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In Situ Bio-Methanation Modelling of a Randomly Packed Gas Stirred Tank Reactor (GSTR)

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Abstract: In situ Bio-Methanation (BM) is a recently developed biogas upgrading technique which finds application also in the Power to Gas (P2G) field. In this study a novel configuration of BM digester, the randomly packed Gas Stirred Tank Reactor (GSTR), was modelled. A 49 L reactor, in thermophilic conditions (55 °C) and at atmospheric pressure, was filled up with random packing on which the microbial populations could adhere. The feedstock used was Second Cheese Whey (SCW), liquid waste of cheese factories, rich in lactose (38 g/L), and its flowrate was chosen to obtain a Hydraulic Retention Time (HRT) of 30 days. The process was analyzed for different hydrogen inlet flowrates of 10 mL/min and 50 mL/min. The produced biogas was also recirculated in the reactor in order to transfer, into the liquid phase, as much hydrogen as possible. The model parameters were estimated by means of stationary state information of the reactor working without hydrogen injection, while a dynamical fitting was necessary to evaluate the value of the hydrogen mass transfer coefficient during BM. The model well described the reactor behavior and, by means of a dimensionless analysis in which the numbers of Stanton (St) and β were defined, it was found out that the mass transfer coefficient is the limiting step of the process.

Keywords: in situ bio-methanation; anaerobic digestion; bioreactors; modelling; dimensionless analysis

1. Introduction

Anaerobic digestion (AD) is a biological process used to convert organic waste material into biogas, a gaseous mixture mainly composed of CH_4 (~50–60%) and CO_2 (~50–40%). Due to its abundance in methane, biogas is suitable for energy and heat generation purposes [1,2]. Moreover, the removal of CO_2 from biogas by means of upgrading processes makes AD a sustainable source of CH_4. In such cases, the methane produced is directly injected in the already existent natural gas grids [3]. Currently, the most used biogas upgrading techniques such as membrane separation and adsorption are performed in specific units downwards the anaerobic digester. To avoid the costs related to the presence of additional units, an attractive solution which exploits the ability of anaerobic microorganisms to convert hydrogen and carbon dioxide in methane was proposed: the in situ bio methanation. The in situ bio-methanation consists of injecting hydrogen into an anaerobic digester in order to reduce and convert CO_2, produced during AD, to CH_4. This process finds a possible application also in the Power to Gas (P2G) field for the chemical storage of electrical energy produced by renewable sources [4,5]. In many studies, it was observed that the limiting step of the process is the hydrogen mass transfer to the liquid

phase [6,7], but to achieve a complete comprehension of the system behavior, the development of a mathematical model is crucial. The AD is composed of a series of different syntrophic biological pathways. Several AD models were developed with a different degree of complexity. Andrews et al. [8] developed a model considering only two distinct types of microbial populations which could predict the reactor failure for the decrease in pH due to Volatile Fatty Acids (VFAs) accumulation. Angelidaki et al. [9] realized a model which included the presence of three different types of bacteria and ammonia inhibition on the acetoclastic step. Zaher et al. [10] considered in the simulation of AD the hydrogenotrophic biomass. The most complete and complex models for the high numbers of parameters and for considering almost all the phenomena that take place in the AD are the "BioModel" developed by Angelidaki et al. [11] and the "ADM1" proposed by Batstone et al. [12]. In literature only a few models are available for the description of in situ biological methanation (BM). In particular, Lovato et al. [13] integrated the "BioModel" with the presence of the hydrogenotrophic pathway proposed by Hill et al. [14] for the description of a conventional CSTR with a working volume <2 L. Bensmann et al. [15] used the ADM1 model together with the characteristic growth kinetics of the *Methanobacterium thermoautotrophicum* obtained from the work of Shill et al. [16] to detect the limit working conditions (related to biological and mass transfer phenomena) of an anaerobic digester performing the biological methanation.

In order to reduce the computational complexity due to the high number of parameters that usually occur in biological models, the aim of this work is to propose a simple biological methanation model for the description of a randomly packed Gas Stirred Tank Reactor (GSTR) including in the model the presence of suspended attached growth media. Once estimated all the parameters of the model, a dimensionless analysis is performed to obtain information about the limiting steps of the process.

2. Materials and Methods

2.1. The Influent

The experiments were performed with a Second Cheese Whey (SCW) gotten from Caseificio Santa Maria s.r.l., a small dairy factory located in Rome, Italy. SCW is a lactose rich wastewater stream obtained from the production process of ricotta (a typical soft cheese of the Mediterranean region). According to this, the collected SCW had some fluctuations in its composition, but for modelling purposes, the average concentration of lactose was used as input of the system, and it was set to 38 g/L. Further details on the SCW used are reported in Lembo et al. [17].

2.2. The Process

The pilot plant is made of a 61 L randomly packed Gas Stirred Tank Reactor (GSTR) working in thermophilic (55 $\pm$ 1 °C) and atmospheric pressure conditions [17]. The liquid feed of SCW is introduced in the lower part of the reactor, while the digestate is collected in the upper part of the reactor in order to maintain a constant the liquid working volume of 49 L. The liquid flow rate was set to achieve a Hydraulic Retention Time (HRT) of 30 days. Part of the biogas stream is recirculated into the reactor to guarantee a homogeneous mixing of the nutrients into the liquid phase and to enhance the mass transfer between the liquid and the gaseous phases. Random packing material (SAGM 600, HDPE), with a specific surface area of 800 m^2/m^3, was placed in the central part of the reactor. The immobilized area occupied a volume of about 30 L.

From the bottom of the reactor, pure H_2 ($\geq$99%) obtained from an electrolyzer (DBS, model PGH2 100, Linde Gases Division, Pullach, Germany) was injected. Depending on whether H_2 was flowed into the reactor or not, in situ Bio-Methanation (BM) mode or Anaerobic Digestion (AD) mode were run respectively in separate steps. The AD was conducted until stationary conditions were reached (working time > 3 HRT (day)) setting the recirculation biogas flow to 4 L/min. For the BM a period of 43 days was investigated: 10 mL/min of H_2 was injected and the recirculation biogas flow was fixed to 4 L/min

for the first 7 days while 50 mL/min of H_2 was introduced, and the recirculation biogas flow was fixed to 6 L/min for other 34 days. A complete sketch of the process is given in Figure 1.
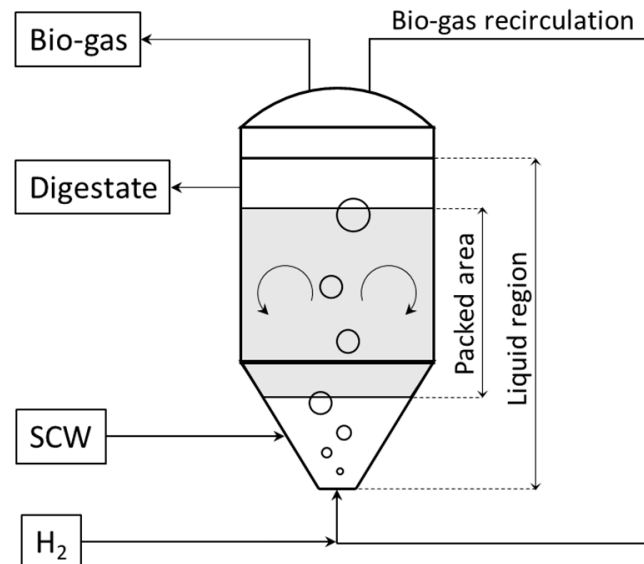

Figure 1. Schematic representation of the randomly packed gas stirred tank reactor. (GSTR).

2.3. Analytical Methods

Biogas composition was measured by an online Micro Gas Chromatograph Varian (GC4900). Biogas and H_2 flow rate were monitored by a digital online flow meters (EL-Flow select series, Bronkhorst High-Tech B.V, Ruurlo, The Netherlands). The VSS of the SCW, of the reactor effluent and of the immobilized part were analyzed following standards methods [18].

2.4. Model Assumptions and Description

2.4.1. Bio-Methanation Modeling

The complexity of the model is directly related to the number of bacteria populations taken into account [19]. In a conventional anaerobic digester, it was observed that the main contribution to methane generation is given by acetoclastic methanogens archaea while hydrogenotrophic methanogens played a minor role. The scenario changes when hydrogen is directly injected into the digester as happens with in situ bio-methanation where hydrogenotrophic methanogens are responsible of the conversion of CO_2 and H_2 into CH_4. In order to well represent the two working operation modes of the reactor under analysis (in situ BM and AD), it was decided to develop a suitable model for (AD) and extend its validity toward in situ BM by introducing a little modification. According to this consideration, the model developed takes into account two main groups of microbial populations for the description of AD and considers an additional biomass to deal with hydrogen consumption during in situ BM. More in detail, AD is completely reproduced in two phases: acidogenesis is the first phase of the process in which acidogenic bacteria (X_1) consume the organic substrate available $(S_1$, in this case lactose) and produce CO_2 and VFAs (S_2); methanogenesis is the last phase where acetoclastic methanogen archaea (X_2) consume the volatile fatty acids, which are assumed to behave like pure acetic acid, to generate CO_2 and CH_4. A crucial aspect of AD is the substrate inhibition of acetoclastic methanogen archaea which may cause a strong pH decrease and eventually may lead to process failure [8]. It is important to underline that no kind of ion inhibition was assumed in this model, thus the ion balances were not considered. Since lactose is a dimer, the hydrolytic step, carried out by extracellular enzymes [20], was assumed to be faster than all the others and therefore neglected. The same assumptions for AD modeling were

already adopted by Bernard et al. [19]. In situ BM was reproduced including the biomass of *Methanobacterium thermoautotrophicum* (X_3) as responsible for H_2 and CO_2 consumption and CH_4 generation. Such microbial species, belonging to the *Methanobacteriaceae* family, was found to be, in previous analysis made on the same reactor [17], the most abundant among all the hydrogenotrophic community. A block diagram of the whole biological model is provided in Figure 2.
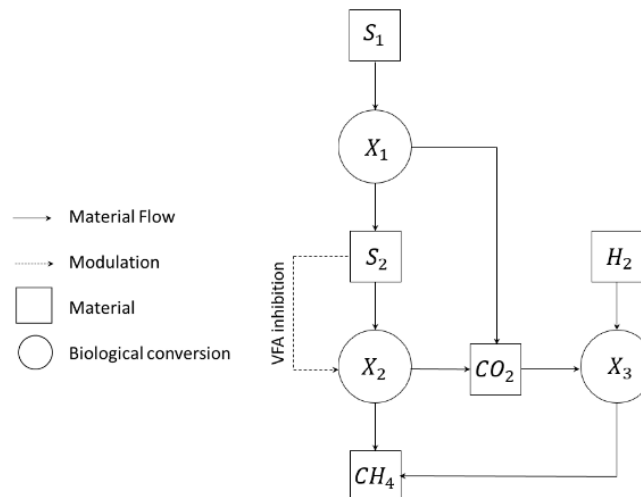

Figure 2. Outline of the biogas model.

2.4.2. Biological Reactions: Stoichiometry and Kinetics

The biological reactions that occur in the system are related to the metabolism of the different types of microorganisms considered.

*Acidogenesis* stoichiometry is described in Equation (1) as follows:

$$k_1 S_1 \rightarrow X_1 + k_2 S_2 + k_4 CO_2 \tag{1}$$

where S_1 is the organic substrate consumed by the biomass X_1, S_2 are the volatile fatty acids which were assumed to behave like pure acetic acid, and k_i are the reaction yields referred to the biomass growth. The reaction rate of Equation (1) is (r_1), and it was assumed to follow the Monod kinetics depending on the availability of substrate S1 in the liquid bulk, thus (Equation (2))

$$(r_1) = \mu_{1,max} \frac{S_1}{K_{S_1} + S_1} X_1 \tag{2}$$

where $\mu_{1,max}$ is the maximum specific growth rate, K_{S_1} is the half saturation constant, and X_1 is the acidogenic biomass concentration.

Acetoclastic methanogenesis stoichiometry is reported in Equation (3) as follows:

$$k_3 S_2 \rightarrow X_2 + k_6 CH_4 + k_5 CO_2 \tag{3}$$
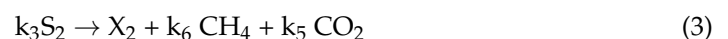
The reaction rate of Equation (3) is (r_2), and it was modelled by the Haldane expression which is commonly utilized for substrate inhibited kinetics:

$$(r_2) = \mu_{2,max} \frac{S_2}{K_{S_2} + S_2 + \frac{S_2^2}{K_I}} X_2 \tag{4}$$

where $\mu_{2,max}$ is the maximum specific growth rate, K_{S2} is the half saturation constant, K_I is the inhibition constant, and X_2 is the acetocastic methanogen concentration.

Hydrogenotrophic methanogenesis results from the metabolism of *Methanobacterium thermoautotrophicum* which is composed of a reaction for cellular growth (ν_G) and a reaction

for cellular maintenance [21] (ν_M) that are reported in Equation (5) and Equation (6), respectively.

$$H_2 + k_9 \, CO_2 \rightarrow k_8 \, X_3 + k_{10} \, CH_4 \tag{5}$$

$$H_2 + \frac{1}{4} \, CO_2 \rightarrow \frac{1}{2} \, H_2O + \frac{1}{4} \, CH_4 \tag{6}$$
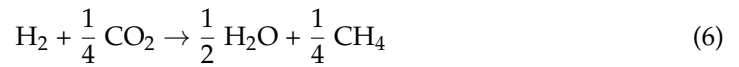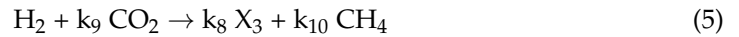
The overall hydrogen rate of consumption was assumed to have a saturation kinetics as adopted by Schill et al. [16]

$$\left(r_{H_2}\right) = \nu_G + \nu_M = q_{max} \frac{C_{H_2}^L}{K_{H_2} + C_{H_2}^L} X_3 \tag{7}$$

where q_{max} is the maximum hydrogen specific growth rate, K_{H_2} is the half saturation constant, $C_{H_2}^L$ is the hydrogen concentration dissolved in the liquid phase, and X_3 is the hydrogenotrophic methanogen bacteria concentration. The rate of maintenance is expressed as follows:

$$\nu_M = m \cdot X_3 \tag{8}$$

where m is the maintenance coefficient. According to Equation (7) and Equation (8) the rate of CH_4 production, CO_2 consumption and biomass growth are:

$$\left(r_{CH_4}\right) = k_{10}\left(r_{H_2} + \nu_M\right) + \frac{1}{4}\nu_M \tag{9}$$

$$\left(r_{CO_2}\right) = k_9\left(-r_{H_2} + \nu_M\right) - \frac{1}{4}\nu_M \tag{10}$$

$$\left(r_X\right) = k_8\left(r_{H_2} - \nu_M\right) \tag{11}$$

The kinetic parameters used in this work referred to all the biological reactions are listed in Table 1.

Table 1. Value of the kinetic parameters referred to all the biological reactions.

Parameter	Meaning	Unit	Value	Reference
$\mu_{1,max}$	Maximum acidogenic biomass growth rate	d^{-1}	1.2	[22]
K_{S1}	Half saturation constant associated with S_1	$g \, L^{-1}$	0.78	[19]
$\mu_{2,max}$	Maximum acetoclastic methanogenic biomass growth rate	d^{-1}	7.1	[19]
K_{S2}	Half saturation constant associated with S_2	$mmol \, L^{-1}$	9.28	[19]
K_I	Inhibition constant associated with S_2	$mmol \, L^{-1}$	256	[19]
q_{max}	Maximum hydrogen specific production rate	$mol \, g^{-1} \, d^{-1}$	21.3	[16]
K_{H2}	Half saturation constant associated with C^L_{H2}	$\mu mol \, L^{-1}$	5.6	[16]
m	maintenance coefficient	$mol \, g^{-1} \, d^{-1}$	1.72	[16]

2.4.3. Mass Balances

In this section are provided all the mass balances of the system which are crucial to evaluate the performances of the process. Both the liquid and the gaseous phases are assumed to be perfectly mixed as happens in a Continuous Stirred Tank Reactor (CSTR). The macroscopic mass balances were written including in the control volume the gas recirculation stream which influences only the gas-liquid mass transfer coefficient.

Liquid phase. All the biochemical reactions take place in the liquid phase. In order to consider the presence of random packing, it was assumed that only a fraction of the total biomass is homogeneously dispersed in the liquid phase while the other part is immobilized on the packing rings. Such fraction of "free" biomass was identified introducing the parameter α as done in the work of Bernard et al. [19]. According to such consider-

ation, the mass balances for biomass growth, one for each type of microbial population (X_1, X_2, X_3), are:

$$\frac{dX_i}{dt} = (r_i) - \alpha D X_i \tag{12}$$

where D represents the dilution factor which is the reciprocal of the HRT. The material balances for the substrates and the dissolved gaseous components are expressed in Equation (13):

$$\frac{dC_i^L}{dt} = D\left(C_{in,i}^L - C_i^L\right) + \sum_j^N (k_{i,j} r_j) + N_i^{L-G} \tag{13}$$

where $k_{i,j}$ is the yield coefficient of the component i referred to the reaction j, N is the total number of biological reactions and N_i^{L-G} is the volumetric transfer rate for the component i from the liquid to the gaseous phase.

Gaseous phase. The mass balances are written in Equation (14), one for each component present in the gaseous phase (CO_2, CH_4, H_2):

$$\frac{dn_i}{dt} = F_i^{in} - F_i^{out} + V_L N_i^{L-G} \tag{14}$$

where, n_i are the moles of component i in the gaseous phase, V_L is the liquid volume. The inlet and outlet molar flow rates are F_i^{in} and F_i^{out}, respectively. According to the control volume considered, only for hydrogen $F_i^{in} \neq 0$ when it is injected in the system during BM. The volumetric transfer rate N_i^{L-G} is defined as follows

$$N_i^{L-G} = k_{L,i} a \left(C_i^L - C_i^{L*}\right) \tag{15}$$

where $k_{L,i} a$ is the overall volumetric mass transfer coefficient referred to the component i in the liquid phase, C_i^L is the concentration of the gaseous component i dissolved in the liquid, and C_i^{L*} represents the hypothetic liquid concentration of component i in equilibrium with the gas bulk. The concentration C_i^{L*} is linked to the partial pressure of the related component P_i by means of the Henry's law

$$C_i^{L*} = P_i / H_i \tag{16}$$

where H_i is the Henry's constant referred to the component i. Assuming valid the penetration theory for the gas–liquid mass transfer, for each component the mass transfer coefficient is proportional to the square root of its diffusivity. The latter consideration permits to link all the mass transfer coefficients as described in Equation (17):

$$k_{L,i} a = k_{L,j} a \left(D_i / D_j\right)^{1/2} \tag{17}$$

where D_i is the diffusivity of the component i. The values of the diffusivities and of the Henry's constants for every component are reported in Table 2.

Table 2. Values of the Henry's constant and of the diffusivity referred to all the gaseous components.

Gas	Diffusivity Coefficient [1] (D_i) (cm^2 s^{-1}) (10^5)	Henry's Constant [1] $(1/H_i)$ (mol L^{-1} Pa^{-1})
H_2	4.65	7.40×10^{-9}
CH_4	1.57	1.12×10^{-9}
CO_2	1.98	2.70×10^{-9}

[1] From Pauss et al. [23].

2.5. Parameter Estimation

The model was developed in order to be suitable to both Anaerobic digestion and Bio-Methanation operation mode. For this reason, it was possible to split the parameter estimation in two steps:

1. Parameter Estimation related to the Anaerobic Digestion by evaluating the stationary state of the system. As will be explained in the result section, the unknown parameters to determine are: all the yield parameters related to the reactions r_1 and r_2, the mass transfer coefficients $k_{L,i}a$ and the parameter α.
2. Evaluation of the remaining parameters by a dynamic simulation fitting during Bio-methanation. It will be later explained how the only parameter to fit will be the hydrogen mass transfer coefficient $k_{L,H_2}a$.

2.5.1. Determination of the AD Stationary State

The equations that describe the stationary state of the reactor are crucial to build the algebraic system necessary for the parameter evaluation. The same strategy was applied by Bernard et al. [19].

Liquid phase. Starting from setting to zero the time derivative of Equation (16), it is possible to derive the values of the concentration of lactose (S_1) and VFA (S_2) at stationary state.

$$S_1^* = K_{S_1} - \frac{\alpha D}{(\mu_{1,max} - \alpha D)} \tag{18}$$

$$\frac{S_2^2}{K_I} + (1 - \mu_{1,max}\alpha D)S_2 + K_{S_2} = 0 \tag{19}$$

Equation (19) shows that there are two possible values of S_2^*. The difference between them depends on whether the system is working or not in a VFAs inhibited mode. Since in the system under analysis this situation did not occur, the minimum value of S_2 solving Equation (19) was taken in consideration as S_2^*. Once obtained S_1^* and S_2^*, from Equation (17), it is possible to obtain the expression for the biomass X_1 and X_1 concentration at the stationary state:

$$X_1^* = \frac{(S_{1,in} - S_1^*)}{\alpha k_1} \tag{20}$$

$$X_2^* = \frac{\frac{k_2}{k_1}(S_{1,in} - S_1^*) - S_2^*}{\alpha k_3} \tag{21}$$

In order to determine k_1, the knowledge of α and one between X_1^* or S_1^* is essential. For this reason, it was assumed (as suggested in the work of Bernard et al.) as a (rough) estimation of the total biomass concentration the total amount of Volatile Suspended Solids (VSS) in the digestate at stationary conditions. Moreover, the ratio between the acidogenic microbial population respect to the total one (γ) was taken from literature [24] and fixed to $\gamma = 0.2$. According to the above-mentioned considerations and to Equations (20) and (21) it follows:

$$\frac{X_1^*}{X_1^* + X_2^*} = \gamma = \frac{S_{1,in} - S_1^*}{\alpha k_1 \cdot VSS} \tag{22}$$

At this point, only the value of α must be evaluated to calculate k_1. For this purpose, it was exploited the definition of α previously given in Section 2.4.3. Considering the liquid volume filled with random packing ($V_1 \sim 30$ L) and the volume of the reactor occupied only by liquid ($V_2 \sim 20$ L), α is the solution of the system reported below:

$$\begin{cases} X_L V_2 = \alpha \cdot VSS \cdot V_L \\ X_{im}V_1 = (1 - \alpha) \cdot VSS \cdot V_L \end{cases} \tag{23}$$

where X_{im} and X_L are the concentration of biomass in the immobilized and liquid part, respectively. Once calculated k_1, from Equation (21), k_3 was obtained by fixing k_2 from

literature. In fact, from the sensitivity analysis developed in the work of Bernard et al. [19], k_2 was find out to have a minor influence on the system.

Gaseous phase. The information on the reactor behavior that is available by means of the experimental operations is biogas flowrate and composition. The utilization of such information in the stationary state Equations of the system is crucial for the evaluation of the parameters k_4, k_5, k_7. Those parameters do not depend on the values of $k_{L,CO_2}a$ and $k_{L,CH_4}a$ for two different reasons that will be verified in the result section:

- The concentration of CO_2 in the liquid phase was supposed to be at equilibrium conditions since it is a highly soluble gas [23]. Combining Equations (13) and (14) and setting to zero the time derivative terms, the following equation was obtained were k_4 was fixed from literature since it is low impact on the system sensitivity again considering the work of Bernard et al. [19].

$$k_5 = \frac{F_{CO_2}^{out}}{V_L D\alpha X_2^*} + \frac{P_{CO_2}}{H_{CO_2}\alpha X_2^*} - k_4\frac{X_1^*}{X_2^*} \tag{24}$$

- The concentration of CH_4 in the liquid solution happens to be so low that it was neglected the term that it takes it into account for the evaluation of the parameter k_6. The latter was calculated by combining Equations (13) and (14) and setting to zero the time derivative terms which led to the expression reported below.

$$k_6 = \frac{F_{CH_4}^{out}}{V_L D\alpha X_2^*} \tag{25}$$

2.5.2. Dynamic Fitting

All the yield parameters related to *Methanobacterium thermoautotrophicum* bacteria metabolism (reported in Table 3) were taken from the work of Schill et al. [16]. Since all the other yield parameters were calculated using the stationary state of the Anaerobic Digestion, the only parameter to evaluate remained $k_{L,H2}a$ in the case of 4 L/min and 6 L/min biogas flow recirculation. A non-linear regression, based on the minimization of the Mean Square Error, was performed with the aid of the process simulator gProms (Process System Enterprises, London, UK) to fit the experimental data.

Table 3. Yield parameters of the *Methanobacterium thermoautotrophicum* bacteria metabolism.

Parameter	Meaning	Unit	Value
k_8	Yield for *Methanobacterium thermoautotrophicum* growth	g_{VSS}/mol	0.443
k_9	Yield for CO_2 consumption	mol_{CO2}/mol_{H2}	0.166
k_{10}	Yield for CH_4 production	mol_{CH4}/mol_{H2}	0.179

3. Results and Discussion

3.1. Anaerobic Digestion Mode

During the Anaerobic Digestion Mode, the system reached stable conditions already after 10 days. The amount of biogas produced was of 30.8 ± 2.1 mL/min with a volumetric composition of $51.21\% \pm 1.04$ and $49.28\% \pm 2.36$ for CH_4 and CO_2, respectively. The value of VSS calculated from liquid samples of the digestate was of 6.3 ± 0.1 (g/L), and it corresponds to the value used for X_L. The concentration of biomass in the immobilized part of the reactor (X_{im}) was calculated to be 7.51 ± 0.3 (g/L). As explained in the previous section, those experimental data (reported in Table 4) were fundamental for the estimation of the yield parameters related to the acidogenesis and acetoclastic methanogenesis reactions.

Table 4. Values of the main parameters of the reactor during Anaerobic Digestion.

Parameter	Value $\pm$ SD
Biogas flow rate (mL/min)	30.8 ± 2.1
$CH_4\% \, v/v$	51.21 ± 1.04
$CO_2\% \, v/v$	49.28 ± 2.36
Biomass in digestate (X_L) (g_{VSS}/L)	6.3 ± 0.1
Biomass immobilized (X_{im}) (g_{VSS}/L)	7.51 ± 0.3

By means of the Equation (23), it was calculated $\alpha = 0.36$. Subsequently, from Equations (18) and (19) S_1^* and S_2^* were found to be 71 mg/L and 0.15 mmol/L respectively. According to the Equations (20)–(22) were evaluated the yield parameters k_1 and k_3, while from Equations (24) and (25), the yield parameters k_5 and k_6 were determined. The values of all the yield parameters related to the Anaerobic Digestion Mode are reported in Table 5.

Table 5. Value of the yield parameters referred to the Anaerobic Digestion mode of the reactor.

Parameter	Meaning	Unit	Value	Reference
k_1	Yield for lactose consumption	-	78.2	This work
k_2	Yield for VFA formation	mmol/g	116.5	[19]
k_3	Yield for VFA consumption	mmol/g	29	This work
k_4	Yield for CO_2 formation from acidogenesis	mmol/g	50.6	[19]
k_5	Yield for CO_2 formation from acetoclastic methanogenesis	mmol/g	208	This work
k_6	Yield for CH_4 formation from acetoclastic methanogenesis	mmol/g	275	This work

3.2. Bio-Methanation Mode

Before running the simulation, the initial conditions of the system should be provided. The gaseous phase initial conditions were fixed according to the biogas composition at the beginning of the experiment. Different considerations were necessary to select the appropriate initial conditions for the liquid phase. The initial substrate concentrations were set as S_1^* and S_2^* of the Anaerobic Digestion mode. The microbial population analysis, carried out by Lembo et al. [17] on the same reactor in previous experiments, showed that the 0.1% of the total biomass during AD mode was made of methanogens mainly belonging to the *methanobacteriaceae* family and the latter was mainly composed by hydrogenotrophic *Methanothermobacter thermoautrophicus*. For this reason, X_3 was estimated as 0.1% of the total VSS of the reactor liquid effluent during Anaerobic Digestion. Since the ratio between the acidogenic microbial population respect to the total one (γ) was kept to 0.2, the following initial conditions for the biomass were adopted:

- X_1 = 1.26 g/L
- X_2 = 5.03 g/L
- X_3 = 6.3 mg/L

The results of model fitting are reported in Figure 3, and the values of the hydrogen mass transfer coefficient, derived from fitting, are reported in Table 6. As expected, enhancing the recirculation flowrate, the mass transfer coefficient increases. In particular, modifying the recirculation rate from 4 to 6 L/min, the $K_{L,H_2}a$ changes from 100 to 220 d^{-1}. This phenomenon is explained by considering the reduction of the gas–liquid film resistance due to the induction of higher turbulence conditions.
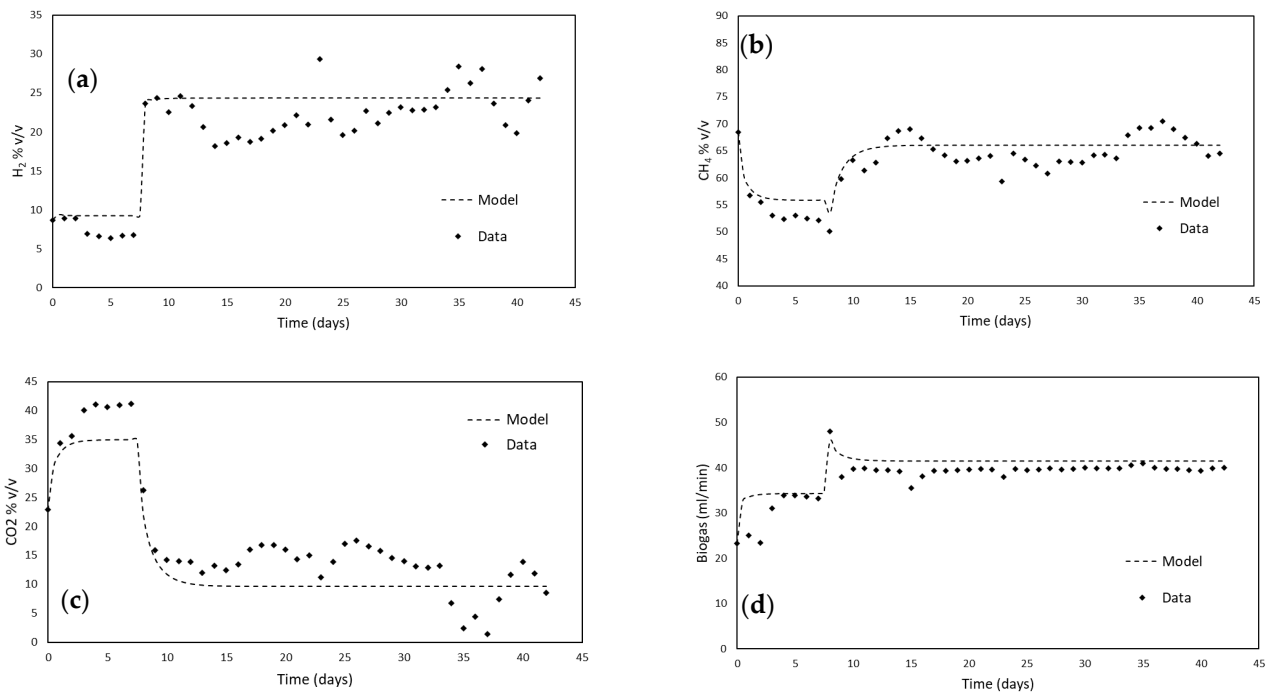
Figure 3. Experimental data fitting related to the composition of (**a**) hydrogen, (**b**) methane and (**c**) carbon dioxide and to (**d**) the outlet biogas flowrate. At day 7 the inlet flowrate of hydrogen in the system was switched from 10 mL/min to 50 mL/min, and the recirculation biogas flowrate was switched from 4 L/min to 6 L/min.

Table 6. Fitted value of the hydrogen mass transfer coefficient depending on the different recirculation flowrate.

$K_{L,H_2}a$ (d^{-1})	Recirculation Flowrate (L/min)
100	4
220	6

As it is possible to notice in Figure 3, the model well represents the dynamic of the system even when a step change in the inlet hydrogen stream is applied. The experimental data fluctuation observed in Figure 3 related to the biogas composition may be caused by the variation of lactose content in the Second Chees Whey (SCW), which was not considered in the model. In order to obtain further information about the system behavior, the dimensionless number of Stanton and β were calculated as follows:

$$St = \frac{\tau_p}{\tau_t} = \frac{K_{L,H_2}aV_L}{Q_R} \tag{26}$$

$$\beta = \frac{\tau_t}{\tau_r} = \frac{q_{max}X_3(t)}{K_{L,H_2}a} \tag{27}$$

where Q_R is volumetric recirculation flowrate, τ_r is the characteristic time of reaction, τ_p is the characteristic residence time of the gas into the reactor, and τ_t is the characteristic time of material transfer from the gaseous to the liquid phase [15]. Both *St* and *β* depend on time. According to this, the model was used to calculate *St* and *β* during time as reported in Figure 4.
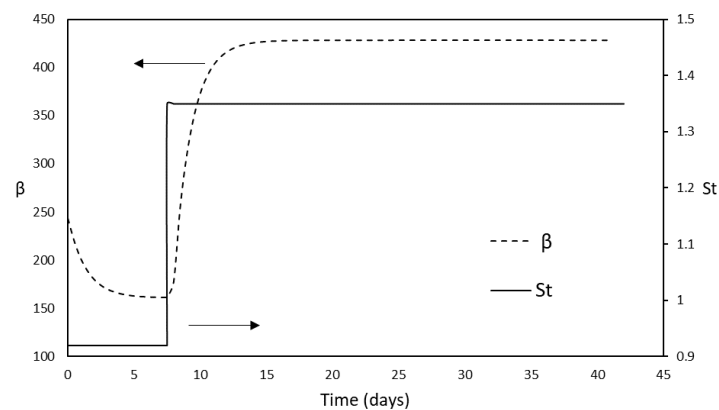
Figure 4. Plot of the dimensionless numbers St (-) and β (–) versus time during the Bio-Methanation working period.

As shown in Figure 4, β >> 1 for the working period. This condition indicates that the consumption of hydrogen in the liquid phase is much higher than its supply. On the other hand, St ≈ 1, which means that the residence time of hydrogen in the reactor is comparable with the characteristic time for its transfer to the liquid phase. More precisely, the value of St changes from 0.92 to 1.35 with the recirculation rate of 4 L/min and 6 L/min, respectively. According to the consideration made on β, a possible strategy to increase the reactor performances is to enhance the liquid to gas mass transfer coefficient in order to exploit the capacity of biomass in converting H_2 and CO_2 into CH_4. It was observed that the recirculation flowrate plays a fundamental role for the regulation of $K_{L,H_2}a$, but the values of St are still too low to obtain a high conversion of the hydrogen injected in the reactor. Since St grows once increased the recirculation flowrate, it is possible to conclude that the benefits obtained by enhancing the recirculation flowrate (increase of $K_{L,H_2}a$) are higher than the drawbacks (decrease of the residence time of the gas in the reactor). However, a significant increase of St could be obtained only if $K_{L,H_2}a$ is adjusted by modifying other design parameters rather than the recirculation rate. In order to clarify this aspect, further studies are necessary to investigate deeper the dependence of $K_{L,H_2}a$ with the biogas recirculation flow. In addition, the mass transfer coefficient can be enhanced with the utilization of a gas sparger which may reduce the dimensions of the flowing bubbles. Another important factor is the influence of biomass immobilization on the system performances. In this work such aspect was taken into account by the introduction of the parameter α. The yield parameters of the Anaerobic Digestion are highly sensible to the value attributed to α. Bearing this in mind, it is possible to affirm that the model it is able to reproduce the behavior of the system, but further experiments should be carried out to really understand the role of bacteria immobilization, and its effect on the yield parameters of the model.

4. Conclusions

In this work, the model proposed was able to reproduce the experimental data of a 49 L randomly packed Gas Stirred Tank Reactor (GSTR). The parameter estimation was performed in two steps applied in the following order: (1) calculation of the yield parameters for the acidogenic and acetoclastic microorganisms using the stationary state of the reactor during anaerobic digestion and (2) evaluation of the hydrogen mass transfer coefficient by means of a dynamical fitting. It was found out that the hydrogen mass transfer coefficient strictly depends upon the recirculation flowrate. Moreover, by defining the dimensionless numbers of St and β, it was understood that the system could be capable of treating a higher amount of hydrogen as long as the hydrogen mass transfer coefficient is increased sufficiently. Further investigations are necessary to better understand how the recirculation flowrate affects the hydrodynamics of the reactor and to quantify the influence

that the introduction of a gas sparger may induce to the hydrogen mass transfer. In addition, also the benefits given by the presence of random packing should be deeply evaluated.

Abbreviations

AD	Anaerobic Digestion
BM	Bio-Methanation
CSTR	Continuous Stirred Tank Reactor
GSTR	Gas Stirred Tank Reactor
HRT	Hydraulic Retention Time
SCW	Second Cheese Whey
VFAs	Volatile Fatty Acids
VSS	Volatile Suspended Solids
Symbols	
D	Dilution (1/d)
D_i	Diffusivity of component i (cm^2/s)
H_i	Henry's constant of component i (mol/L Pa)
K_{H2}	Half saturation constant associated with H_2 (μmol/L)
$K_{L,H2}$	Hydrogen mass transfer coefficient (1/d)
K_{S1}	Half saturation constant associated with S_1 (g/L)
K_{S2}	Half saturation constant associated with S_2 (mmol/L)
K_I	Inhibition constant associated with S_2 (mmol/L)
k_1	Yield for lactose consumption
k_2	Yield for VFA formation (mmol/g)
k_3	Yield for VFA consumption (mmol/g)
k_4	Yield for CO_2 formation from acidogenesis (mmol/g)
k_5	Yield for CO_2 formation from acetoclastic methanogenesis (mmol/g)
k_6	Yield for CH_4 formation from acetoclastic methanogenesis (mmol/g)
k_8	Yield for Methanobacterium thermoautotrophicum growth (g_{VSS}/mol)
k_9	Yield for CO_2 consumption from idrogenotrophic methanogenesis
k_{10}	Yield for CH_4 production idrogenotrophic methanogenesis
m	Maintenance coefficient (mol/g d)
q_{max}	Maximum hydrogen specific production rate (mol/g d)
St	Stanton dimensionless number
S_1	Organic substrate consumed by the biomass X_1 (g/L)
S_2	Volatile Fatty Acids (mmol/L)
V_L	Reactor liquid volume (L)
X_1	Acidogenic bacteria (g/L)
X_2	Acetoclastic methanogen archaea (g/L)
X_3	Methanobacterium thermoautotrophicum biomass (g/L)
Greek symbols	
α	Fraction of the total non-immobilized biomass
β	Dimensionless number
γ	Biomass fration of the acidogenic microbial population
$\mu_{1,max}$	Maximum acidogenic biomass growth rate (1/d)
$\mu_{2,max}$	Maximum acetoclastic methanogenic biomass growth rate (1/d)

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