
alp_0i  = p(1);       % Percentage recycle
alp_ij  = p(2);
alp_ik  = p(3);

% j
alp_0j  = p(4);       % Percentage recycle
alp_ji  = p(5);
alp_jk  = p(6);

% k
alp_0k  = p(7);       % Percentage recycle
alp_ki  = p(8);
alp_kj  = p(9);


%% Factors
Q0 = 12.99;

gam = ((alp_0j+alp_ij*alp_0i)*(alp_jk+alp_ik*alp_ji)+(alp_0k+alp_ik*alp_0i)
    *(1-alp_ij*alp_ji))/((1-alp_ik*alp_ki)*(1-alp_ij*alp_ji)-(alp_ij*alp_ki+
    alp_kj)*(alp_jk+alp_ik*alp_ji));
Qk = gam*Q0;

eps = ((alp_0i+alp_ki*gam)+(alp_0j+alp_kj*gam)*alp_ji)/(1-alp_ij*alp_ji);
Qi = eps*Q0;

lam = alp_ij*eps+alp_0j+alp_kj*gam;
Qj = lam*Q0;

%% Restrictions
%tss = 3000;
tss_index = 3000/inter; % tss = 3000 [h], but tss will be an index in the
    following vectors, that is why is divided by inter

Fout = [(1-alp_ij-alp_ik)*Qi  (1-alp_ji-alp_jk)*Qj (1-alp_ki-alp_kj)*Qk]';

% Nitrogen Flows
Ni = Fout(1)*(X(tss_index:end,12)+X(tss_index:end,13));
Nj = Fout(2)*(X(tss_index:end,6))*(14*1e3);
Nk = Fout(3)*(X(tss_index:end,18)+X(tss_index:end,19));
%N = (Ni+Nj+Nk)/(Qi+Qj+Qk);
```

```
Ci = Fout(1)*(X(tss_index:end,11));
Cj = Fout(2)*(X(tss_index:end,5))*1e3;
Ck = Fout(1)*(X(tss_index:end,17));

% Inputs
kin   = 0.0523;
NH40  = 48.67; % NH4
NO0   = 0.19;
C0    = 370.32;
Norg0 = kin*C0;

% Output nitrogen requirements
c(1) = mean((Ni+Nj+Nk)/(Fout(1)+Fout(2)+Fout(3)))-15;                    % N_out<15
c(2) = mean((Ci+Cj+Ck)/(Fout(1)+Fout(2)+Fout(3)))-125;                   % COD_out<125
c(3) = mean((Ci+Cj+Ck)/(Fout(1)+Fout(2)+Fout(3)))-0.25*C0;               % Remotion: 75%
c(4) = mean((Ni+Nj+Nk)/(Fout(1)+Fout(2)+Fout(3)))-0.25*(NH40+NO0+Norg0); % Remotion: 75%


% Mass balance
ceq(1) = alp_0i*Q0+alp_ji*Qj+alp_ki*Qk-Qi;
ceq(2) = alp_0j*Q0+alp_ij*Qi+alp_kj*Qk-Qj;
ceq(3) = alp_0k*Q0+alp_jk*Qj+alp_ik*Qi-Qk;
ceq(4) = Fout(1)+Fout(2)+Fout(3)-Q0;
ceq = [];
```

```
function dX = ODEs_W_A_N_p(t,X,p)
kin   = 0.0523;
L     = 0.4;       % Deep of the pond

%% Connections/Recycle
% i
alp_0i  = p(1);      % Percentage recycle
alp_ij  = p(2);
alp_ik  = p(3);
Vi = p(10)*1e3;

% j
alp_0j  = p(4);      % Percentage recycle
alp_ji  = p(5);
alp_jk  = p(6);
Vj = p(11)*1e3;


% k
alp_0k  = p(7);      % Percentage recycle
alp_ki  = p(8);
alp_kj  = p(9);
Vk = p(12)*1e3;

%% Factors

```

```
% Q_k = gam*Q_0
gamK = ((alp_0j+alp_ij*alp_0i)*(alp_jk+alp_ik*alp_ji)+(alp_0k+alp_ik*alp_0i)
    *(1-alp_ij*alp_ji))/((1-alp_ik*alp_ki)*(1-alp_ij*alp_ji)-(alp_ij*alp_ki+
    alp_kj)*(alp_jk+alp_ik*alp_ji));
% Q_i = eps*Q_0
epsI = ((alp_0i+alp_ki*gamK)+(alp_0j+alp_kj*gamK)*alp_ji)/(1-alp_ij*alp_ji);
% Q_j = lam*Q_0
lamJ = alp_ij*epsI+alp_0j+alp_kj*gamK;




%% Inlets
I0    = (max(0,39.8202*sin(0.2666*t -1.7771)))^2;
NH40   = 48.67;
NO0   = 0.19;

C0    = 370.32;
Norg0 = kin*C0;
Q0    = 12.99; % Average in [L/h]
X0    = 0;

b1 = 0.3;
b2 = 0.4;
bI = 0.3;

%% Importantes Variables

% i
Xi    = X(10);    % Microalgae
Ci    = X(11);    % COD
NH4i  = X(12);    % Nitrogen
NOi   = X(13);    % Nitrogen
Norgi = X(14);    % Nitrogen organic

% j
Xj = 0;         % Assumption: Anaerobic microorganism do not survive in other
    ecosystems, in particular, Microalgae
Cj = X(5);      % VFA: S3, in [g/L], to be coupled with any acosystem should
    enter in [mg/L]
Nj = X(6);      % N in [mol NH4/L], to be coupled with any acosystem should
    enter in [N mg/L], as a NH4 = 97% Nj and NO = 3% Nj
Norgj = 0;      % Nitrogen organic

% k
Xk    = X(15);            % Only microalgae. Assumption: Nitrifiers do not
    survive in other ecosystems
Ck    = X(17);            % COD
NH4k  = X(18);            % NH4
NOk   = X(19);            % NO
Norgk = X(20);            % Nitrogen organic



%% %%%%%%%%% j: ANAEROBIC %%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%
%Physio-chemical parameters
```

```
Knh3=1.1e-9;

% Coef stochio alpha_i
a(1) = 12.5;
a(2) = 0.00625;
a(3) = 11.5;
a(4) = 0.03;
a(5) = 9.1;
a(6) = 8.1;
a(7) = 0.054;
a(8) = 0.03;
a(9) = 20;
a(10) = 0.00625;
a(11) = 0.30;
a(12) = 0.20;

% coef kinetic
pa(1) = 0.3/24;
pa(2) = 2.11;
pa(3) = 0.053/24;
pa(4) = 0.03;
pa(5) = 0.14/24;
pa(6) = 0.02;
pa(7) = 16.4;
pa(8) = 0.0018;

ni=X(6);
h=10^-7;
nh3=Knh3/(Knh3+h)*ni;

% Contois
mu1 = pa(1)*X(1)/(X(1)+pa(2)*X(2));
mu2 = pa(3)*X(3)/(X(3)+pa(4)*X(4));
mu3 = pa(5)*(X(5)/(X(5)+pa(6)+(X(5)^2)/pa(7)))*(pa(8)/(pa(8)+nh3));


rhoT9 = a(11)*mu3*X(8);


% ODE Anaerobic
Dj=(Q0/Vj);

% S1[g COD/L]: b1 per cent of X(13)*1e-3 plus COD=X(14)*1e-3 (X(13/14)comes in
    mg/L)
dX(1)=Dj*((alp_0j*X0+alp_ij*epsI*Xi+alp_kj*gamK*Xk)*b1*1e-3+(alp_0j*C0+alp_ij*
    epsI*Ci+alp_kj*gamK*Ck)*1e-3-lamJ*X(1))-a(1)*mu1*X(2);
% X1
dX(2)=(mu1-lamJ*Dj)*X(2);
% S2[g COD/L]: b2 per cent of Xi (in mg/L) and X(14)+X(20)=Norg_{i,k} (in mg N
    /L) from COD.  Using protein: 6.63 [mmol N/g COD]
dX(3)=Dj*((alp_0j*X0+alp_ij*epsI*Xi+alp_kj*gamK*Xk)*b2*1e-3+(alp_0j*Norg0+
    alp_ij*epsI*Norgi+alp_kj*gamK*Norgk)*(1/(14*6.63))-lamJ*X(3))-a(5)*mu2*X(4)
    ;
% X2
dX(4)=(mu2-lamJ*Dj)*X(4);
% S3 [g COD/L]= Cj. Implies X(5) should be recycle with a factor 1e3
```

```
dX(5)=-Dj*lamJ*X(5)+ a(3)*mu1*X(2)+a(6)*mu2*X(4)-a(9)*mu3*X(8);
% NH4[M]=[mol NH4/L]: Coupled with ither ecosystem with a factor of *(18/(14e3
    )) because Ni and Nk come in [mg N/L]
dX(6)=Dj*((alp_0j*(NH40+NO0)+alp_ij*epsI*(NH4i+NOi)+alp_kj*gamK*(NH4k+NOk))
    *(1/(14e3))-lamJ*Nj)+ a(7)*mu2*X(4)-a(2)*mu1*X(2)-a(10)*mu3*X(8);
%dX(6)=Dj*((alp_0j*(NH40)+alp_ij*epsI*(NH4i)+alp_kj*gamK*(NH4k))*(1/(14*1e3))-
    lamJ*Nj)+ a(7)*mu2*X(4)-a(2)*mu1*X(2)-a(10)*mu3*X(8);
% S_I[g/L]:b3 per cent of X(13)*1e-3 (X(13)comes in mg/L)
dX(7)=Dj*((alp_0j*bI*X0+alp_ij*epsI*bI*Xi+alp_kj*gamK*bI*Xk)*1e-3-lamJ*X(7));
% X3
dX(8)=(mu3-lamJ*Dj)*X(8);
% P CH4 gaz
dX(9) = rhoT9*Vj;



%% %%%%%%%%%% i: MP %%%%%%%%%%%%
% Microalgae
umn     = 0.0204;       % umax of u(I)
KIn     = 44.4093;      % KI of u(I)
kn      = 0.1811;       % I=I0*exp(-kxz)
ucod    = 0.0115;       % umax of u(COD)
Kcod    = 21.3288 ;     % Kcod of u(COD)
R1      = 0.001;        % Respiration
Knh41   = 0.0127;       % Half saturation constant
Kno31   = 0.0047;       % Half saturation constant


% Nitrifiers
unh42   = 0.009;
Knh42   = 1.0461;
R2      = 0.0006;
k_NO2   = 2.2723;
k_NH2   = 2.3723;
k_N1    = 0.3871;
k_C     = 3.6537;

% Auxiliar variables to make easier to write equations
f_Im   = (1/(L*kn*Xi))*log((I0+KIn)/(I0*exp(-kn*L*Xi)+KIn));
f_Cm   = (X(11)/(X(11)+Kcod));
f_NHm  = (X(12)/(X(12)+Knh42));
f_NOm  = (X(13)/(X(13)+Kno31))*(Knh41/(X(12)+Knh41));

Di = (Q0/Vi);

% ODE MP
dX(10) = Di*(alp_0i*X0+alp_ji*lamJ*Xj+alp_ki*gamK*Xk-epsI*Xi)+(umn*f_Im+ucod*
    f_Cm)*(f_NHm+f_NOm)*Xi-R1*Xi;                                    %
    Microalgae
dX(11) = Di*(alp_0i*C0+alp_ji*lamJ*Cj*1e3+alp_ki*gamK*Ck-epsI*Ci)-k_C*ucod*
    f_Cm*(f_NHm+f_NOm)*Xi;                                           % COD
dX(12) = Di*(alp_0i*NH40+alp_ji*lamJ*Nj*1*(14*1e3)+alp_ki*gamK*NH4k-epsI*NH4i)
    -f_NHm*(k_N1*umn*f_Im+(k_N1-kin*k_C)*ucod*f_Cm)*Xi; % NH4
dX(13) = Di*(alp_0i*NO0+alp_ji*lamJ*Nj*0*(14*1e3)+alp_ki*gamK*NOk-epsI*NOi)-
    f_NOm*(k_N1*umn*f_Im+(k_N1-kin*k_C)*ucod*f_Cm)*Xi;      % NO
dX(14) = Di*(alp_0i*Norg0+alp_ji*lamJ*Norgj+alp_ki*gamK*Norgk-epsI*Norgi)-kin*
```

```
    k_C*ucod*f_Cm*f_NHm*Xi;
176
178 %% %%%%%%%%%%%%%%%%%%%% k: WWT %%%%%%%%%%%%%%%%%%%%

180
    % Auxiliar variables to make easier to write equations
182 f_Iw  = (1/(L*kn*X(15)))*log((I0+KIn)/(I0*exp(-kn*L*X(15))+KIn));
    f_C   = (X(17)/(X(17)+Kcod));
184 f_NHw = (X(18)/(X(18)+Knh41));
    f_NOw = (X(19)/(X(19)+Kno31))*(Knh41/(X(18)+Knh41));
186 f_NHw2 = (X(18)/(X(18)+Knh42));

188
    Dk = (Q0/Vk);
190
    %ODEs
192
    % Microalgae Balance
194 dX(15) = Dk*(alp_0k*X0+alp_ik*epsI*Xi+alp_jk*lamJ*Xj-gamK*Xk)+(umn*f_Iw+ucod*
        f_C)*(f_NHw+f_NOw)*Xk-R1*Xk;
    % Nitrifiers balance
196 dX(16) = -Dk*gamK*X(16)+unh42*f_NHw2*X(16)-R2*X(16);
    % COD Balance
198 dX(17) = Dk*(alp_0k*C0+alp_ik*epsI*Ci+alp_jk*lamJ*Cj*1e3-gamK*Ck)-k_C*ucod*f_C
        *(f_NHw+f_NOw)*X(15);
    % NH4 Balance: Assumption, 97% of Nj (in g/L) is NH4
200 dX(18) = Dk*(alp_0k*NH40+alp_ik*epsI*NH4i+alp_jk*lamJ*Nj*1*(14*1e3)-gamK*NH4k)
        -f_NHw*(k_N1*umn*f_Iw+(k_N1-kin*k_C)*ucod*f_C)*Xk-k_NH2*unh42*f_NHw2*X(16);
    % NO Balance
202 dX(19) = Dk*(alp_0k*NO0+alp_ik*epsI*NOi+alp_jk*lamJ*Nj*0*(14*1e3)-gamK*NOk)-
        f_NOw*(k_N1*umn*f_Iw+(k_N1-kin*k_C)*ucod*f_C)*Xk+k_NO2*unh42*f_NHw2*X(16);
    % Organic Nitrogen
204 dX(20) = Dk*(alp_0k*Norg0+alp_ik*epsI*Norgi+alp_jk*lamJ*Norgj-gamK*Norgk)-kin*
        k_C*ucod*f_C*(f_NHw+f_NOw)*Xk;

206 dX=dX';
```