Institute for Water Quality
and Resource Management

Master thesis

Organic acid degradation inhibition by high concentrations of hydrogen in an in-situ anaerobic methanation reactor

Submitted by

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Abbreviations

R1, R2	Reactor 1 (with no hydrogen), reactor 2 (with hydrogen)
TS	Dry mass concentration [g/L or g/kg sludge]
oTS	Organic dry mass concentration [g/L or g/kg sludge]
COD	Chemical Oxygen Demand [mg O_2]
TP	Total Phosphate concentration [mg/L]
TKN	Total Kjeldahl Nitrogen
NH_4-N	Ammonium nitrogen concentration [mg/L]
PO_4-P	Ortho Phosphate Phosphor concentration [mg/L]
CH_4	Methane
CO_2	Carbon dioxide
O_2	Oxygen
H_2S	Hydrogen Sulfide
HRT	Hydraulic Retention Time [d]
ORP	Oxidation Reduction Potential
BMR	Biologic Methanation Rate

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Abstract

Biological in-situ methanation is a promising method to produce highly pure methane out of organic residuals and hydrogen from surplus energy. Two lab scale digesters were operated continuously to demonstrate biological in-situ methanation (BM) using raw sludge from a municipal wastewater treatment plant as substrate. The degradation of organic acids and organic macro molecules during the conventional anaerobic digestion process, provides the necessary CO_2 for the methanation reaction. Additional hydrogen for the methanation reaction is provided by water electrolysis and is introduced into the reactor via a fine porous silicon diffuser. However, it was proven that degradation of propionic acid needs very low concentrations of hydrogen to be performed ($<10^{-4}$ atm). Otherwise the reaction is inhibited through thermodynamic reasons. (Fukuzaki et al., 1990; Siriwongrungson et al., 2007). Fang and Xiao-Shan (1998) showed that the presence of H_2 (between 10^{-5} and 10^{-4} atm) does not affect the butyrate degradation, but enhances the formation of propionate (regardless of the initial butyrate's concentration). But it was also demonstrated that the addition of high quantity of H_2 (1 atm) in a batch fed with manure inhibits the propionic acid and butyric acid degradation only under high mixing intensity while more than 90% of H_2 is converted to methane.

The aim of this study is to examine the kinetic behavior of propionic acid, acetic acid and butyric acid degradation during continues operated in-situ methanation under high concentrations of hydrogen. Degradation rates and gas production rates are presented for each acid. Inhibition factors for the degradation of acetic acid, propionic acid, and butyric acid are presented.

Keywords: biological methanation, degradation inhibition, hydrogen, kinetic, organic acids

1. Introduction

1.1. Energetic transition

Energetic transition is supposed to be one of the major challenges of the incoming decades. Since the end of the 19th century the average temperature on the globe has risen between 0.8 °C and 1.2 °C (IPCC, 2014). Most of this increase is caused due to the emission of CO_2 produced by the industry, agriculture, the production of electricity, transportation and mobility, etc. In order to limit the temperature increase, several agreements were ratified by many countries like the Kyoto protocol (1997) or the “Accord de Paris” (2015) with the goal to reduce the global emission of greenhouse gasses. To reach the goals of the Kyoto protocol and the Paris agreement Austria has strongly developed the sectors of photovoltaic and wind-power- in the last decade. In 2018 the accumulated produced power of photovoltaic was 1,438 MWh (PVAustria, 2018) while that of wind-power was 6,030 MWh (IG Windkraft, 2018) which represents 2.1% respectively 8.6% of the electric production of Austria (IEA, 2018). The Austrian energy mix for the production of electric energy is presented in figure 1.

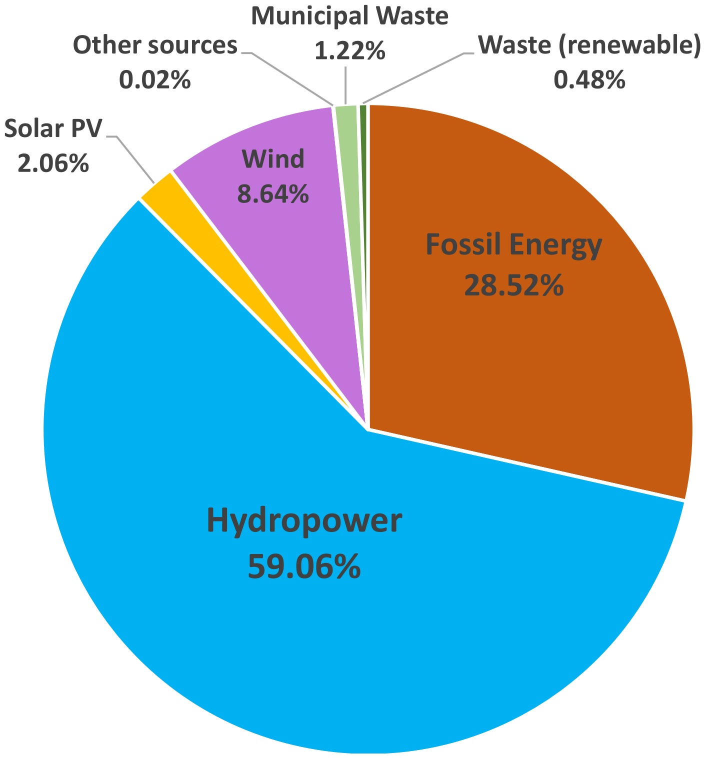

Figure 1 Austrian electricity mix for the year 2018 (IEA, 2018)

However, the issue of the load balancing of the exceeding produced energy is a strong concern and it increases with growing fluctuating energy share. Contrarily to thermic power plants or nuclear power plants, the energy production of wind and PV can’t be adjusted to the demand and depends of the weather. For instance, solar panels don’t produce electricity during the night and their performance is much higher in summer than in winter although the electrical demand is much higher in winter than in summer. That’s why technologies of energy storage are necessary. The concept of Power-to-Gas is an answer to this issue. The exceeded electricity is used to perform a hydrolysis of water which produces oxygen and hydrogen (equation 1).

$$H_2O \quad \overset{\overset{\text{Electricity}}{\Downarrow}}{\rightarrow} \quad 0{,}5\ O_2 + H_2 \qquad \text{(kJ/Mol)} \qquad (1)$$

The produced oxygen can be used in the biological wastewater treatment for aeration or for ozone production to remove organic trace substances from the wastewater (Schäfer et al., 2020). The produced hydrogen can be used for industrial uses or for methanation of CO_2.

1.2. Biological methanation

For energy storage via Power-to-Gas there exist two possible pathways. The first is to inject the hydrogen produced via electrolysis directly into the natural gas distribution network. But the rate of hydrogen in the natural gas is strongly regulated in most of the counties worldwide. In Austria the legal limit is 4 mol % hydrogen of the natural gas. The second possibility is to use hydrogen to produce methane through methanation according to the following equation 2.

$$4H_2 + CO_2 \quad \rightarrow \quad CH_4 + 2H_2O \qquad (2)$$

The obtained methane can be refined to obtain synthetic natural gas (SNG) that can be used directly in the distribution network. The produced biomethane can be stored and transported in the natural gas grid. This methane gas can be used for heating purpose in habitation or industries, it can be used to produce electricity in a gas power plant or as liquid natural gas (LNG) for fuel purposes. Figure 2 shows the Power-to-Gas concept with sector coupling between electric and gas grid.

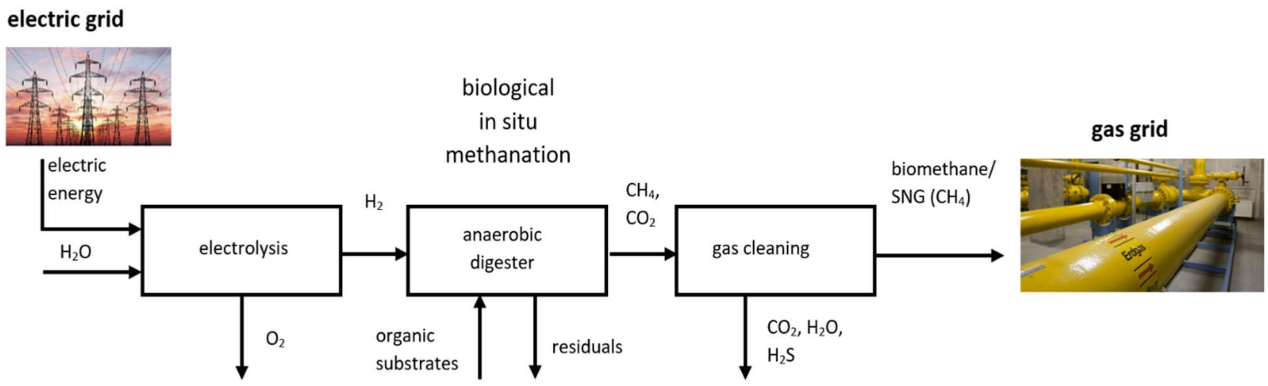

Figure 2 Power-to-gas Concept for In-situ Methanation (adapted after Tauber et al., 2020)

There are several possible processes for methanation. A currently well-established process is the Sabatier process, a thermo-chemical process which is based on the catalytic hydrogenation of CO_2 to methane. It operates at high temperatures of 250-400 °C and under pressures of 1-80 bar. It uses nickel- and ruthenium-based catalysts for enabling the reaction. Another possibility is the biologic methanation using hydrogenotrophic archaea for the conversion of carbon dioxide and hydrogen to methane. Which is declined in two versions: ex-situ and in-situ. In the ex-situ version, the methanation is realized in a separate reactor of the bioreactor ("outside"). The bioreactor provides a biogas, a mix of CO_2 and methane which is injected in the methanation reactor. Hydrogen is also injected in the reactor to allow the biological methanation. With this process it is possible to obtain a biogas with methane concentrations of 98% (Bassani et al., 2017). In the in-situ version the methanation is directly realized in the bio-reactor – the so called digestor, where the fermentation reactions of the organic substrate take place. Additional hydrogen is introduced as well to perform the methanation reaction. Biogas with a methane concentration of 82% can be achieved (Bassani et al, 2016). The advantage of the ex-situ process over the in-situ process is that it's much easier to maintain the stability of the system in an ex-situ process. And for the ex-situ process, much smaller reactors are required. But only one reactor is required for the in-situ process in which the anaerobic digestion and the biologic methanation are realized in parallel.

The aim of the project BioMAra in which this master's thesis takes place, is to investigate conditions under which a biologic methanation in digesters of municipal wastewater treatment plants can occur. Additionally, the economic viability of such a project is evaluated. The methanation in municipal digesters is an in-situ process using existing bioreactors under mesophilic anaerobic conditions. In this study two lab scale digesters were operated continuously for more than two years to demonstrate biological in-situ methanation (BM) using raw sludge from a municipal wastewater treatment plant as substrate. One reactor was operated without hydrogen injection (reactor 1) and served as a reference. The second reactor was operated as an in-situ methanation reactor with additional hydrogen injection (reactor 2). In the BM reactor the anaerobic digestion process provides the necessary CO_2 for the methanation reaction from the degradation of organic acids and organic macro molecules in the substrate (raw sludge). Additional hydrogen for the BM is provided by water electrolysis and is introduced into the reactor via a silicon diffuser. In Table 1 collected data of performance of methane production measured in the project BioMAra are compared to reference data from different authors.

Table 1 Obtained production rate of CH_4 from several references and for five combinations of hydraulic retention time and H_2-injection in the project BioMAra (adapted after Tauber et al., 2020)

Reference	BM-Rate [L_{CH4} /(L·d)]	CH_4-content [%]	Reactor-form	Temp. [°C]	Pure Culture
Luo et al. (2012)	0,08	65,0	CSTR	55	Yes
Bassani et al. (2016)	0,18	81,3	UASB	55	Yes
Luo and Angelidaki (2013a)	0,34	68,0	CSTR	55	No
	0,39	75,0	CSTR	55	No
Luo and Angelidaki (2013b)	0,38	96,1	CSTR	55	No
(Strubing *et al.* 2017)	15,4	98,5	Trickle-bed	55	No
Schönberg and Busch (2012)	0,52	81,3	Column	55	No
	0,60	85,8	Column	60	No
BioMAra 25d HRT Keramik Diffusor	0,18	53,8 – 69,4	CSTR	38	No
BioMAra 25d HRT Diffusor 50 Poren	0,21 – (0,68)	56,92 – 73,42	CSTR	38	No
BioMAra 25d HRT Diffusor 100Poren	0,39	57,17 – 73,75	CSTR	38	No
BioMAra 20d HRT Diffusor 100Poren	0,17	57,32 – 73,94	CSTR	38	No
BioMAra 15d HRT Diffusor 100Poren	0,43	53,91 - 69,6	CSTR	38	No

1.3. Anaerobic digestion

Anaerobic digestion is a technology to treat sewage sludge from biological wastewater treatment and organic residuals which allows to produce biogas. This technology uses microorganism in the sludge which degrade the organic matter in the sludge and convert it into biogas, a mixture mainly composed of methane, carbon dioxide, with traces of hydrogen and hydrogen sulfide (H_2S). This biogas is then collected, cleaned and can be used to produce electricity or heat. Figure 3 illustrate the main steps of anaerobic digestion in a waste water treatment plant.

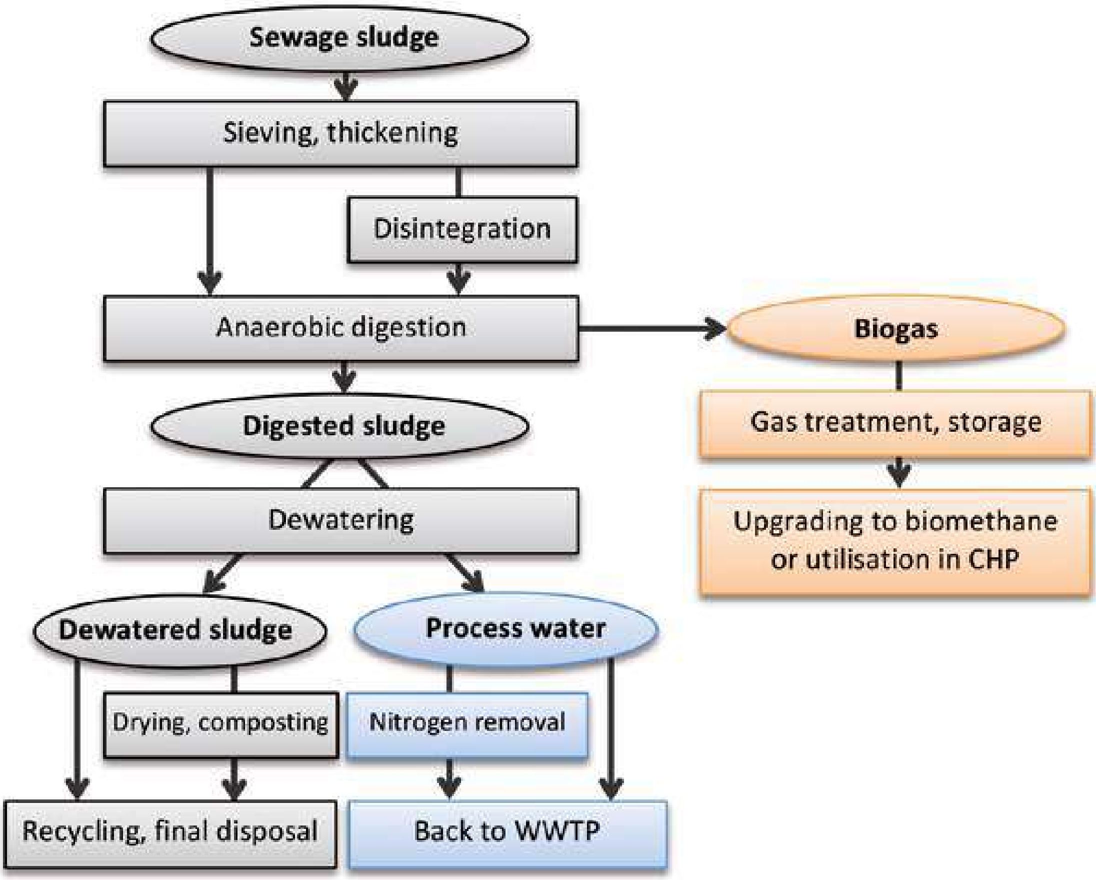

Figure 3 Main steps of anaerobic digestion at waste water treatment plants (Bachmann, 2015)

Figure 4 Two digesters at the waste water treatment plant of Bruck an der Leitha, Austria

Gujer and Zehnder (1983) identified six different conversion processes of degradation of organic material in a continuous flow reactor at steady state that they called a “mature digester”, which is similar to the BM reactors used in this project.The first process is the hydrolysis of biopolymers such as proteins, carbohydrates and lipids into monomers and fatty acids. The second process is the fermentation of amino acids and sugars into intermediary products (like propionic acid or butyric acid), acetic acid and hydrogen. The third process is the anaerobic oxidation of long chain fatty acids and alcohols into intermediary products like acetic acid and hydrogen. The fourth process is the anaerobic oxidation of intermediary products like organic acids (propionic acid, butyric acid) into acetic acid, hydrogen and CO_2. The following stoichiometry for the propionic acid’s degradation, presented in equation 3 was suggested by Gujer and Zehnder (1983):

$$\mathrm{CH_3CH_2COO^-} + 3\,\mathrm{H_2O} \rightarrow \mathrm{CH_3COO^-} + \mathrm{HCO_3^-} + \mathrm{H^+} + 3\,\mathrm{H_2} \qquad (3)$$

The fifth process is the methanogenesis from acetic acid by the acetogen bacteria. According to Gujer and Zehnder, the decarboxylation of acetic acid generates 70% of the produced methane. The stoichiometry of this reaction is given by the following equation 4:

$$C^{*}\mathrm{H_3COO^-} + \mathrm{H_2O} \rightarrow C^{*}\mathrm{H_4} + \mathrm{HCO_3^-} \qquad (4)$$

The last identified process is the methanogenesis from hydrogen by the methanogen bacteria. This is the reaction of methanation described in equation (2). In addition, to these six process of degradation Gujer and Zehnder also identified factors that inhibits degradation of organic acids and the methanogenesis from hydrogen. The degradation process of propionic acid is thermodynamically inhibited by high concentration of hydrogen. While the methanation of hydrogen is extremely sensitive to the pH-value in the digester. The optimal pH-domain of growth of the methanogen archaea was identified between 6.8 and 7.8 (Kroiss, 1988). Outside of this domain the growth rate of the archaea dramatically sinks with the pH-value rising or sinking. Gujer and Zehnder (1983) noted that a first reason of degradation of the pH-value is the accumulation of acetic acid which decreased the pH-value and thus decreases the efficiency of the methanation of hydrogen. This leads to an inhibition of oxidation of propionic acid, which starts to accumulate and thus decreases successively the pH-value. Those mechanism of inhibition make difficult to maintain the stability of the reactor since it can easily switch to the acid domain of pH and thus inhibits the methanation. Also, hydrogen inhibits the degradation of organic acids above a certain concentration. In this study, it represents a difficulty since we inject in our reactor hydrogen which boost the methanation but a priori inhibits the propionic acid degradation which then leads to inhibition of methanation. Moreover, this problem of pH stability can also be taken in the other way. If the process of degradation of organic acids and of methanation from hydrogen are “too efficient” the amount of acids in reactor will strongly decrease, leading to pH rise und thus inhibiting methanation. The following scheme summarize the processes that occur in a digester.

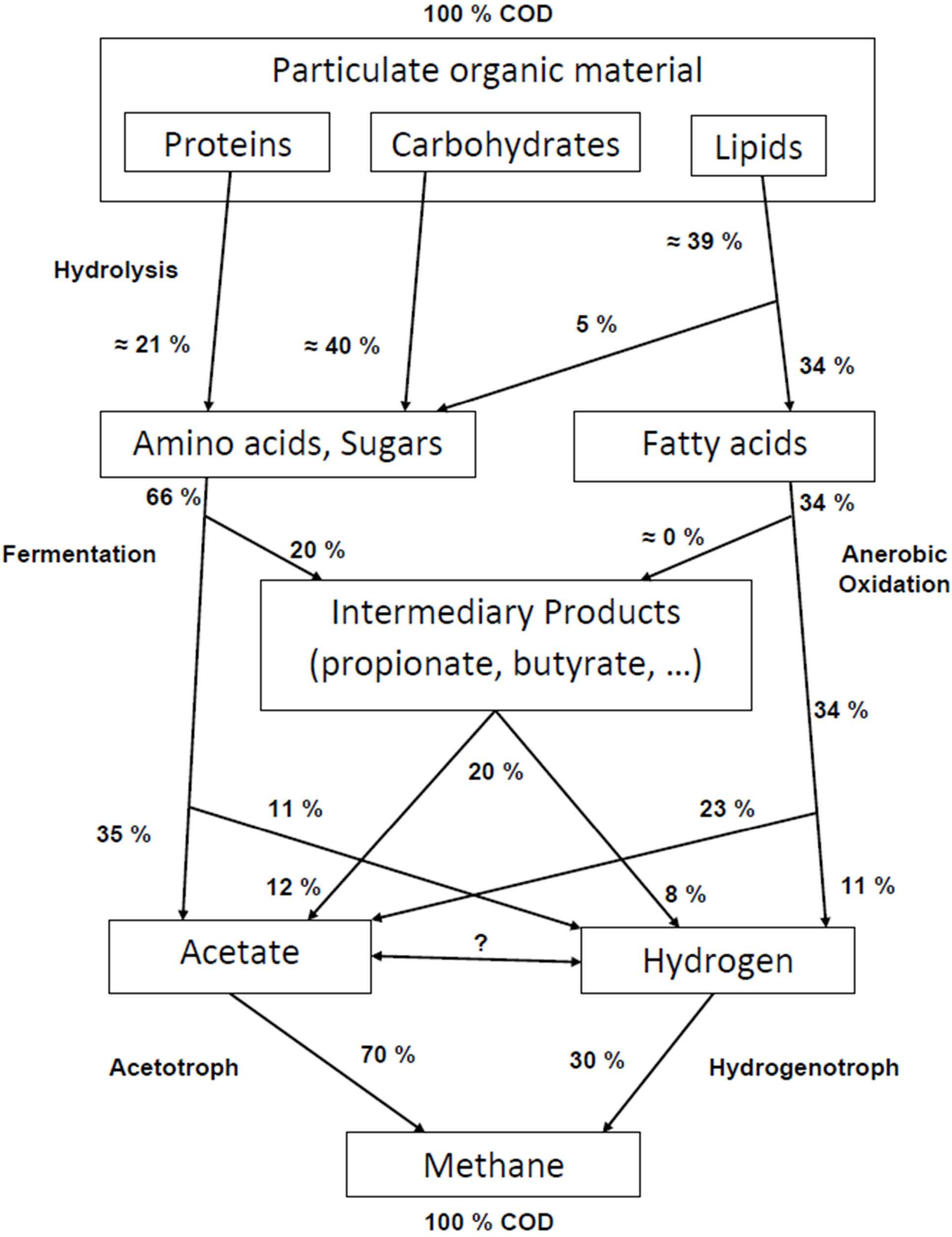

Figure 5 Reaction scheme for the anaerobic digestion of domestic sludge adapted after Gujer and Zehnder (1983). Percentages indicate substrate flow (stoichiometrically) in the form of COD or CH_4 equivalents. Only the net flow of substrates (degradation minus biomass formed) through cell external pools is indicated.

Several other authors have noticed these inhibition's mechanism. Fukuzaki et al. (1990) noted that a high initial concentration of propionic acid (more than 10 mM) in a digester filled with a mesophilic propionic acid-acclimatized sludge strongly decreases the propionic acid disappearance's rate. Moreover, a pH-value below pH 7 also decreases this rate (and the lower the stronger). They supposed that this inhibition is caused by undissociated propionic acid. The same behavior was observed by addition of acetic acid (2,5 mM). An addition of 0,91 atm of H_2 in mesophilic sludges resulted in inhibition of propionic acid degradation which was later reverted by addition of methanogen archaea. Fang and Xiao-Shan (1998) reported that the presence of H_2 (between 10^{-5} and 10^{-4} atm) in batch of anaerobic butyric acid-utilizing sludge doesn't affect the butyric acid degradation, but enhances the formation of propionic acid (regardless of the initial butyric acid's concentration). Luo et al. (2011) showed that in a batch fed with manure, the addition of high quantity of H_2 (1 atm) inhibits propionic acid and butyric acid degradation only under high mixing intensity (shaking speed of 300 rpm) while more than 90% of H_2 is converted to methane. Furthermore, they showed that continuous addition of H_2 (flow rate of 28,6 mL/L · h) to an anaerobic reactor fed with manure doesn't affect the propionic acid and butyric acid level. In another study, Luo et al. (year) conducted an experiment on an anaerobic reactor for in situ biogas upgrading by using hollow fiber membranes to add H_2 to the process by diffusion. They observed that increasing the flow of H_2 increases both the methanogenic activity and the pH-value. However, a decrease of the methanogenic activity for substrate with acetic acid was detected for a flowrate of H_2 of 1,760 mL/(L · day). It was interpreted as the result of inhibition of acetic acid degradation by high pH-value (8.31).

During the project BioMAra, the concentrations of organic acids contained in the raw sludge and in the digester sludge were quantified. The detected organic acids were acetic acid, lactate, butyric acid, propionic acid valeric acid and pentanoate. It appeared that their quantity and their composition varied during the project in a significative way. Their composition in the digester sludge of reactor 2 and in the raw sludge is described in the following diagrams (Figure 6 and Figure 7).

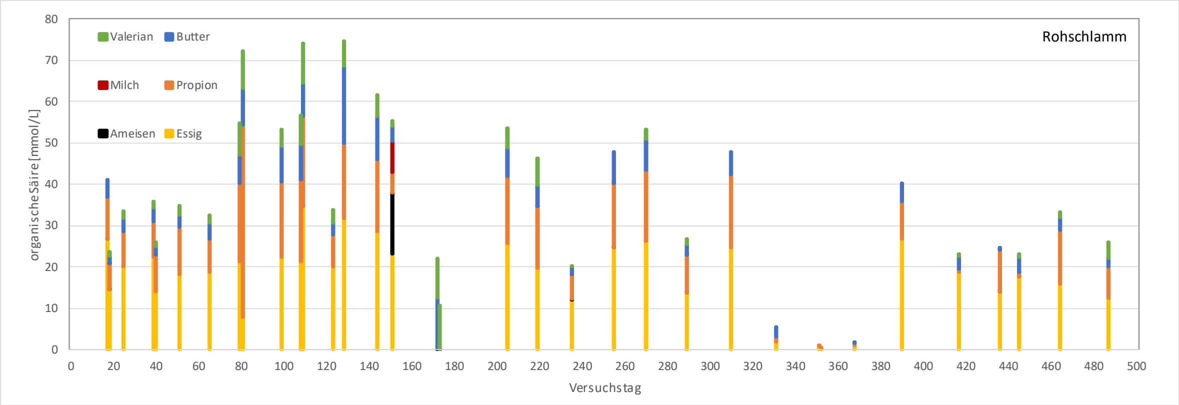

Figure 6 Concentration of organic acids in raw sludge

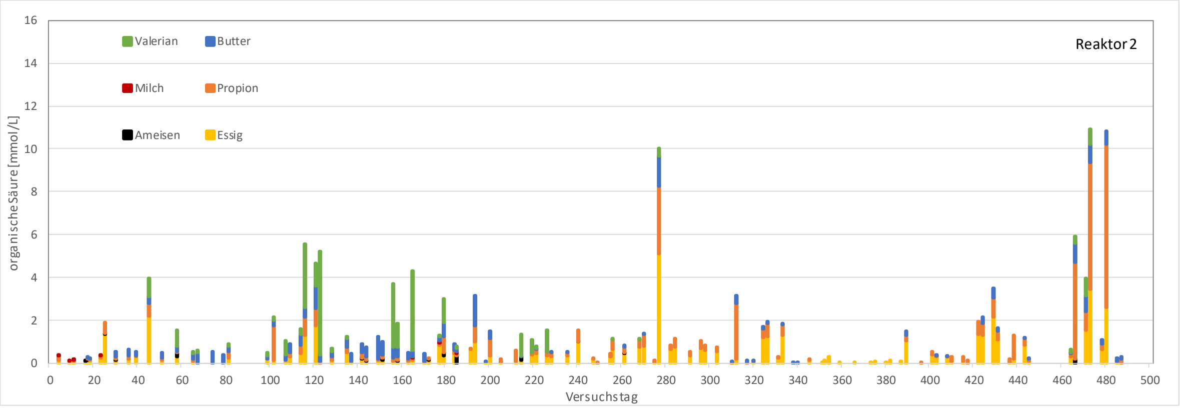

Figure 7 Concentration of organic acids in reactor 2

Periods with high amounts of organic acids were associated with inhibition of the gas production. As the composition of the organic acids in the raw sludge strongly vary and is not configurable, it is necessary to investigate how much high quantity of organic acid can impact on the gas production. And as these organic acids react differently one from another, it has to be done for each of them.

The aim of this thesis is to examine the kinetic behaviour of degradation of acetic acid, propionic acidic acid and butyric acidic acid during in-situ methanation under high concentrations of hydrogen. It is chosen to focus on these three acids, because they represent quantitatively the majority of organic acids during anaerobic digestion. There is currently no study about the effect of organic acids on the gas production of a digester with presence of hydrogen. That’s why this study is undertaken with the hope to bring clarification about those phenomena.

2. Methods and materials

2.1. Experimental facility

2.1.1. Description of the experimental facility

Figure 8 (left) shows the experimental facility with the two reactors in the digestor and for each one a drum gas counter, a system controller (Siemens LOGO 8.2), a gas washer and a PEM water electrolyzer (company, design). In Figure 8 (right) the two reactors with their connection tubes, their probes, their mixers and their heaters are presented. On the top of the reactors are connection tubes which collect the product gas in addition to allow the continuous measurement of this production. A schematic representation of the facility is represented in Figure 9.In this scheme the measure-loop for the gas measurement, the gas-dryer, the gas-filter, the gas-pump and the IR-spectrometer (Saxon Junkalor IR 5000 und IR 7000) for the continuous measurement of CO_2- CH_4 and O_2 concentration are presented. After the concentration measurement the gas is re-humidified with a washing bottle and re-introduced in the reactors. This humidification is necessary because of the drying-step which eliminates the water vapor in the gas, and the introduction of dry gas in the column would result in a diminution of the water content in the digester sludge. Both reactors have a column-volume of 14 L and a gas-space of 1 L (intern diameter: 12,5 cm, height: 120 cm).

A mixer (with three mixing-plates) is placed in the center of the reactor (figure 7). The produced gas is evacuated through the top of the reactor. The volume of the produced gas is measured by a drum gas counter (Ritter, TG 05) and stored in a gas-cylinder for the evaluation of its composition. A peristaltic pump allows the recirculation of the produced gas in the reactor (Ismatec ecoline, easyload II 77202-200). The pressure in the gaseous phase is measured once time a week with a hand-held meter (testo 512 Digital-Manometer). The combined pH/Redox-probes are installed horizontally in the upper part of the column (Endress + Hauser pH-/Redox-Sensor Memosens CPS16D) (figure 6, right). A valve is installed in the lower part (resp. upper part) of the column to allow the feed (resp. the evacuation) of the raw sludge (resp. column sludge) in the reactor. A mixing of the reactor's content by recirculation of sludge is possible via those valves with the use of a peristaltic pump (Watson Marlow, 323 D). However, it was never used but is necessary if a hollow-fiber membrane is used in the reactor. A heating mantel is used to assure a temperature of 38°C inside the column. The heating is done by an electric resistance (Arnold Rak HK-5,0 Heizkabel 230 V 75 W 5.0 m). The temperature is controlled by lighting up or lighting down the mantel via the control panel. On the figure 6 (left) the two H_2S-washing-bottle mounted in series are easily recognizable (in blue).

Figure 8 Experimental installation with the hydrogen generator and the gas collector (picture on the left). Reactors with connection tubes, probes, heating mantel and stirring drive (picture on the right).

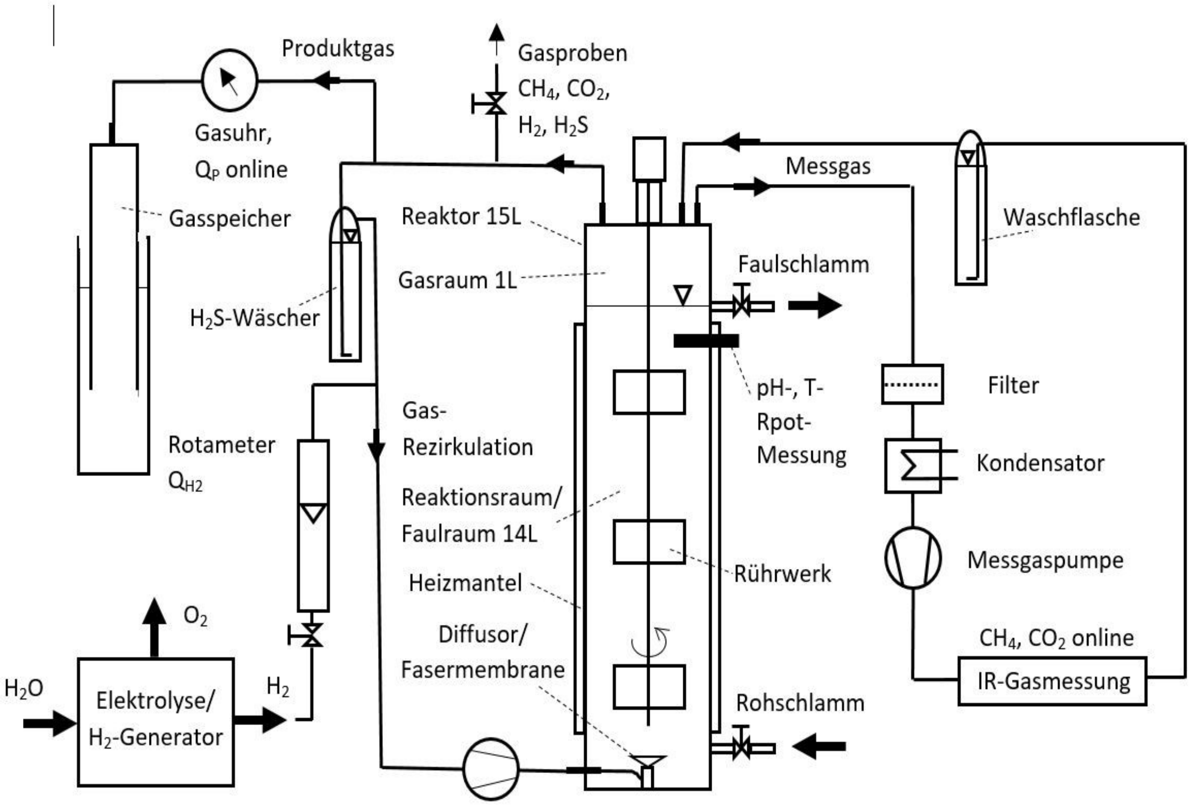

Figure 9 Schematic representation of the experimental installation of the BioMAra project.

2.1.2. Hydrogen generator

The hydrogen generator used in this experiment is based on the PEM-electrolysis. This device uses a Proton-Exchange-Membrane (PEM) as electrolyte and is directly connected to the electrode. The temperature of the process is generally limited to 80°C and hydrogen can be generated until 100 bar while at the same time oxygen is generated at atmospheric pressure. The following picture illustrate the PEM-principle and shows the device used the project (Parker, 20H). This water electrolyser delivers a maximum hydrogen-flow of 160 mL/min with a purity of H_2 of 99.9995%. The hydrogen is introduced between 2 and 7 bar.

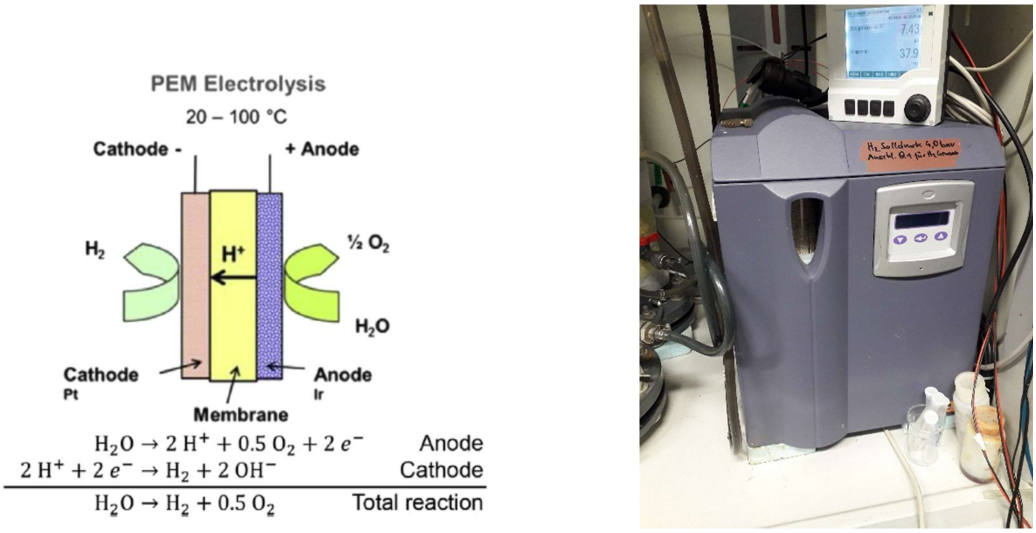

Figure 10 Principle scheme (left) and photo (right) of the proton exchange membrane (PEM) electrolysis

2.1.3. Centrifuge

During the experiments two centrifuges were used to separate the solid part of the sample from the liquid part in order to prepare it for the HPLC analysis. The centrifuge on the left of Figure 11 (SIGMA 3K30) was used with a speed of 14000 rpm, during 10 minutes and with a temperature of 20 °C. The one on the right side was used with a speed of 4200 rpm and during 15 minutes (temperature could not be programmed on this model). The centrifuges were used according to their availability.

Figure 11 Lab centrifuges used for sample preparation

2.1.4. Organic acids

The organic acids used in this experimentation were acetic acid, propionic acid and butyric acid. The acetic acid was under the form of a commercial colored and aqueous solution of vinegar with 5 % of acetic acid and an amount of protein smaller than 0,5 g per 100 mL. Propionic acid and butyric acid were used as pure liquid.

2.2. Experimental operation

2.2.1. Feeding with organic substrate

The sludge used in the experiment come from a local municipal waste water treatment plant. The sludge used during the whole experiment is a mixture from primary and secondary sludge from the biological wastewater treatment. the sludge is collected once every two or three weeks and stored in a fridge at 5°C until it is used. Before the utilization, the sludge is sieved with a hole sieve with hole diameters of 4 mm to eliminate big impurities which could plug or damage the tubes or mixers.

2.2.2. Hydraulic Retention Time

It is possible to calculate the hydraulic retention time (HRT) through the measure of the average daily sludge input in the reactors. The formula for calculation of HRT is given by the equation (5). HRT is an important parameter for the degree of sludge stabilization. Organisms inside the sludge need indeed enough time to develop, this is the minimal necessary HRT. It depends from several factors like temperature or biodegradability of the substrate. For a municipal waste water treatment plant this minimal HRT is around 25 days while the performed dosing experiments de HRT was set to 25 days. In this study the reactors were fed once a day, five times a week (from Monday to Friday). The sludge input per week is considered in the calculation of the HRT. A reactor volume of 14 liters and a feed of 600 mL of raw sludge two times a week gives an HRT of 50 days.

$$\mathrm{HRT} = \frac{\mathrm{V_R}}{\mathrm{Q_{RS}}} \tag{5}$$

HRT	Hydraulic Retention Time	$[\mathrm{d}]$
$\mathrm{V_R}$	Reactor’s volume	$[\mathrm{m}^3]$
$\mathrm{Q_{RS}}$	average daily volume flow	$[\mathrm{m}^3/\mathrm{d}]$

The feed is performed after reading of gas production, of the pH-value and of temperatures. A syringe of 100 mL is used to inject the raw sludge inside the reactors. The opening of a valve on top of the column allows the evacuation of sludge in the column through a drain hose. Every charge of injected raw sludge is weighed before injection. After injection the empty and uncleaned beaker is weighed to determine the exact mass of sludge injected in the reactor (so the loss of sludge during the injection are taken in count). The handling of this experimental device includes the feed of reactors, the verification of gasometers, the sampling of raw and digester sludge and the calibration of pH-, temperature- and Redox-probes.

2.2.3. Temperature control

A heating jacket with a power of 75 W was used to allow the control of temperature by switching on/off the jacket and to reach the desired temperature. (Arnold Rak HK-5.0 Heizkabel 230 V 75 W 5.0 m). The desired temperature for both reactors was 38.0 °C (mesophilic conditions). This temperature was chosen because it is the optimal temperature for bacteria to perform methanation.

2.2.4. Impact of the pH-value on the process performance

The pH-value in reactors has a strong importance like in every other bioprocess. The activity of bacteria depends strongly from this value and the tolerance range for methanogen archaea is narrow (between 6.8 pH and 7.8 pH). A smaller acid capacity with a higher charge of substrate can lead to increased organic acid concentrations with results in a sink of the pH value below 6.8 pH, which causes the inhibition of the methanogen archaea.

2.2.5. Organic acid dosing

A syringe of 100 mL was used to introduced organic acids in the reactors. For acetic acid, the weight of acid to introduced was measured with a kitchen balance (Crofton 96650, precision: 1 g). Then the acetic acid was directly introduced in reactors through the upper valve. Propionic acid and butyric acid were collected with a syringe of 10 mL and weighted with a laboratory balance (Mettler AE 200, precision: 10^{-4}g). Then they were diluted into 50 mL of water and introduced in reactors through the upper valve.

For some experimentations, a mix of sludge and acid were introduced in reactors. For this experimentations 300 g of raw sludge were first introduced. Then for propionic acid and butyric acid the acid was mixed with around 100 g of sludge and introduced. For acetic acid the solution was directly injected in reactors. Then 200 g of sludges (resp. 300 g for acetic acid) were injected for a total of 600 g. The raw sludge naturally contains acetic acid, propionic acid and butyric acid. Those mix were realized to study the behavior of the column when both three acids were in presence.

The quantities of injected acids were determined by the desired concentration of acid in reactors. For acetic acid dosing with concentration of 10 mmol/L were made. For propionic acid and butyric acid dosing of 5 mmol/L were made.

The hydrogen was injected by the hydrogen’s generator in R2. The amount of injected hydrogen was for each dosing 0.15 atm (15% of the gaseous volume in R2) with the exceptions of the two last dosing where the quantity of injected hydrogen was 0.10 atm (10% of the gaseous volume in R2).

2.2.6. Experimental Overview

The Table 2 summarized the dosing that were made during this thesis.

Table 2 Experiments realized during the project

Date	Dosing hour	Dosing (acids)	Hydrogene (atm)	Sample collection	Comment
18/05/2020	11:35:00	Acetate (10 mmol/L); 169g	0.15	5*2	5*2 = 1 sample every hour
19/05/2020	10:30:00	Acetate (10 mmol/L); 169g	0.15	5*2	2*6 = 3 samples in the morning and 3 in the evening
20/05/2020		nothing			
25/05/2020	11:00:00	Acetate (10 mmol/L); 169g	0.15	5*2	
26/05/2020	11:30:00	Acetate (10 mmol/L); 169g	0.15	5*2	
27/05/2020		nothing			
03/06/2020	13:45:00	Propionate (5mmol/L); 5.18g (R2 mixer broken)	0.15	5*2 + 1 point with 3 measures	Mixer in R2 broken (only R1 was operated)
08/06/2020	12:00:00	Propionate (5mmol/L); 5.18g	0.15	5*2	
09/06/2020	12:30:00	Propionate (5mmol/L); 5.18g	0.15	5*2	
10/06/2020	13:00:00	Propionate (5mmol/L); 5.18g	0.15	5*2	
15/06/2020		nothing			
16/06/2020	11:30:00	Propionate (5mmol/L); 5.18g	0.15	5*2	
18/06/2020	11:15:00	Acetate (10 mmol/L); 169g	0.15	5*2	
22/06/2020	12:15:00	Butyrate (5 mmol/L); 6.27/6.19 g	0.15	5*2	
24/06/2020	11:30:00	Butyrate (5 mmol/L); 6.27/6.19 g	0.15	6*2	
25/06/2020		nothing			
29/06/2020	14:50:00	Butyrate (5 mmol/L); 6.16/6.18 g	0.15	6*2	
30/06/2020		nothing			
01/07/2020	12:15:00	Propionate (5 mmol/L); 5.18/5.19 g	0.15	5*2	
07/07/2020	12:30:00	Raw sludge(600 g) + Propionate (5 mmol/L); 5.18/5.19 g	0	6*2	no H2 in R2
08/07/2020		nothing			
10/07/2020	11:30:00	Raw sludge(600g) + Propionate (5 mmol/L); 5.18/5.18 g	0.15	6*2	
13/07/2020	12:30:00	Raw sludge(600g) + Propionate (5 mmol/L); 5.20/5.19 g	0.15	5*2	
15/07/2020	12:00:00	Raw sludge(600g) + Acetate (10 mmol/L); 168/168 g	0.15	5*2	
17/07/2020	11:30:00	Raw sludge(600g) + Butyrate (5 mmol/L); 6.16/6.22 g	0.15	5*2	
20/07/2020	11:30:00	Propionate (5 mmol/L); 5.20/5.25 g	0.1	5*2	0.10 atm H2 in R2
22/07/2020	11:30:00	Propionate (5 mmol/L); 5.20/5.18 g	0.1	5*2	0.10 atm H2 in R2

2.3. Analytics

2.3.1. Reactor online data

Temperature and pH-value

The reactor's temperature is measured online with the temperature sensor of the pH-Redox-electrode (Endress + Hauser pH-/Redox-Sensors Memosens CPS16D). So, the performance of the heating jacket can be adjusted. The setpoint for the temperature in the reactor is 38°C. In addition to the online measurement, the temperature is also measured for both reactors (R1 and R2) during the daily handling.

Figure 12 shows the temperature for reactor 1 and 2 for one week, from 13th of July to 20th of July. The black curve is for R1 and the red one for R2. As it can be seen the temperature is maintained most of the time at 38 ± 0.2°C. The drops in the measured temperature are caused by introduction of 600 g of cold raw sludge from the fridge (5°C) inside both reactors. As the sludge is introduced at the level of the sensor, the variation in temperature is only local. Therefor the mixing and heating of the reactors brings back quickly to a temperature of 38°C. It is to noticed that those drops vary slightly from one reactor to the other and from one injection to another.

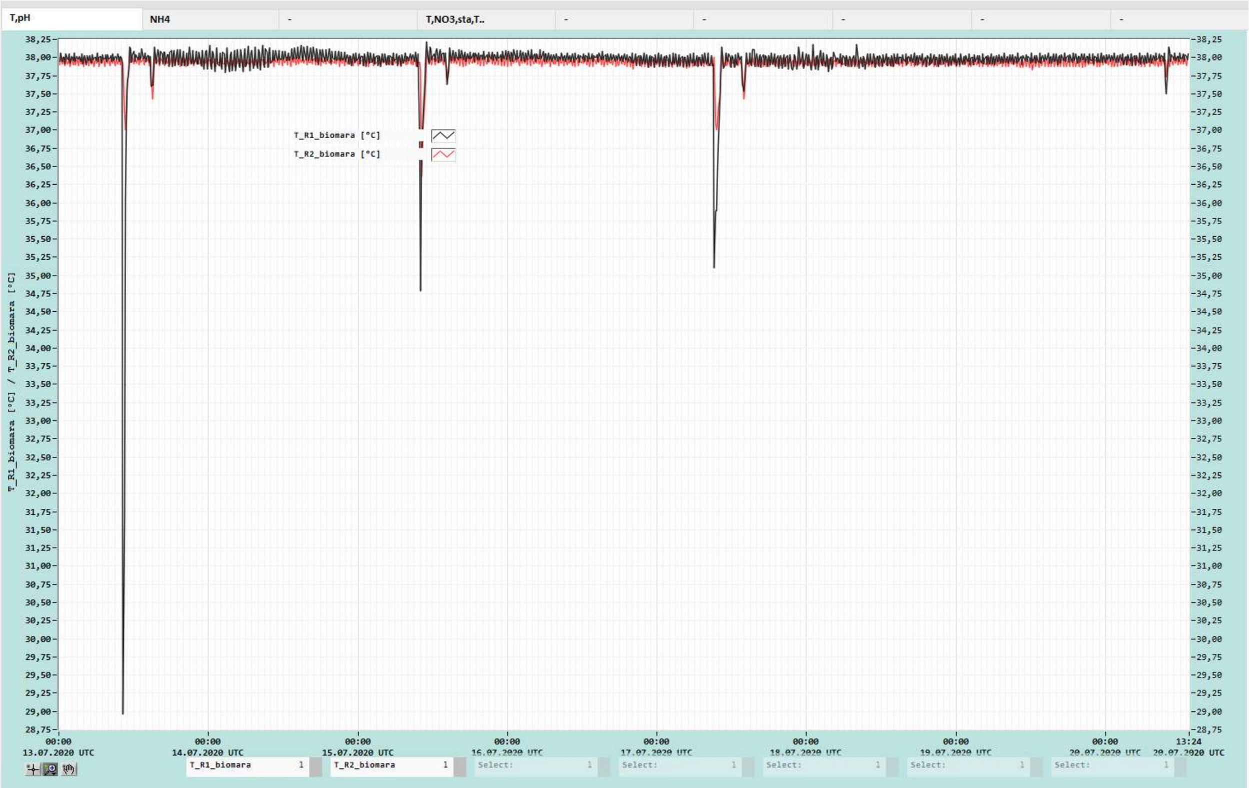

Figure 12 Online measure of temperature

The pH-value of the reactors is measured with an online sensor (Endress + Hauser pH-/Redox-Sensor Memosens CPS16D) and each measurement point is saved every 30 seconds. The pH-value is also daily controlled with a hand-sensor (Hach Lange HQ40d). This measure is used to calibrate the online sensor with an offset if a deviation (as of +/- 0.05 pH) between the hand-sensor and the online-sensor is detected. Every month the online pH-sensor is calibrated with a two-point method (buffer at pH 4 and pH 7). Figure 13 shows exemplarily the evolution of the pH-value in reactor 1 and 2 between the 13.07 and the 20.07.

Figure 13 Online measure of pH

The black curve shows the pH value of R1 and the red one the pH value for R2. The pH value varies between 7.15 pH and 7.25 pH for reactor 1, while the pH value varies between 7.45 pH and 7.65 pH for reactor 2. This difference is explained by the introduction of hydrogen in reactor 2, which induces a reduction of the partial pressure of CO_2. According to Henry's law, this leads into a reduction of the concentration of dissolved CO_2 inside the digester sludge and therefor to a rise of the pH-value. Also, pH drops can be noticed in **Error! Reference source not found.**. This is due to the combined introduction of raw sludge (pH value of the raw sludge varied between 4.8 pH and 5.6 pH) (naturally acid) and of the dosed organic acids. The pH then rises back within 12-14 hours after substrate dosage to its initial value. Caused by the degradation of the organic acids by the bacteria and to the mixing inside the reactors which homogenizes the content of the reactors.

Produced gas volume and gas production rate

As shown in Figure 8, the produced gas volume is measured with a drum gas counter (Ritter, TG 05 PVC). The produced gas volume is measured in steps of 10 mL and counted with an inductive pulse generator, which sends the pulses to a controller (Siemens Logo, 8.2). The controller sends the data to a SQL database, where it is saved as 10 second values. From this data the gas production rate can be deduced. Figure 14 shows the gas production for both reactors exemplarily for one week. On the 15.07. and the 17.07. the counter was reset to 0 after introduction of acids. As it can be seen, the introduction of a mixture of raw sludge and organic acid leads to a rise of the gas production. The production rate and the amount of the gas produced, depends from the type of the introduced acid. More details will be discussed in the following sections. To determine the composition of the gas mixture an IR-photometer was used (Saxon Junkalor Infralyt 80, Figure 15).

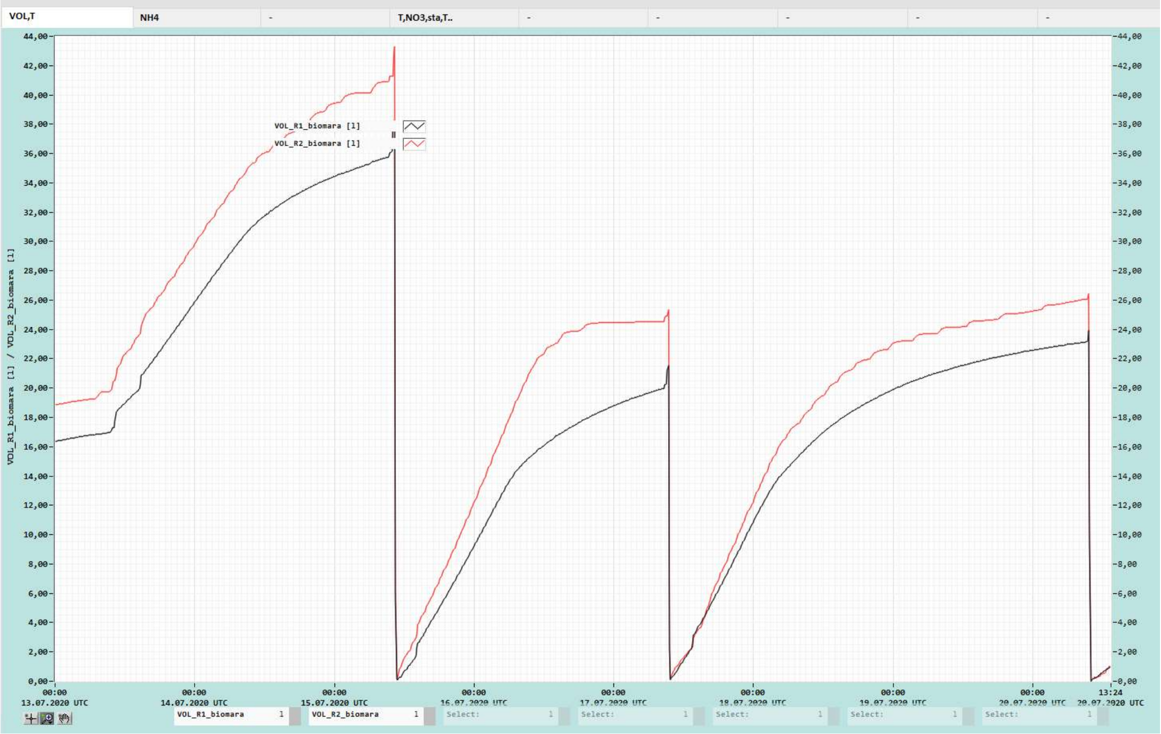

Figure 14 Online measure of gas production

Figure 15 IR-spectrometer (Saxon Junkalor Infralyt 80)

2.3.2. Raw and digester sludge samples

The raw sludge and digester sludge samples are taken on regular basis and analyzed to determine their dry mass (TS), their organic dry mass (oTS), their amount of chemical oxygen demand (COD), total phosphor (GP) and Total Kjeldahl nitrogen (TKN). Digester sludge samples are taken from the reactors two times a week. Raw sludge from each charge is tested during their first and last utilization approx. one's a week. The scale and frequency of the sampling is given in the Table 3. The concentrations of the components NH_4-N, PO_4-P and organic acid concentrations are also measured. The organic acid composition was determined with an HPLC chromatograph (HITACHI Primaid, Figure 16). The concentrations of CH_4, CO_2, O_2 and H_2S in the biogas are determined at regular intervals. And the gas is analyzed with a hand-held-meter (GFM series 100). All gas and sludge samples were analyzed in the laboratory of the Institute for water quality and resource managements at TU Wien. All used analytical methods are summarized in Table 4.

Table 3: Analyze plan for raw- and digester sludge samples

Weekday	TS	oTS	NH_4-N	PO_4-P	COD	TKN	TP	org. acids
Digester sludge								
Tuesday	x	x	x	x	x	x	x	x
Thursday	x	x	x	x	x	x	x	x
Raw sludge								
Every charge min. 3 times	x	x	x	x	x	x	x	x

Table 4: Used analytic method

Parameter	Methods	Note
COD	DIN 38409	DEV H 43-1 short time process
TKN, TP	DIN 19684 Part 4	DEV H11 photometric variant
NH_4	DIN 38406	DEV D5-1 photometric
PO_4	DIN 38405	DEV D11-3 photometric
Organic acids	Aminex HPX-87 H column, 5 mN H_2SO_4 Solvent front, UV-Detector And measure with 625 nm	High Performance Liquid Chromatography (HITACHI Primaid)
TS, oTS	DIN 38409 Part 1	DEV H1 Annealing temperature 550°C
pH-value	DIN 38404 Part 15	DEV C5 WTW Combination electrode SenTix20

Figure 16 High performance liquid chromatography HITACHI Primaid, including Aminex HPX-87 H column and UV-Detector

2.3.3. Sampling and sample preparation before the organic acid measurement

Two different processes were used for the collection of samples. At the first process one sample for each reactor was collected 30 minutes after the dosing of organic acid in the reactors. Then one sample was collected every hour for each reactor. This process is described in the Table 5.

Table 5 Schedule of the probes after the start of a dosing (first process).

Sample	1	2	3	4	5
Time after dosing	+ 30 min	+ 1 h	+ 2h	+ 3 h	+ 4 h

For the second process, 3 samples spaced from 10 min were collected 30 minutes after the dosing. Then 3 samples (spaced from 10 min) were taken 5 hours after the dosing and 3 samples (spaced from 10 min) were taken the next day on the morning (approx. 24 h after the dosing). This process is described in Table 6.

Table 6 Schedule of the probes after the start of a dosing (second process)

Sample	1,2,3	4,5,6	7,8,9
Time after dosing	30-40-50 min	5h00-5h10-5h20	22h00-22h10-22h20

After the sampling, the 45 ml sludge samples were immediately centrifuged at 10,000 g in the lab centrifuge. Then 1 mL of supernatant was filtered with a syringe of 10 mL, filtered with a disposable filter (CHROMAFIL GF/RC-45/25) and stored in the fridge up to the HPLC analysis.

Figure 17 Syringe attachment filters, two steps 180µm and 0.45µm pore wide (CHROMAFIL GF/RC-45/25)

2.4. Experimental calculation

2.4.1. Determination of the quantities of acid to inject in reactors

The calculation of the mass needed for organic acid is given by formula 8.

$$\mathbf{m = M * C * V_r} \qquad (8)$$

m	Dosed organic acid mass	[g]
M	Molar mass of the organic acid	[g/mol]
c	Concentration of the organic acid in the reactor	[mol/L]
V_r	Volume of the reactor (14 L)	[L]

Formula (8) is enough for the propionic acid and the butyric acid that were used pure. For the acetic acid another calculus is necessary since it was under the form of an aqueous solution with a concentration in acetic acid of 5% in term of mass. The formula (9) gives the mass for the solution of acetic acid.

$$\mathbf{m = \frac{M * c * V_r}{0,05}}$$

The Table 7 summarize the value of mass for each acid:

Table 7 Molar mass, desired concentration in reactors and injected mass of acetic acid, propionic acid and butyric acid

	Acetic acid	Propionic acid	Butyric acid
Molar Mass	60.052 g/mol	74.08 g/mol	88.11 g/mol
Concentration in reactor	10 mmol/L	5 mmol/L	5 mmol/L
Mass	168 g	5,18 g	6,17 g

2.4.2. Stoichiometric calculations

The stoichiometry for the conversion of each organic acid into biogas is given in Table 8.

Table 8 Stoichiometry of conversion process of acetic acid, propionic acid and butyric acid

Acetic acid	$C_2H_4O_2 = CH_4 + CO_2$
Propionic acid	$C_4H_8O_2 + \frac{1}{2} H_2O = \frac{7}{4} CH_4 + \frac{5}{4} CO_2$
Butyric acid	$C_4H_8O_2 + H_2O = \frac{5}{2} CH_4 + \frac{3}{2} CO_2$

Using the chemical stoichiometry and the ideal gas law, it is possible to calculate the volume of gas that a certain quantity of acid will theoretically produce. Table 9 gives the theoretical value of the produced gas volume, obtained for the quantity of each acid introduced in the reactor. The temperature and pressure taken for the calculus were 25°C respectively 1 bar.

Table 9 Theoretical produced gas volume for acetic acid, propionic acid and butyric acid

	Acetic acid	Propionic acid	Butyric acid
Dosed organic acid [mmol]	140 mmol	70 mmol	70 mmol
Theoretic biogas yield [L]	6.94 L	5.20 L	6.94 L

3. Results

Online measures in the case of these study gave data on the amount of gas produced during each dosing. The regular measures made before each dosing inform about the quantity of organic acids inside each column before dosing. The results indicate that these amounts of acids were in general negligible (< 10 mg/L) (see appendix). The measures on the organic acids were made to monitor the evolution of their concentration during the dosing.

3.1. Acetic acid dosing

3.1.1. Pure Acetic acid

In the following diagrams the evolution of the concentration of acetic acid in the digester sludge for six dosing experiments is presented. Those dosing experiments were performed on the 18/05, the 19/05, the 25/05, the 26/05 and the 18/06. The dosed acid was acetic acid and digester sludge samples were taken according to the method described in part 2.3.3. The same amount of acetic acid was introduced in each reactor 140 mmol (169 g stock solution).

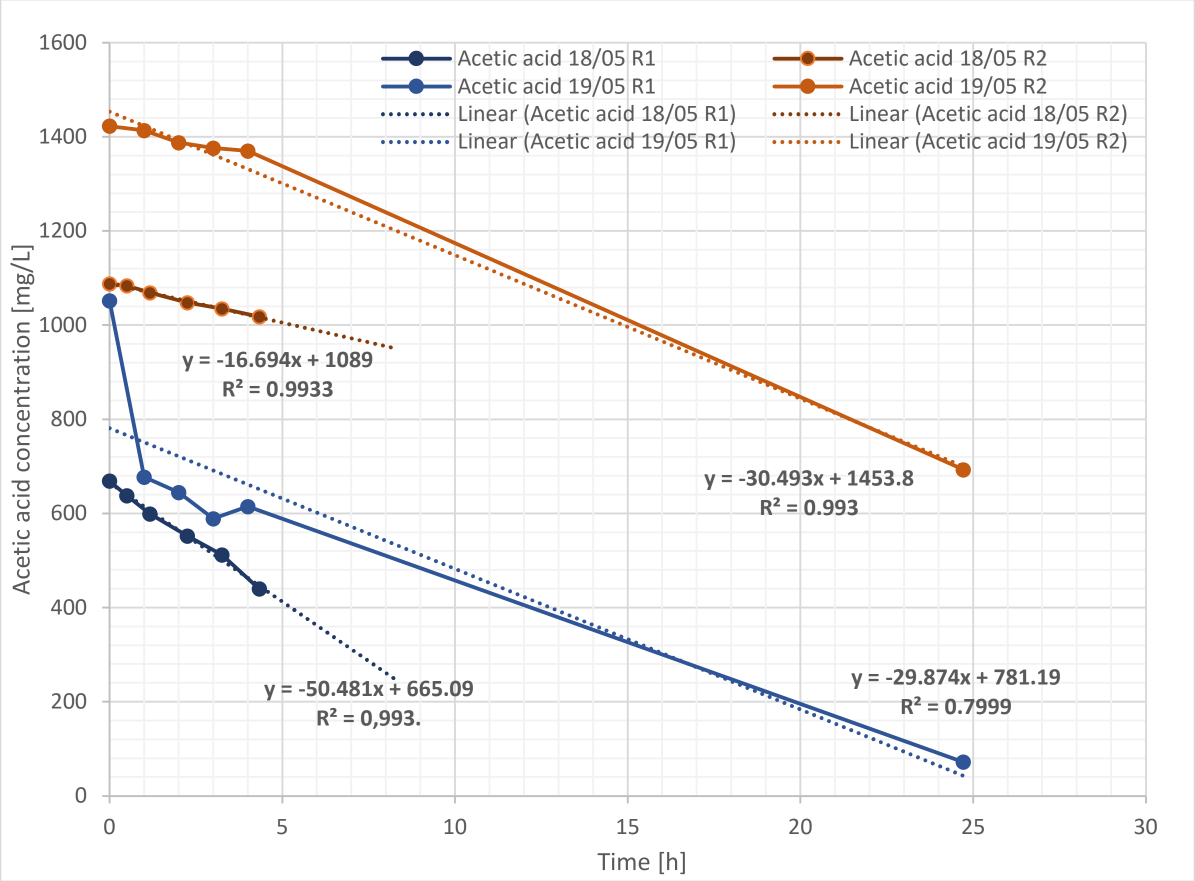

Figure 18 acetic acid concentration for R1 and R2 for the dosing experiment on the 18/05 and the 19/05.

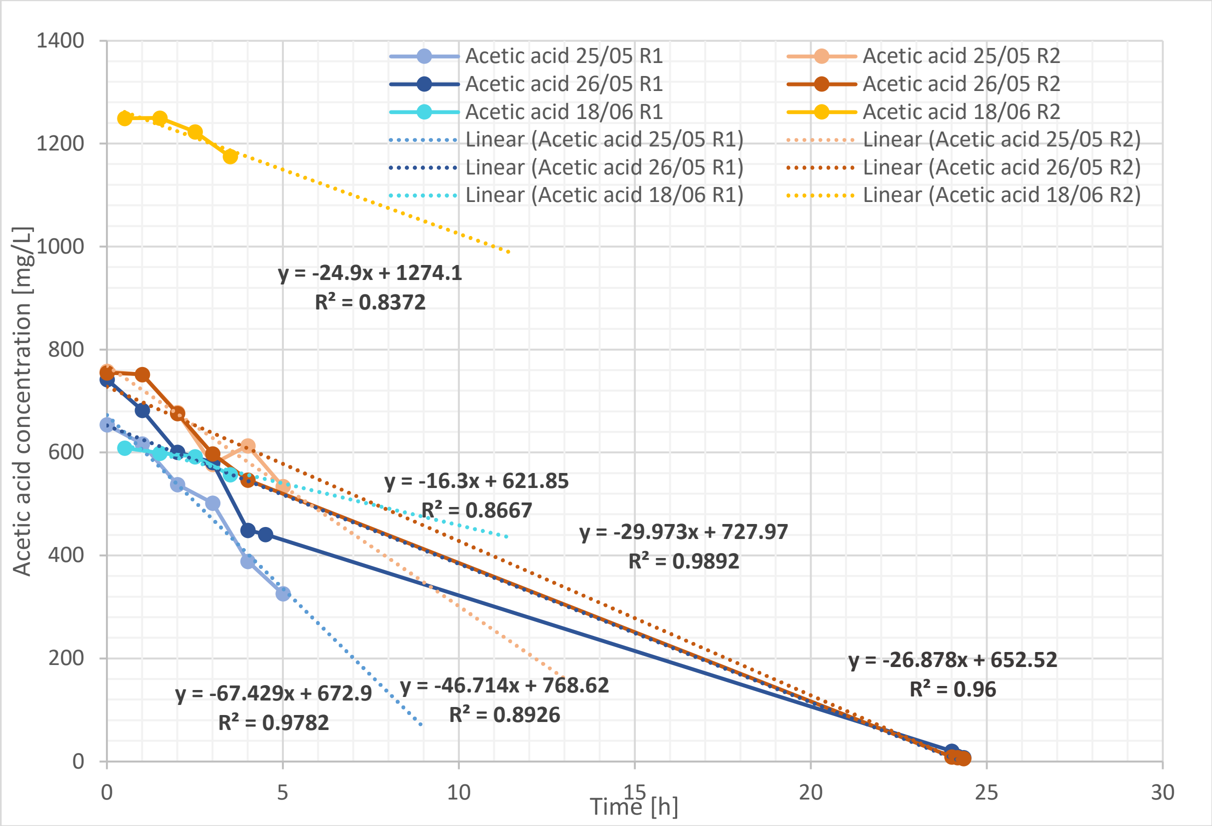

Figure 19 acetic acid concentration for R1 and R2 for the dosing experiment on the 25/05, the 26/05 and the 18/06

As it can be seen in the Figure 18 and in the Figure 19 the initial value of the concentration can strongly vary from one reactor to another (or from one dosing to the other) due to the initial concentration of acetic acid in the reactor. As an example, the concentration in R2 of acetic acid before the dosing of the 18/05 was 383 mg/L which results in a much higher concentration of acetic acid compare to R1. The same observation can be made for the dosing on the 18/06. The value of 1052 mg/L measured for R2 during the dosing of the 19/05 is high when compared to the value of the 18/05. This is certainly due to the interval of only 24 hours between this dosing and the previous one. Contrarily to R1, R2 hadn’t enough time to completely eliminate the acetic acid introduced the previous day which resulted in an accumulation of the acetic acid introduced during this dosing and during the previous dosing and therefore a much higher concentration in acetic acid. The abnormally high initial value for R1 for the dosing of the 19/05 can be explained through inhomogeneity of the sludge inside R1 at the beginning of the dosing

Most of the time, the initial concentration of acetic acid varied between 600 and 800 mg/L. Which is consistent with the amount of introduced acetic acid since a concentration of 10 mmol/L represents a concentration of 600 mg/L. The orders of magnitude are equivalent. The acetic acid is degraded during the dosing and it can be estimated that it takes approximatively 25 hours for both reactors to completely degrade the dosed 140 mmol acetic acid. A linear model was chosen according to the zero-order reaction to simulate the evolution of the concentration during the dosing. The rates of degradation of acetic acid are deduced from this model. They vary from 16.3 mg/(L · h) to 67.43 mg/(L · h). They are summarized with the coefficient of determination (R²) in the Table 10.

Table 10 Degradation's rate of R1 and R2 for the dosing with acetic acid

Date	Degradation rate [mg/(L · h)]		Degradation rate ratio	R^2	
	R1	R2	R2/R1	R1	R2
18/05/2020	**-50.481**	**-16.694**	**33.07%**	**0.9935**	**0.9933**
19/05/2020	**-29.874**	**-30.493**	**102.07%**	**0.7999**	**0.9933**
25/05/2020	**-67.429**	**-46.714**	**69.28%**	**0.9782**	**0.8926**
26/05/2020	**-26.119**	**-29.973**	**114.76%**	**0.96**	**0.9892**
18/06/2020	**-16.3**	**-24.9**	**152.76%**	**0.8667**	**0.8372**
Average	**-38.0406**	**-29.7548**	**78.22%**		
Average (without 18/05)	**-34.9305**	**-33.02**	**94.53%**		

It can be noticed that the degradation rates vary strongly from one reactor to the other and from one dosing to another. The ratio between the degradation rates of R1 and R2 informs about their difference. A ratio of 100% indicates that the degradation rates in R1 and R2 are the same. A ratio of 50% indicates that the rate of R1 is twice higher as the rate of R2, while a ratio of 200% indicates the contrary. In this case the ratios vary strongly from one dosing experiment to another but it doesn't seem that a reactor has systematically a higher degradation rate than the other. The average ratio between R1 and R2 is 78% and if the dosing of the 18/05 is not taken in count (since the ratio is really weak compare to the others) it rises to 94.5%. These results indicate that there are no big differences in the degradation of acetic acid between R1 and R2. This difference can be explained by difference in the biomass concentration between R1 and R2. Which leads to the conclusion that the introduction of hydrogen inside R2 leads to no inhibition for the acetic acid degradation.

The coefficient of determination R^2 varies from 0.7999 to 0.9935. Most of the time this coefficient is very close from 1 for both reactors which signifies that the linear model is a good model to describe the evolution of the concentration of acetic acid during the dosing experiments in both reactors.

3.1.2. Mix of acetic acid and raw sludge

In the following diagram the acetic acid concentration of during the dosing experiment with raw sludge on the 15/07 is displayed. The amount of introduced acetic acid was the same as the previous dosing (140 mmol) but in addition 600 g of raw sludge were also introduced. And a linear model was used to simulate the evolution of the acetic acid's concentration.

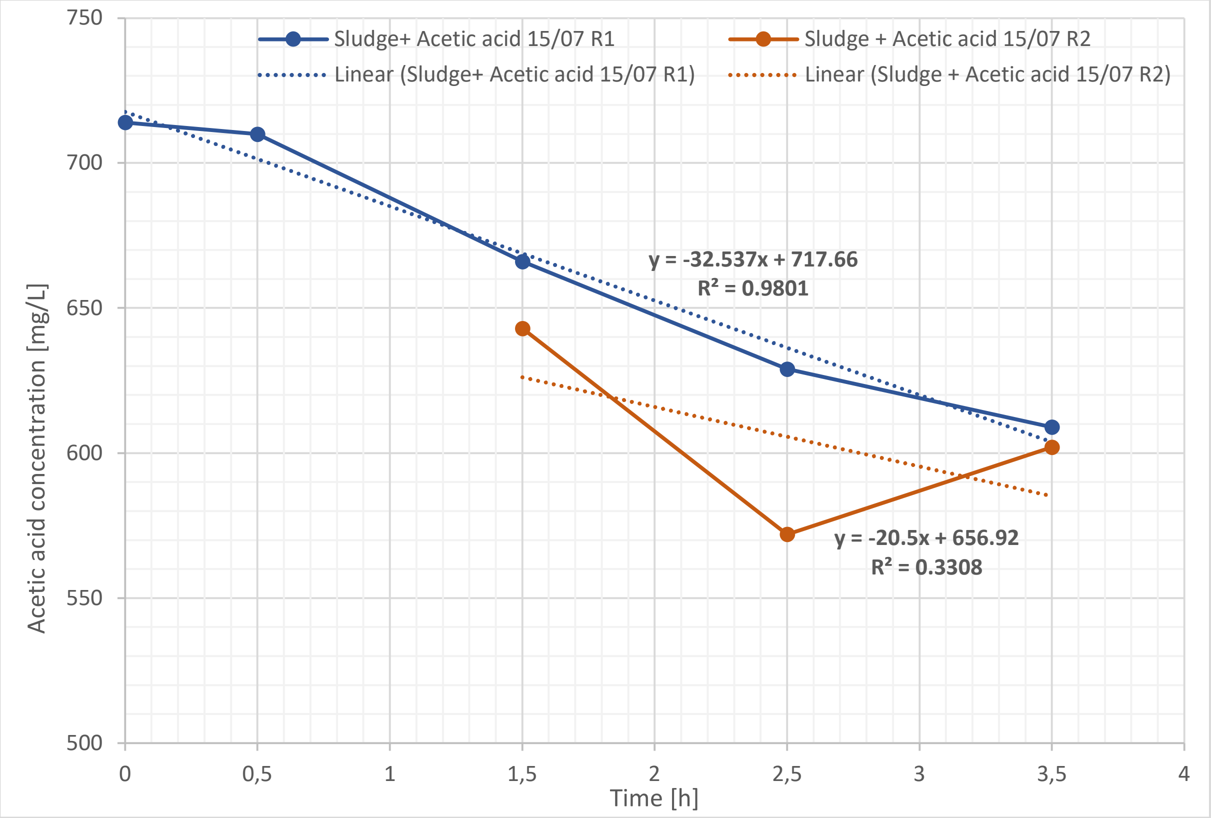

Figure 20 Diagram of evolution of the concentration of acetic acid for R1 and R2 for the dosing with sludge of the 15/07

The two first measurement points of R2 are not represented in Figure 20 due to some inconsistency. However, the acetic acid concentration before the dosing test was null for both reactor (see appendix). It can be assumed that the acetic acid concentration in both reactors at the beginning of the dosing was the same. The concentration of acetic acid in R2 after 3.5 hours was the same as the concentration in R1 so through extrapolation it can be deduced that their degradation rates were very close. It can also be noticed that this degradation rate is close to the values observed for the dosing of pure acetic acid. Furthermore, there are no difference between both reactors. The addition of 600 g raw sludge in the reactor doesn't seem to impact the behavior of the acetic acid degradation in the reactors.

The coefficient of determination R^2 is very close from 1 for R1 (0.9801) which indicates that the linear model is a good model to describe the evolution of the acetic acid concentration during the dosing with sludge in R1. The value of R2 (0.3308) is very low. It can be explained by the fact that the data for R2 are inconsistent for this dosing. It can be due to inhomogeneity in the reactor, error of measure.

3.1.3. Gas production

During the several dosing the volume of the produced gas was measured with a pycnometer. The collected data were then exported and treated with an excel sheet. The following diagrams show the volume of the produced gas during the dosing with acetic acid made during the 18/05, the 19/05, the 25/05 and the 26/05. Each curve starts at the beginning of the dosing which doesn't correspond to the reset of the gas counter (which explains why the curves don’t start at zero). Some curves present a form of oscillation. It is due to a technical default of the pump that send the gas from R2 to the pycnometer.

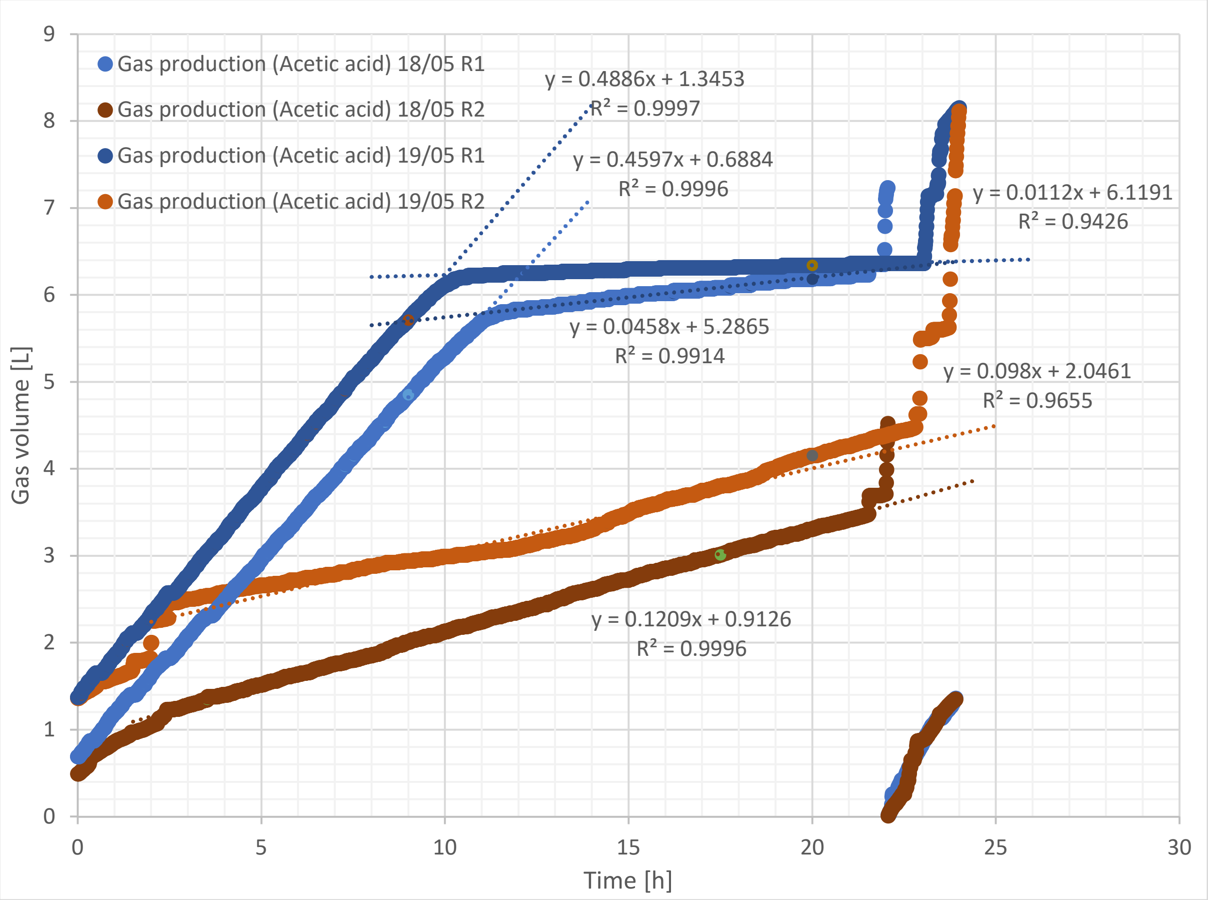

Figure 21 Diagram of evolution of the gas production for R1 and R2 for the dosing of the 18/05 and 19/05

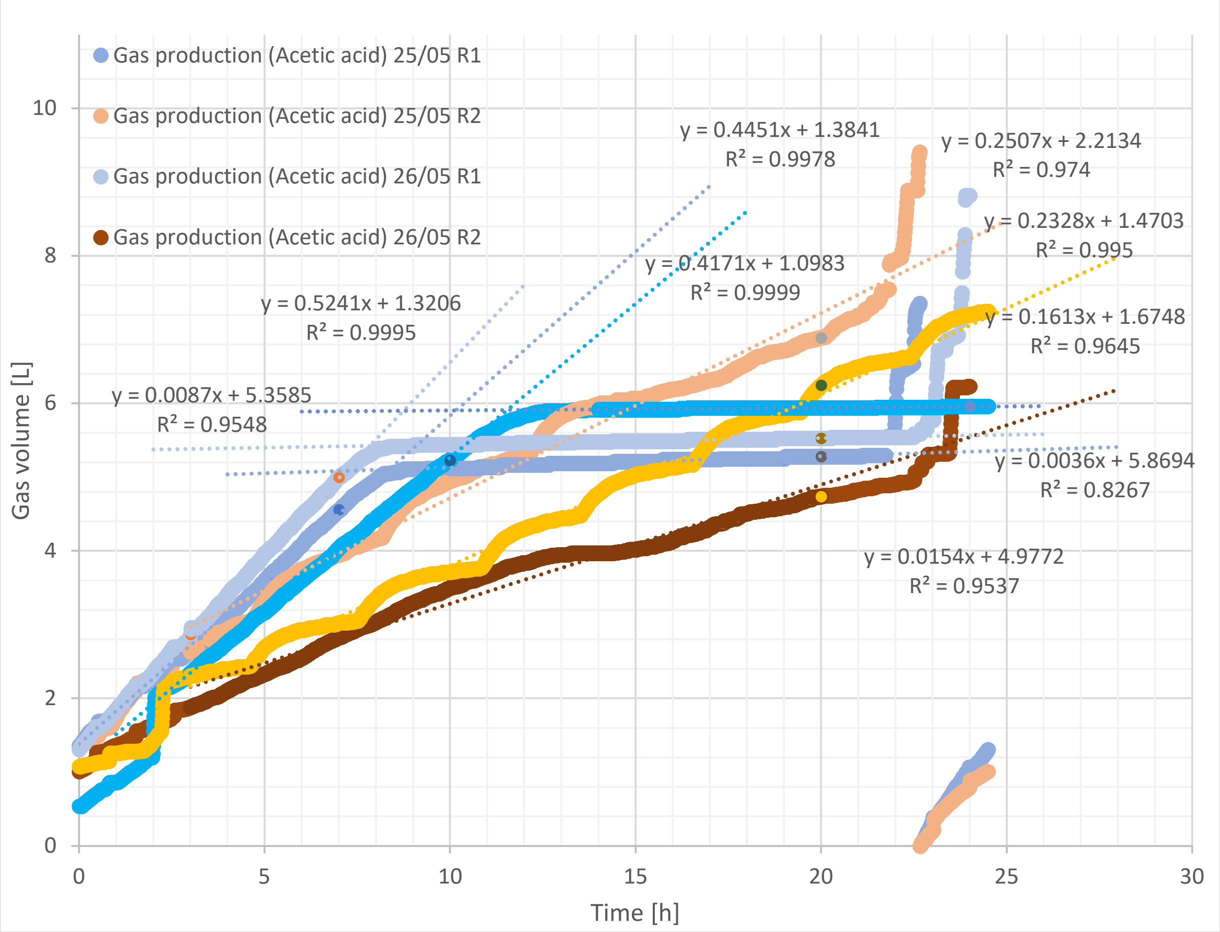

Figure 22 Diagram of evolution of the gas production for R1 and R2 for the dosing of the 25/05, 26/05 and 18/06

The production rates of gas for both reactors and each dosing are gathered in the Table 11. The ratio between the rates of R2 and the rates of R1 are also given.

Table 11 Gas production rates of R1 and R2 for the dosing with acetic acid

Date	Step	Production Rate (L/h)		Production (L)		Ratio R2/R1
		R1	R2	R1	R2	
18/05/2020	**1**	**0.4597**	**0.1209**	**5.06**	**2.83**	**26.30%**
	2	**0.0458**				
19/05/2020	**1**	**0.4886**	**0.098**	**4.82**	**2.79**	**20.06%**
	2	**0.0112**				
25/05/2020	**1**	**0.4451**	**0.2507**	**3.72**	**5.53**	**56.32%**
	2	**0.0154**				
26/05/2020	**1**	**0.5241**	**0.1613**	**4.03**	**3.66**	**30.78%**
	2	**0.0087**				
18/06/2020	**1**	**0.4171**	**0.2328**	**5.32**	**5.23**	**55.81%**
	2	**0.0036**				

For each reactor it can be observed two different behaviors. For R1 the curve is separated in two parts for every dosing. The first part is linear and shows a high production of gas with rates that vary between 0.41 L/h and 0.53 L/h. Those rates are also higher than the rate of R2. The ratio between the rate of R1 and R2 illustrate very well this fact. This first part goes from the beginning of the dosing to approximatively 8-12 hours later (depending of the dosing). The second part is characterized by a production of gas nearly reduced to zero for R1. For R2 the curve keeps its aspect from the beginning of the dosing to the end with a constant production's rate. These behaviors are observed for each dosing made with acetic acid. It can also be noticed than the total amounts of produced gas during the dosing are lower than the theoretical value of 6.94 L calculated in the previous part. This is due to the use by the bacteria of a part of the sludge for biomass build up rather than gas production. Although the introduction of hydrogen doesn't inhibit the degradation of acetic acid it does however impacts the gas production.

At the end of the curves it can be observed a very strong rise of the gas production for both R1 and R2. And a jump at the beginning of some curves can also be observed. The causes for both of it were not identified during the experimentations.

The Figure 23 shows the gas production for the dosing made during the 15/07 with acetic acid and sludge. Like the previous diagram the curves start at the beginning of the dosing and not at the moment where the gas counter was reset.

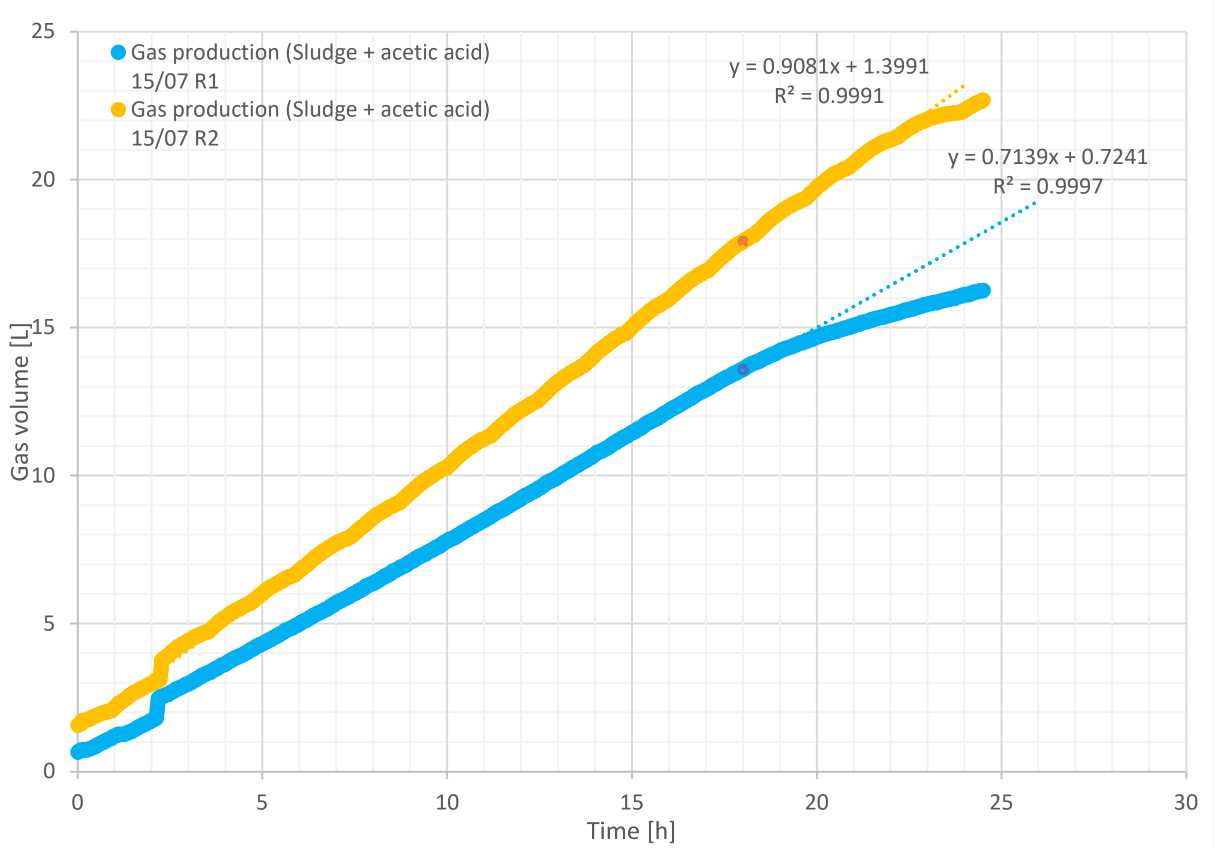

Figure 23 Diagram of evolution of the gas production for R1 and R2 for the dosing with raw sludge of the 15/07

The production rates of gas for both reactors are gathered in the Table 12. The ratio between the rates of R2 and the rates of R1 is also given.

Table 12 Gas production rates of R1 and R2 for the dossing with acetic acid and raw sludge

Date	Production Rate (L/h)		Production (L)		Ratio R2/R1
	R1	R2	R1	R2	
15/07/2020	**0.7139**	**0.9081**	**15.55**	**21.07**	**127.20%**

It can be observed that contrarily to the previous diagrams the gas production is quite similar from one reactor to another. The curves are linear and the production rate are respectively for R1 and R2 of 0.7139 L/h and 0.9081 L/h. The production rate of R2 is higher than the one of R1. The ratio of 127.2% between R1 and R2 illustrates this fact very well. Furthermore, the quantity of produced gas is higher with sludge than without sludge. It can be seen that after 20 hours the amount of produced gas was higher than 15 L for both reactors. While for the dosing with only acetic acid this quantity was lower than 7 L. It can also be noticed that 20 hours after the beginning of the dosing experiment the gas production starts to decrease in both reactors after the acetic acid is degraded.

3.2. Butyric acid

3.2.1. Pure butyric acid

During the dosing with butyric acid, 6.19 g of this acid was injected in each reactor. So, the butyric acid concentration in the reactor would reach 5 mmol/L. In the following diagram butyric acid concentrations are represented for the dosing experiments made during the 22/06 and the 24/06. A concentration of 5 mmol/L is equivalent to a concentration of 440 mg/L. Furthermore, the amounts of butyric acid inside both reactors before the dosing were close to zero (see appendix). Therefore, the initial concentrations in the diagram are consistent with the theoretical one.

It is noticed that the sample collection for the dosing of the 24/06 was made according to the method described in Table 6. The values displayed on the diagram for this dosing are the mean value of the three measures made at the beginning and at the end of the dosing. A linear model was used to simulate the evolution of the butyric acid's concentration for each dosing in both reactors.

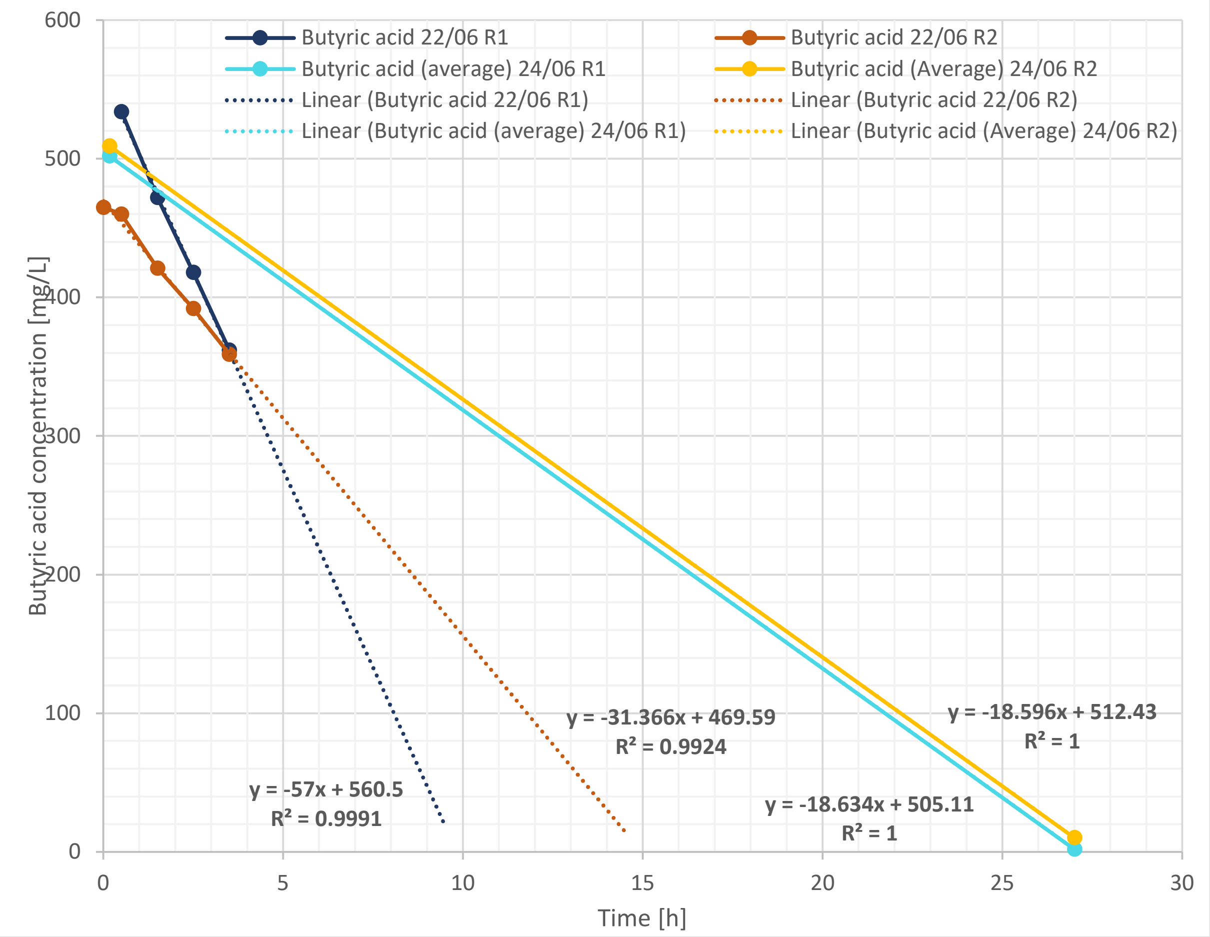

Figure 24 Diagram of evolution of the concentration of butyric acid for R1 and R2 for the dosing of the 22/06 and the 24/06

For the dosing experiment on the 24th of June, just at the beginning and 27h after the dosing samples were taken. So, this experiment cannot be used for the calculation of inhibition factors. The degradation's rate for each dosing for both reactors are summarized in the Table 13.

Table 13 Degradation's rate of R1 and R2 for the dosing with butyric acid

Date	Degradation rate [mg/(L · h]		Degradation rate ratio	R^2	
	R1	R2	R2/R1	R1	R2
22/06/2020	**-57**	**-31.366**	**55.03%**	0.9991	0.9924
24/06/2020*	**-18.634***	**-18.596***	**99.80%***	1*	1*

*data not considered in the inhibition calculation (to little data)

The value of this rate vary from 32 to 57 mg/(L · h). The first dosing indicates that the time for R1 to completely degrade the butyric acid is around 10 hours and for R2 15 hours. It suggests also that there is an inhibition of the degradation of butyric acid in R2 as it can be seen with the ratio of 55% between the degradation rates of R1 and R2. With these results it can be deduced that the curve for the dosing of the 24/06 may be not representative of the actual behavior of both column during this dosing. This likely due to the fact that the measure points are too spaced and don't reflect the evolution of the butyrate concentration in the reactor.

The correlations coefficient R^2 for the dosing of the 22/06 is very closed from 1 for both reactors which indicates that the linear model is adapted to describe the evolution of the butyric acid's concentration during this dosing. The value of R^2 for the other dosing is equal to 1 which is due to the fact that only two points were used to make the diagram for this dosing.

3.2.2. Mix of butyric acid and sludge

In the Figure 25 the evolution of the butyric acid concentration in both reactors for the dosing made during the 17/07 with a mixture of raw sludge and butyric acid is presented. The amount of introduced butyric acid was the same as in the previous part (5 mmol/L) and the amount of introduced raw sludge was 600 g. A linear model was used to simulate the evolution of the butyric acid's concentration in both reactors.

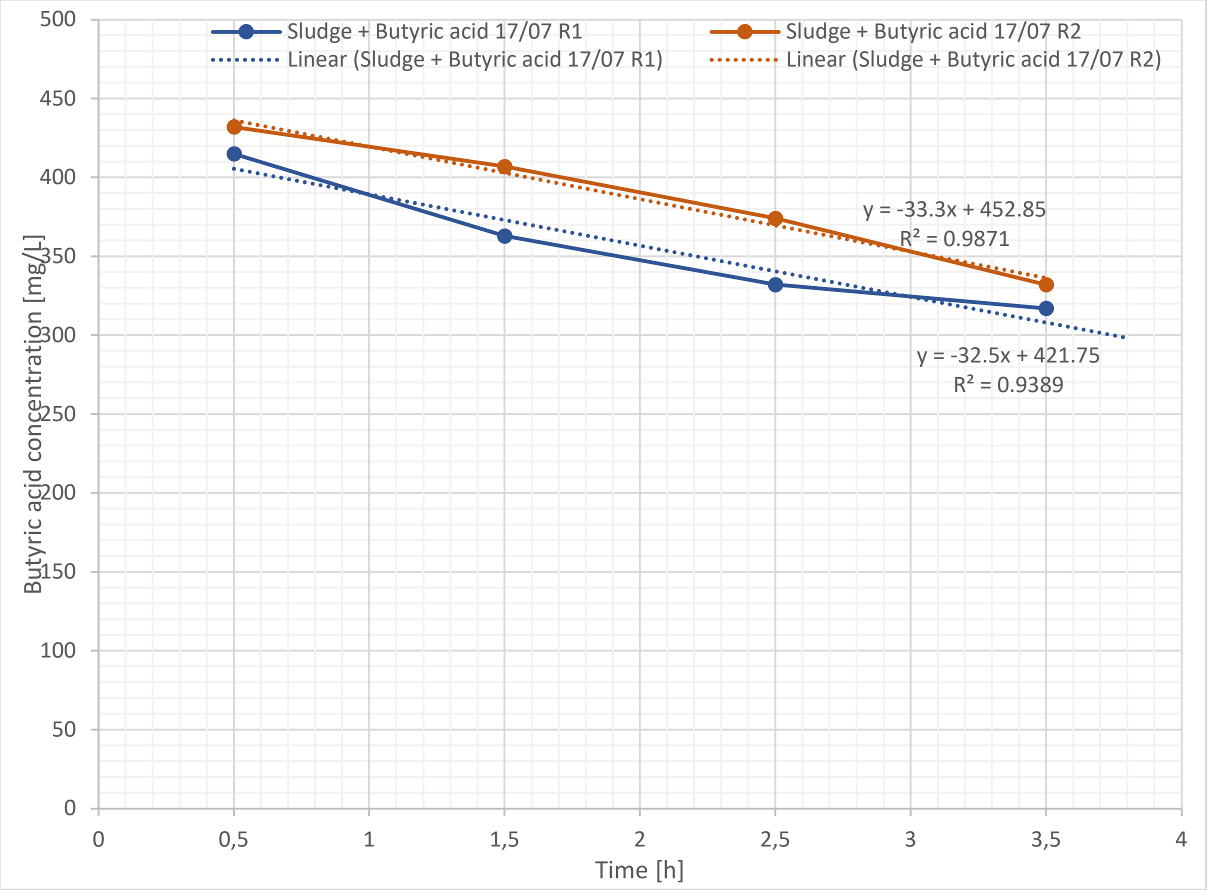

Figure 25 Diagram of evolution of the concentration of butyric acid for R1 and R2 for the dosing with raw sludge of the 17/07

The two first points (at 0 on the abscise) for each reactor were not taken into count due to values lower than the other points. This was certainly caused by the fact that at the time of the first sample collection the butyric acid wasn't homogenously mixed inside the reactors. It can be noticed that the curves of the concentration of butyric acid for R1 and R2 are similar and the degradation's rate are very close with a value of 32.5 mg/(L · h) for R1 and 33.3 mg/(L · h) for R2. The values of the correlation coefficient R^2 are close from 1 for both curves which indicates that the linear model is adapted to describe the evolution of butyric acid's concentration in both reactors for this dosing. With these data it can be assumed that the introduction of H_2 doesn't inhibit the degradation of butyric acid.

3.2.3. Gas production

The Figure 26 shows the volume of the produced gas during the dosing experiments with butyric acid made during the 22/06 and the 24/06. Each curve starts at the beginning of the dosing. Some curves present a form of oscillation. It is caused by hydrogen dosing in the gas circulation pipe of reactor 2, which influences the drum gas counter. It affects only the curves of R2. A jump at the beginning of some curves can also be observed. The cause of those jumps is the introduction of 600 mL of raw sludge at the beginning of the dosing.

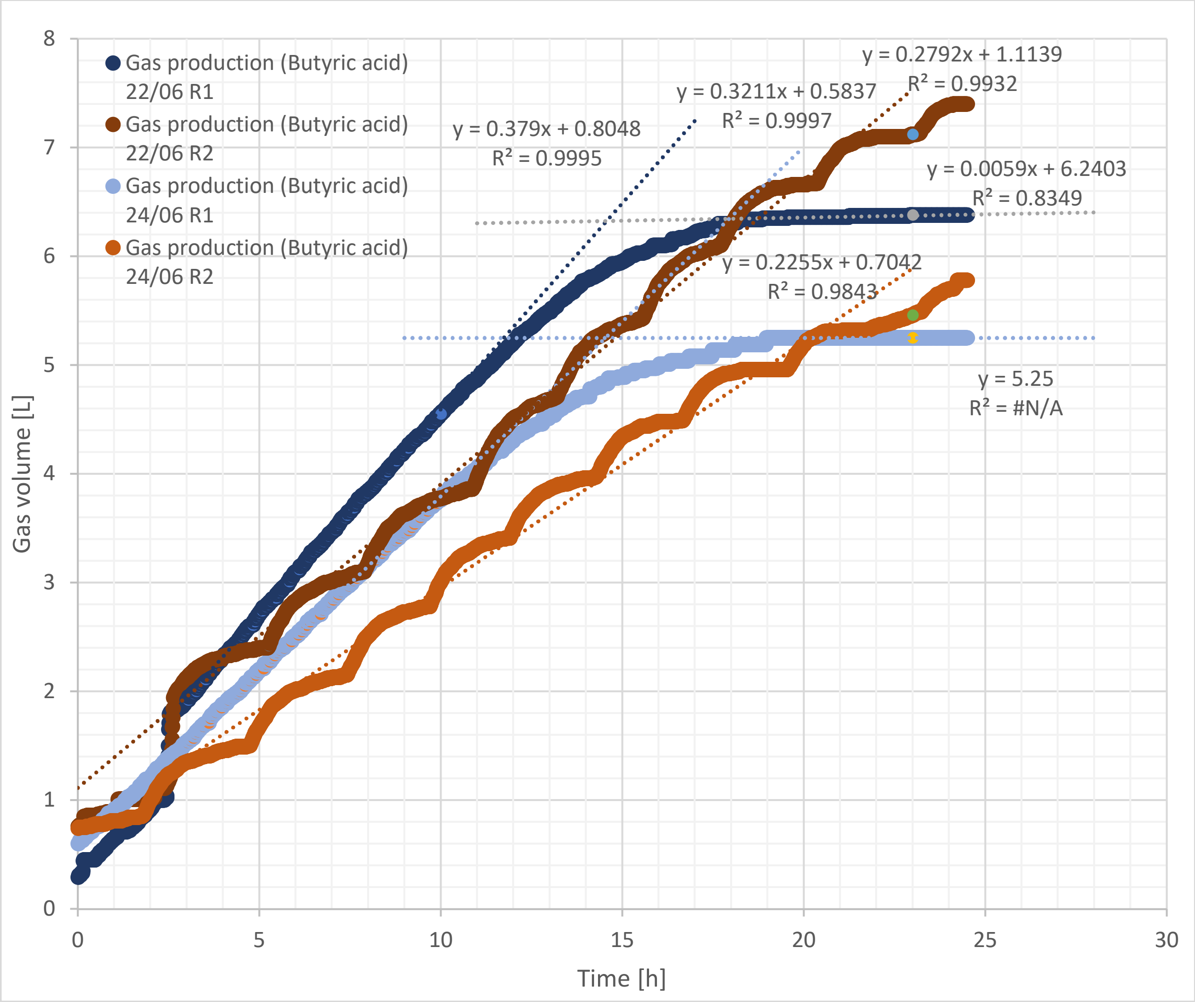

Figure 26 Diagram of evolution of the gas production for R1 and R2 for the dosing of the 22/06 and 24/06

The production rates of gas for both reactors and each dosing are gathered in the Table 14. The ratio between the rates of R2 and the rates of R1 are also given.

Table 14 Gas production rates of R1 and R2 for the dossing with butyric acid

Date	Step	Production Rate (L/h)		Production (L)		Ratio R2/R1
		R1	R2	R1	R2	
22/06/2020	**1**	**0.379**	**0.2792**	**5.96**	**5.8**	**73.67%**
	2	**0.0059**				
24/06/2020	**1**	**0.3211**	**0.2255**	**4.62**	**4.49**	**70.23%**
	2	**0.0059**				

It is to notice that the behavior of the curves is very similar with the one observed with the dosing of acetic acid. For R1 the curve is separate in two parts: a first part with a high production rate and a second one with a production rate practically reduce to zero. The difference is about the time where the transition from one part to another happens. It is around 15 hours here (while with acetic acid it is around 8-12 hours). For R2 the curves keep the same gradient from the beginning to the end. It can also be notice that the ratio between

R2 and R1 are much higher (73.67% and 70.23%) than the ones with acetic acid. The production rates for the first step of R1 are also lower than the ones with acetic acid. While the production rate for R2 are similar to the one made with acetic acid. It can also be noticed than the total amounts of produced gas during the dosing are lower than the theoretical value of 6.94 L calculated in the previous part. The reasons stay the same (part of the sludge used for biomass build up).

The Figure 27 shows the gas production for the dosing made during the 17/07 with butyric acid and sludge. Like the previous diagrams the curves start at the beginning of the dosing.

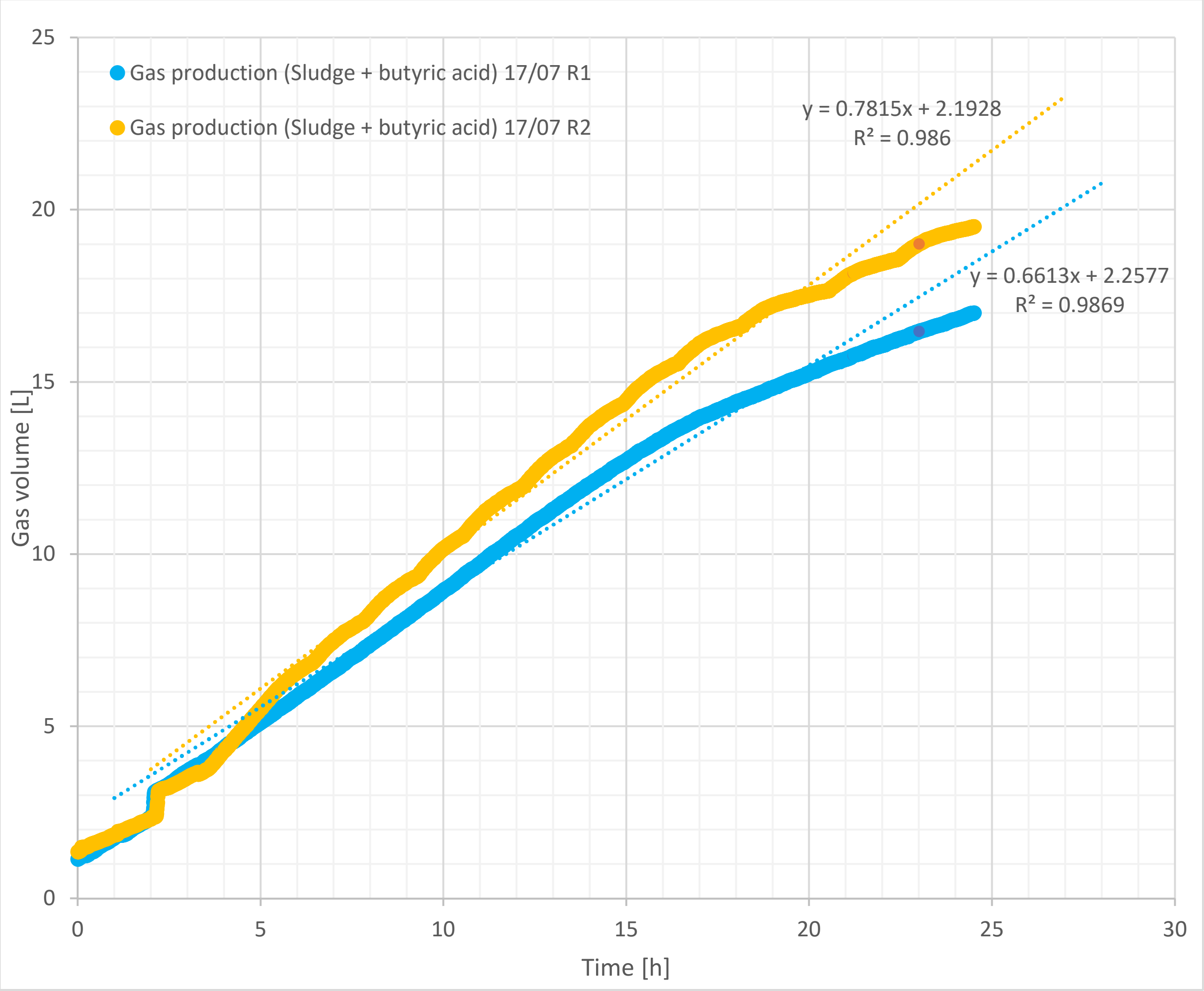

Figure 27 Diagram of evolution of the gas production for R1 and R2 for the dosing with raw sludge of the 17/07

The production rates of gas for both reactors and each dosing are gathered in the Table 15. The ratio between the rates of R2 and the rates of R1 are also given.

Table 15 Gas production rates of R1 and R2 for the dossing with butyric acid and raw sludge

Date	Production Rate (L/h)		Production (L)		Ratio R2/R1
	R1	R2	R1	R2	
17/07/2020	**0.6613**	**0.7815**	**15.61**	**18.1**	**118.18%**

It can be observed that the behavior of the gas production with butyric acid and sludge is similar to the gas production with acetic acid and sludge. The production rate for R1 is 0.6613 L/h and for R2 0.7815 L/h. The rate of R2 is higher than R1. And the volume of produced gas with sludge is higher than 15 L after 20 hours for both reactors. While for the dosing with only butyric acid the volume of produced gas was lower than 7 L.

3.3. Propionic acid

3.3.1. Pure propionic acid

Several dosing experiments with propionic acid were performed. The Figure 28 and the Figure 29 represent the propionic acid concentration during the dosing tests on 3/06, 8/06, 9/06, 10/06 and 16/06 for both reactors. For each dosing 5.18 g of propionic acid (70 mmol) was injected to reach a concentration of 5 mmol/L in both reactors. Which is equivalent to a concentration of 370 mg/L. The propionic acid concentration before dosing varied between 0 and 220 mg/L (see appendix). The propionic acid concentration at the beginning of the dosing are therefore consistent with the exception of the dosing of the 16/06 for R2. A linear model was used to simulate the evolution of the propionic acid concentration for each dosing test in both reactors.

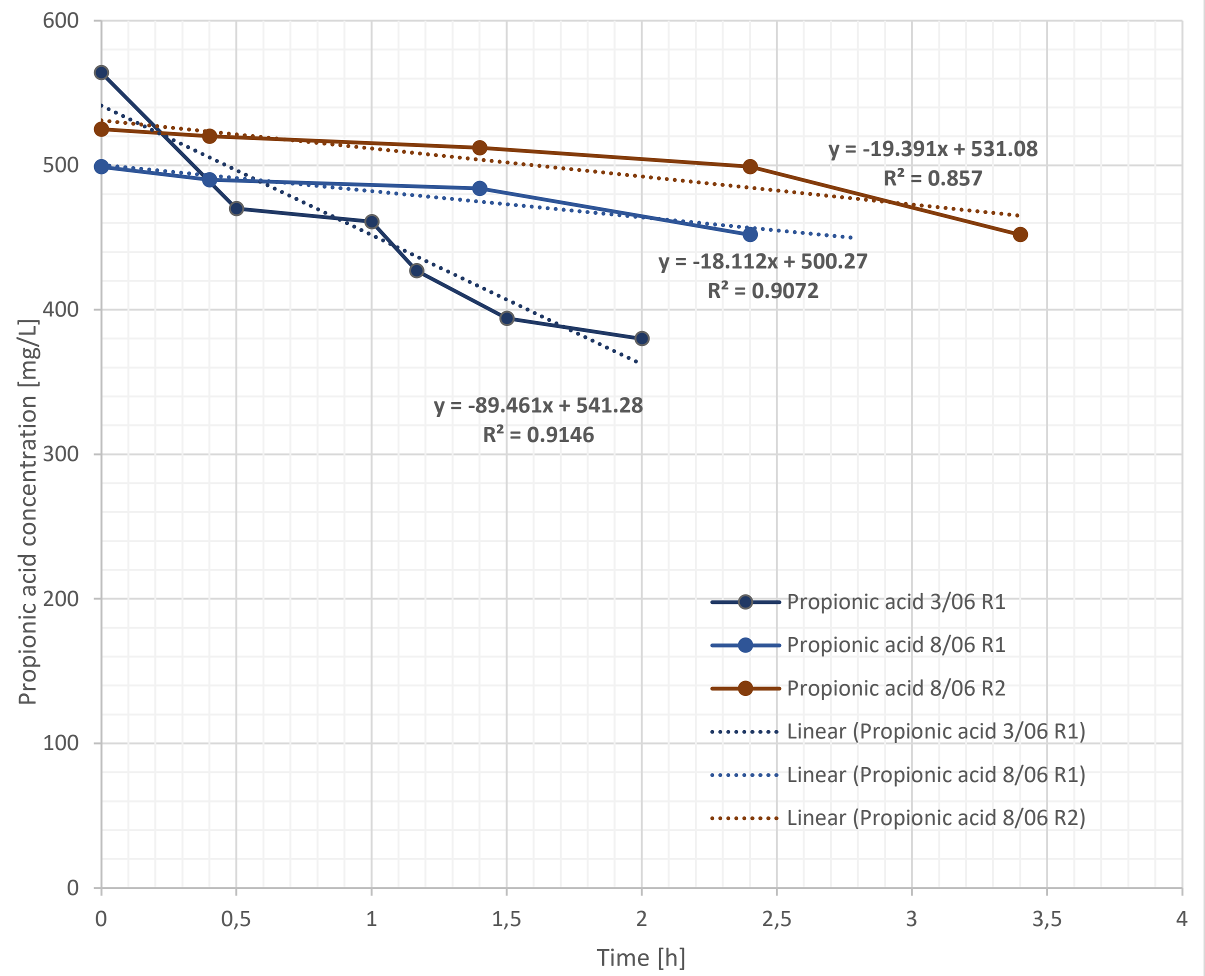

Figure 28 Diagram of evolution of the concentration of propionic acid for R1 and R2 for the dosing of the 03/06 and the 08/06

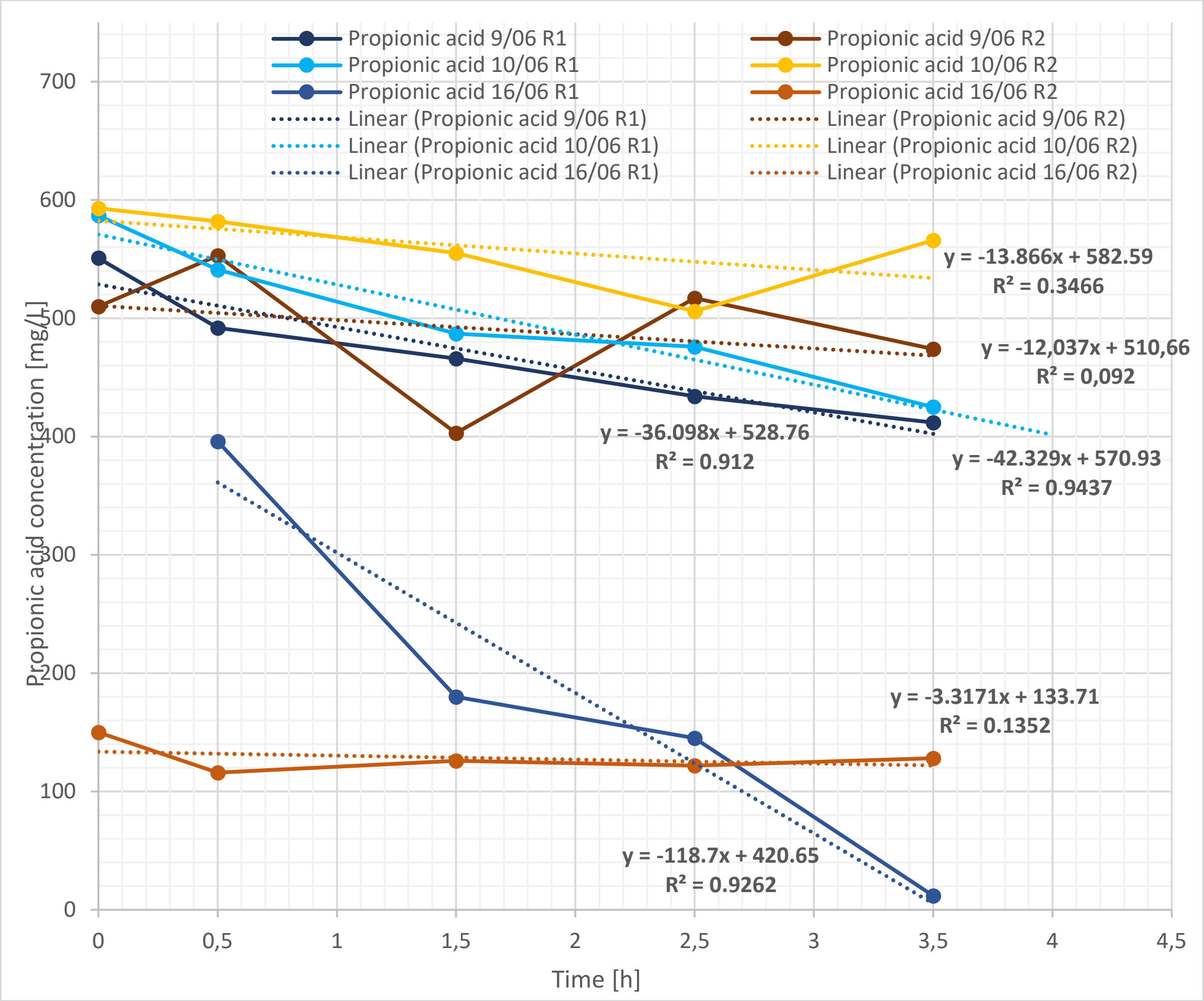

Figure 29 Diagram of evolution of the concentration of propionic acid for R1 and R2 for the dosing of the 09/06, the 10/06 and the 16/06

The degradation rates of propionic acid vary between 18 and 118 mg/(L · hour) for R1 and vary between 3 and 20 mg/(L · hour) for R2. These rates are summarized in the Table 16.

Table 16 Degradation's rate of R1 and R2 for the dosing with propionic acid

Date	Degradation Rate (mg/(L.h))		Degradation rate ratio	R^2	
	R1	R2	R2/R1	R1	R2
03/06/2020	**-87.224**	*****		**0.9146**	
08/06/2020	**-18.112**	**-19.391**	**107.06%**	**0.9072**	**0.857**
09/06/2020	**-36.098**	**-12.037**	**33.35%**	**0.912**	**0.092**
10/06/2020	**-42.329**	**-13.866**	**32.76%**	**0.9437**	**0.3466**
16/06/2020	**-118.7**	**-3.31**	**3%**	**0.9262**	**0.1352**
Average	**-53.80975**	**-12.151**	**22.58%**		

It can be observed that for all dosing, with the exception of the dosing of the 08/06, the degradation's rate of R2 is strongly lower than the rate of R1. The ratio between their rate vary from 3% to 33.35% (if the dosing of the 08/06 is not taking in count). This suggests that there is an inhibition of the degradation of propionic acid in R2. The results for the dosing of the 08/06 indicates that there was an inhibition of the propionic acid's degradation in R1 for this dosing since the value of the degradation's rate for R1 was rather low compare to the value of R1.

The value of R^2 was higher than 0.9 for each dosing in R1 which indicates that the linear model was adapted to describe the evolution of the propionic acid's concentration during the dosing in R1. For R2 the value varies between 0.092 and 0.857 which indicates that the linear model is not a good model to describe this evolution in R2.

In the Figure 30 are displayed the dosing of the 20/07 and the 22/07. During this dosing the amount of hydrogen introduced in R2 was only 0.1 atm (10% of the gaseous volume in R2).

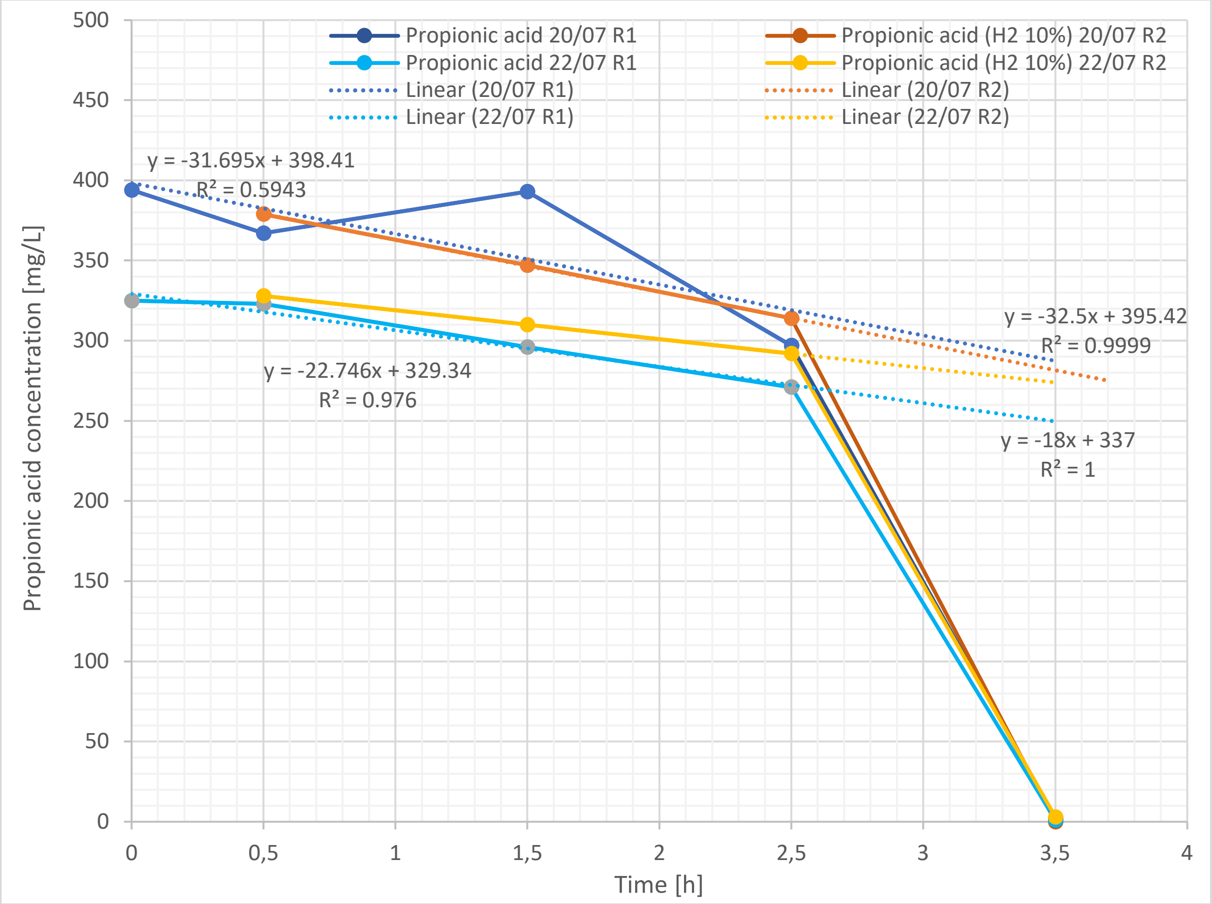

Figure 30 Diagram of evolution of the concentration of propionic acid for R1 and R2 for the dosing with 10% of hydrogen of the 20/07 and the 22/07

The first two point for the dosing of R2 are not displayed since they are much lower than the three next point. This is certainly due to some inhomogeneity in the reactor. The degradation's rates were calculated for the interval of time between 0 and 2.5 h and are summarized in the Table 17.

Table 17 Degradation's rate of R1 and R2 for the dosing with propionic acid and 10% of hydrogen

Date	Degradation rate [mg/(L.hour)]		Degradation rate ratio	R^2	
	R1	R2	R2/R1	R1	R2
20/07/2020	**31.695**	**32.5**	**102.54%**	**0.5943**	**0.999**
22/07/2020	**22.746**	**18**	**79.13%**	**0.976**	**1**
Average	**27.2205**	**25.25**	**92.76%**		

It can be observed that for each dosing the evolution of the propionic acid's concentration is separated in two steps. A first step from 0 to 2.5 hours where the degradation of the propionic acid is rather low. And a second step from 2.5 to 3.5 hours where the degradation is very quick and leads to the total elimination of propionic acid. This behavior seems to be the same for both reactors. And the degradation's rates of propionic acid are very close from one reactor to another as it is illustrated by the ratio R2/R1 (102.54% for the dosing of the 20/07 and 79.13% for the dosing of the 22/07). It contrasts with the behavior observed during the previous dosing where the degradation stayed the same all along the dosing. The causes for this original behavior are difficult to identify. However, it can be deduced from these results that the introduction of 0.1 atm of hydrogen doesn't induce an inhibition inside R2 (or just a slightly one).

3.3.2. Mix of propionic acid and sludge

In the Figure 31 is represented the evolution of the concentration of propionic acid for the dosing made during the 7/07, the 10/07 and the 13/07 with sludge and propionic acid. For the dosing of the 7/07 no hydrogen was injected inside R2 du to a technical issue. For each dosing 5.18 g of propionic acid was injected in both reactors with 600 g of raw sludge. And a linear model was used to simulate the evolution of the butyric acid's concentration for each dosing in both reactors.

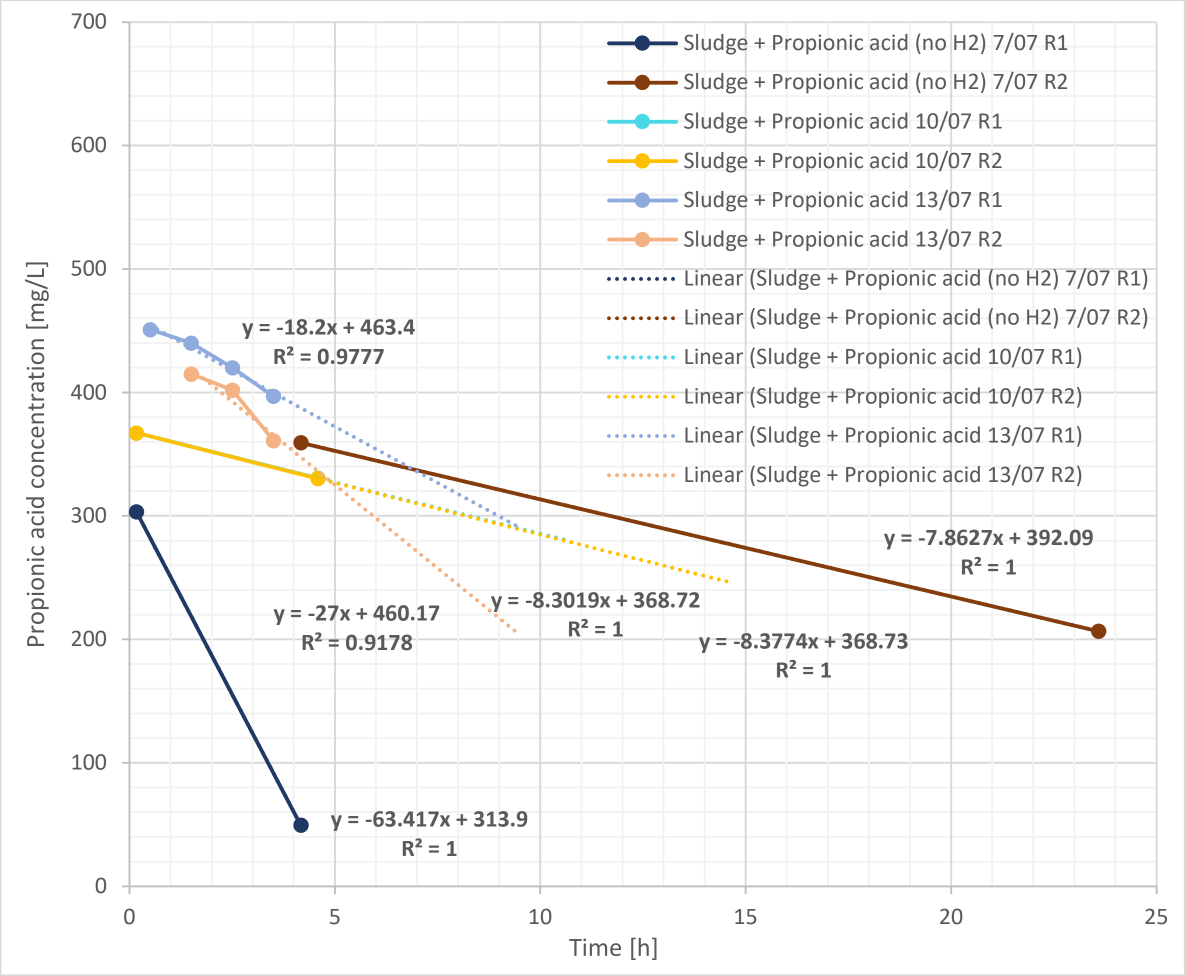

Figure 31 Diagram of evolution of the concentration of propionic acid for R1 and R2 for the dosing with raw sludge of the 07/07, the 10/07 and the 13/07

The degradation's rate of propionic acid vary from 8.3 to 27 mg/(L · hour). All the rates are summarized in the Table 18.

Table 18 Degradation's rate of R1 and R2 for the dosing with propionic acid and raw sludge

Date	Degradation rate [mg/(L · hour)]		Degradation rate ratio	R^2	
	R1	R2	R2/R1	R1	R2
10/07/2020	**-8.3**	**-8.38**	**100.96%**	**1**	**1**
13/07/2020	**-18.2**	**-27**	**148.35%**	**0.9777**	**0.9178**
Average	**-13.25**	**-17.69**	**133.51%**		
07/07/2020	**-63.417**	**-7.86**	**12.39%**	**1**	**1**

The results suggest that the dosing with propionic acid and sludge leads to an inhibition of the propionic acid degradation in both reactors. The values of R1 and R2 are indeed close or lower than the ones of R2 measured during the dosing with only propionic acid. This inhibition is stronger for R1 than R2 according to the ratio between their rate.

The results for the dosing of the 07/07 are however strange. As there was no hydrogen injected in R2 the two reactors functioned therefor under the same conditions. The measured rates should have been the same or at least close, which is not the case. So, concerning this case it is hard to come to a conclusion. A uncontrol parameter could be responsible for this result.

The value of R² is higher than 0.9 for both reactors for the dosing of the 13/07 which indicates that the linear model is adapted to describe the evolution of the concentration of propionic acid during this dosing for both reactors. For the others the value of 1 is due to the use of only two point to make the diagram.

3.3.3. Gas production

The Figure 32 and the Figure 33 shows the volume of the produced gas during the dosing with propionic acid made during the 8/06, the 9/06, the 10/06 and the 16/06. Each curve starts at the beginning of the dosing. Some curves present a form of oscillation, caused by drum gas counter. It affects only the curves of R2. At the end of some curves it can be observed a very strong rise of the gas production. This is due to the reintroduction of the sludge taken for the sampling.

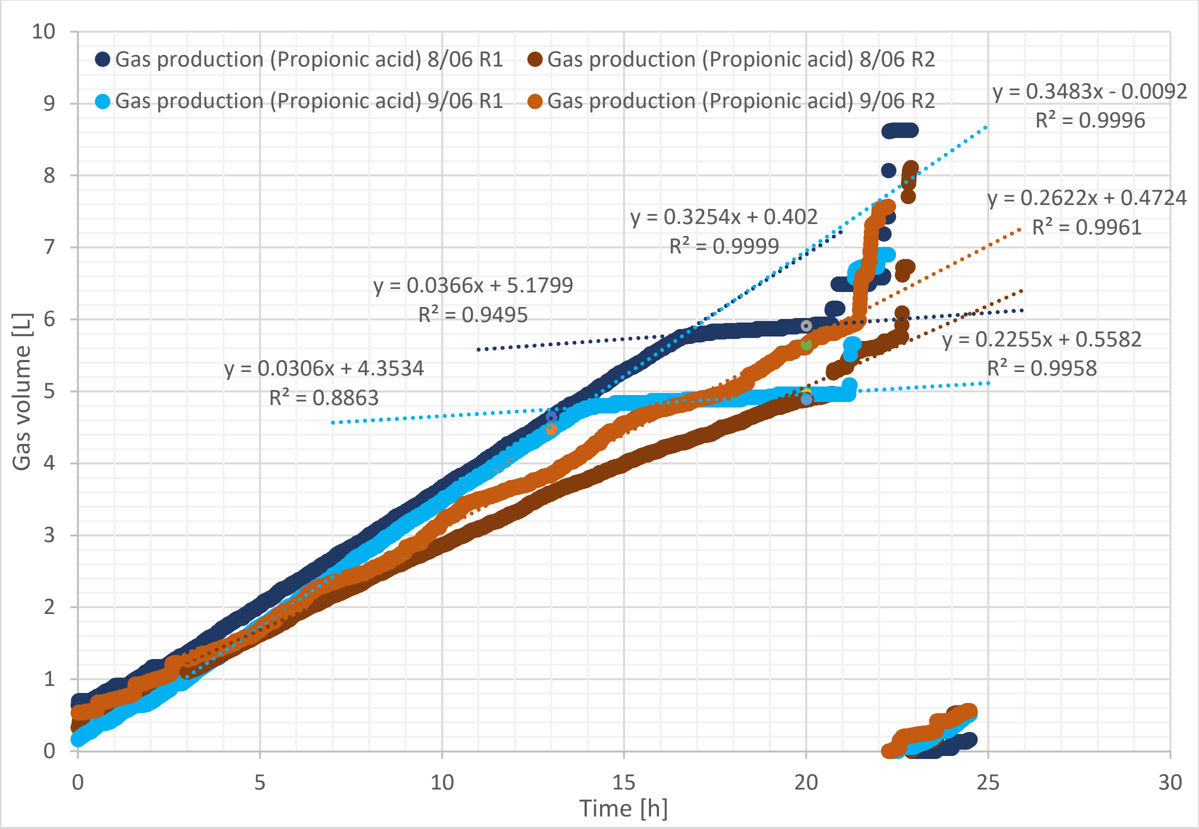

Figure 32 Diagram of evolution of the gas production for R1 and R2 for the dosing of the 08/06 and the 09/06

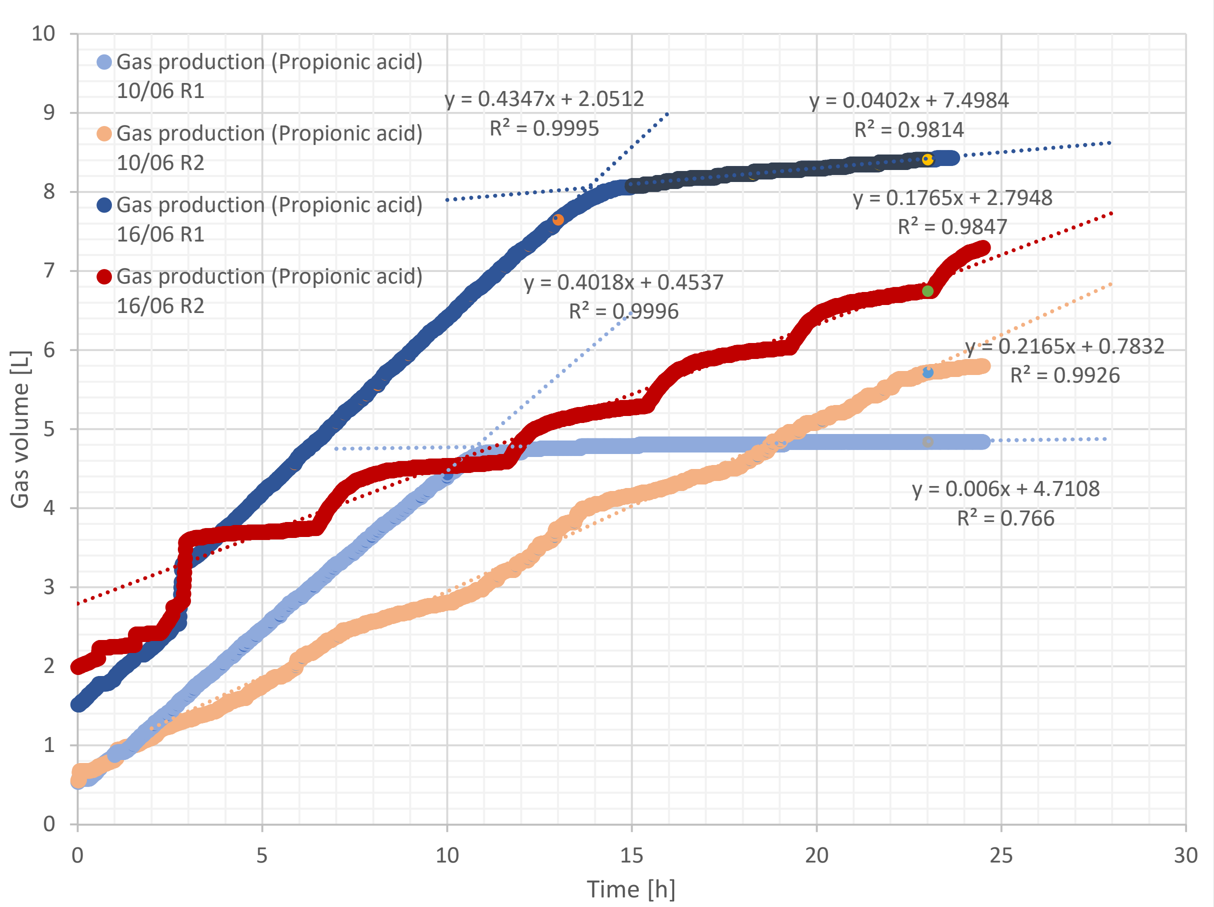

Figure 33 Diagram of evolution of the gas production for R1 and R2 for the dosing of the 10/06 and the 16/06

The gas production rates for both reactors and each dosing experiment are gathered in the Table 19. The ratio between the rates of R2 and the rates of R1 are also given.

Table 19 Gas production rates of R1 and R2 for the dosing with propionic acid

Date	Step	Production Rate (L/h)		Production (L)		Ratio R2/R1
		R1	R2	R1	R2	
08/06/2020	**1**	**0.3254**	**0.2255**	**5.06**	**4.71**	**69.30%**
	2	**0.0366**				
09/06/2020	**1**	**0.3483**	**0.2622**	**4.58**	**5.15**	**75.28%**
	2	**0.03306**				
10/06/2020	**1**	**0.4018**	**0.2165**	**4.15**	**4.55**	**53.88%**
	2	**0.006**				
16/06/2020	**1**	**0.4347**	**0.1765**	**5.68**	**4.5**	**40.60%**
	2	**0.0402**				

It can be observed that the behavior of the gas production is similar to the behavior of the gas production with acetic acid or butyric acid. For R1 the two-steps curve is present and for R2 the curves possess an homogenic gradient. The rate of R1 during the first step are also higher than the rate of R2. It is illustrated by the ratio between R2 and R1 whose value is between 40% and 76%. It can also be noticed than the total amounts of produced gas during the dosing are consistent with the theoretical value (5.20 L) calculated in the previous part, considering the yield of the methanogenic archaea. It is slightly lower than 5.20 L because of the biomass build up. For the dosing of the 16/06, the high volume of produced gas might be explained by the introduction of higher than expected quantity of acid.

The Figure 34 shows the gas production for the dosing of the 20/07 and of the 22/07. In those dosing, propionic acid was used but instead of injecting 15% of hydrogen into R2, it was 10% of hydrogen that were injected.

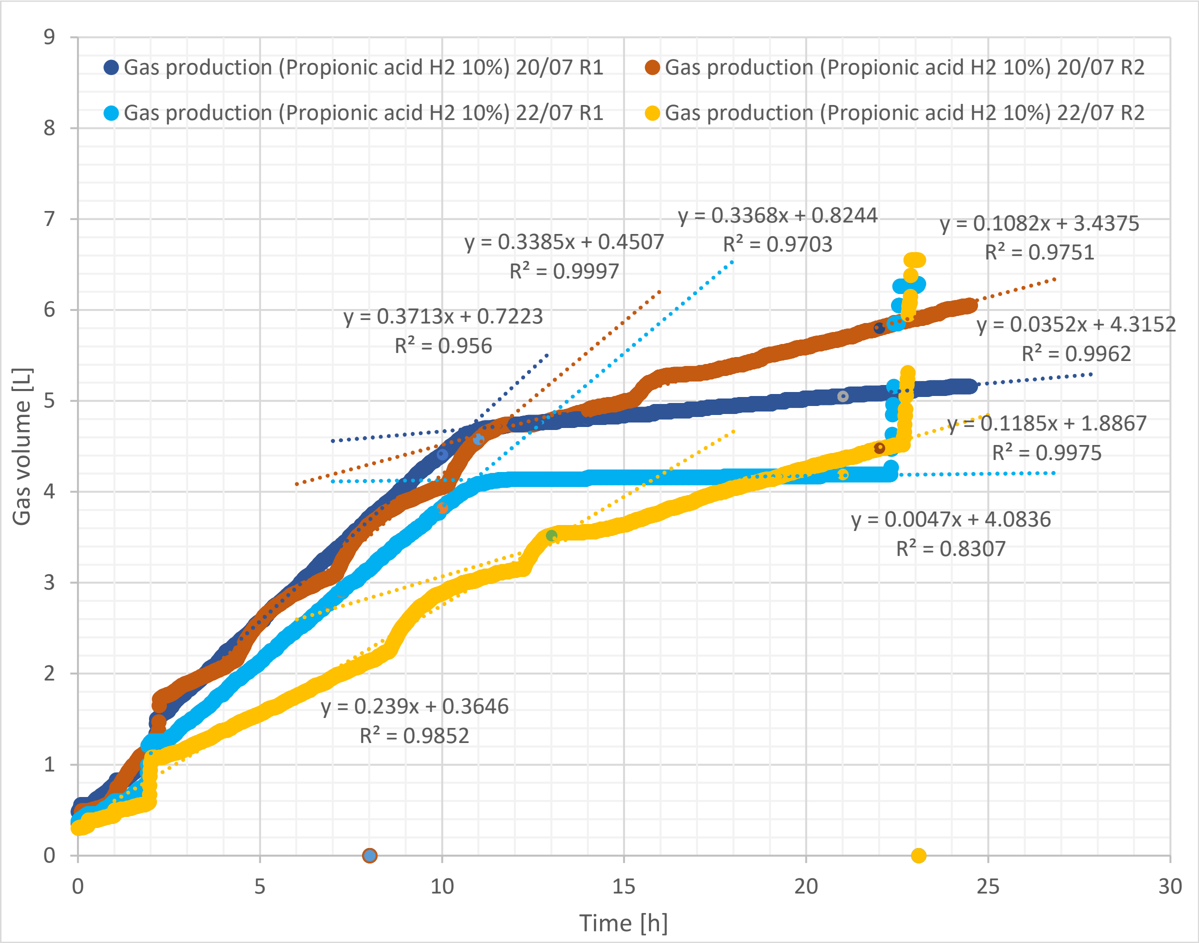

Figure 34 Diagram of evolution of the gas production for R1 and R2 for the dosing with 10% of hydrogen of the 20/07 and the 22/07

The gas production rates for both reactors and each dosing test are gathered in the Table 20. The ratio between the rates of R2 and the rates of R1 are also given.

Table 20 Gas production rates of R1 and R2 for the dosing with propionic acid and 10% of hydrogen

Date	Step	Production Rate (L/h)		Production (L)		Ratio R2/R1
		R1	R2	R1	R2	
20/07/2020	**1**	**0.3713**	**0.3385**	**4.47**	**5.25**	**91.17%**
	2	**0.0352**	**0.1082**			
22/07/2020	**1**	**0.3368**	**0.239**	**3.94**	**3.97**	**70.96%**
	2	**0.0047**	**0.1185**			

It can be observed that the curves for R1 are unchanged (as expected). However, the curves for R2 have a different aspect than at the dosing experiment with 15% hydrogen. They present a “two-step” structure like the curve of R1 but with a difference of the rate between the two steps less strong than for R1. For R1 the rate during the second step is nearly null. While for R2 the rate is only divided by 2 or 3 compare to the first step. For the dosing of the 20/07, the rates of R2 for the two steps are respectively 0.3385 L/h and 0.1082 L/h. And for the dosing test of the 22/07 they are respectively 0.239 L/h and 0.1185 L/h. The gaps between the rate of R1 and R2 during the first step are also smaller as it is illustrated with the ratio between R1 and R2 (91.17% and 70.96%). The total amounts of produced gas during the dosing tests are with approx. 4 L also consistent with the theoretical value (5.20 L) calculated in the previous part.

The Figure 35 shows the production of gas for the dosing with sludge and propionic acid made during the 7/07, the 10/07 and the 13/07.

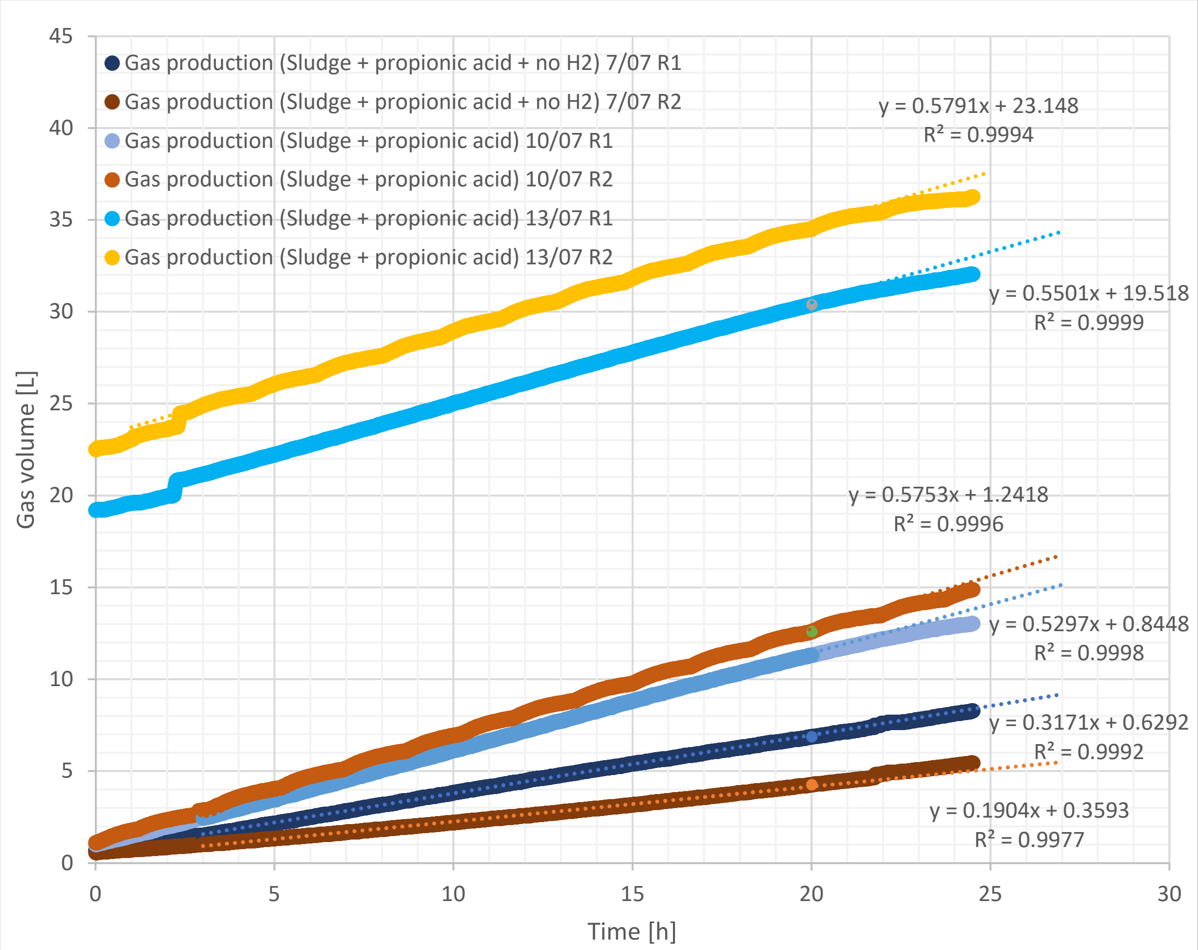

Figure 35 Diagram of evolution of the gas production for R1 and R2 for the dosing with raw sludge of the 07/07, the 10/07 and the 13/07

The gas production rates for both reactors and each dosing experiment are gathered in the Table 21. The ratio between the rates of R2 and the rates of R1 are also given.

Table 21 Gas production rates of R1 and R2 for the dosing with propionic acid and raw sludge

Date	Production Rate (L/h)		Production (L)		Ratio R2/R1
	R1	R2	R1	R2	
07/07/2020	**0.3171**	**0.1904**	**7.06**	**4.23**	**60.04%**
10/07/2020	**0.5297**	**0.5753**	**11.89**	**13.64**	**108.61%**
13/07/2020	**0.5501**	**0.5791**	**12.81**	**13.63**	**105.27%**

Like for the dosing tests performed with a mixture of raw sludge and acetic acid or raw sludge and butyric acid, the behavior of the gas productions for the dosing of the 10/07 and the 13/07 are similar from one reactor to another. The productions are linear and the curves are nearly parallel. The ratio between R1 and R2 are very close from 1. The volumes of produced gas are also superior to the volume produced with only propionic acid. The dosing of the 10/07 generated after 20 hours more than 10 L of gas and the dosing of the 13/07 generated after 20 hours more than 12 L for both reactors. While the dosing with only propionic acid didn't generate more than 7 L after 20 hours (with only the exception of the dosing of the 16/06 for R1). The introduction of raw sludge lead to the same behavior in both reactors in terms of gas production and increased the amount of produced gas.

The dosing of the 7/07 was made without hydrogen in R2 (due a technical problem). It can be observed that contrarily to the other dosing made with sludge and propionic acid, sludge and acetic acid or sludge and butyric acid, the production rate of R2 is lower than the production rate of R1 with a ratio of only 60% between them.

4. Conclusion

The power-to-gas concept is a promising method of storage of electricity produced through photovoltaic or wind. The organic matters used in digesters are sludges obtained from wastewater treatment plant. Those sludges naturally contain high quantity of organic acids such as acetic acid, propionic acid and butyric acid. In the scientific literature, several authors noticed that high level of acids with presence of hydrogen was associated with various behaviors and more specifically with inhibition's behavior.

The dosing experiments made with acetic acid indicated that the introduction of hydrogen lead to a very limited inhibition of the acetic acid's degradation and no inhibition at all. While the dosing experiment with propionic acid indicated a strong inhibition (70%) of the propionic acid degradation (ratio R2/R1 around 30%). For the dosing experiments with butyric acid, there are not enough valid data to draw a conclusion. From the few gathered data, it can be assumed that the degradation of the butyric acid has the same behavior as the one with propionic acid but with an inhibition less pronounced (ratio R2/R1 around 60%). The introduction of 0.1 atm in R2 during the dosing with propionic acid didn't induce a strong inhibition (ratio R2/R1 around 90%). Only a slight inhibition and an absence of inhibition were observed during these dosing experiments. In conclusion it can be deduced that only for acetic acid there no inhibition was found of the acetic acid degradation under a partial pressure of hydrogen of 0.15 atm. For propionic acid, there is a strong inhibition under a hydrogen partial pressure of 0.15 atm but this inhibition disappears for a partial pressure inferior to 0.10 atm. And for butyric acid there might be a moderate inhibition under a hydrogen partial pressure of 0.15 atm, although more data would be needed to prove this assumption.

The gas production during the dosing experiments with acetic acid, propionic acid and butyric acid was also monitored. The data obtained from this monitoring show for each reactor a distinct behavior. And these behaviors are the same for each acid. For R1 the gas production is separated in two steps: a first step where the gas production is very strong and higher than the gas production of R2, suspected of higher organic dry substance concentration in reactor 1. And a second step where the gas production is nearly reduced to zero, were the dosed organic acid is converted to biogas and just hard degradable organic matter from the raw sludge is degraded. While for R2 the gas production stays constant during the whole dosing experiment. The dosing experiment performed at 0.10 atm hydrogen partial pressure and propionic acid highlighted the fact that the behavior of the gas production could be pilot by changing the amount of introduced hydrogen. The obtained diagram shows that the behavior of R2 for this dosing is in an intermediary state between the behaviors previously described for R1 and R2. The production is clearly separated in two steps. A first step with a strong production of gas and a second one with a lower production. However, the gap of production between the two steps is not as pronounced as for R1. It is to notice that the amount of produced gas is consistent with the theoretical values calculated with the stoichiometric formula. From these results it can be deduced that the gas production is influenced by the amount of introduced hydrogen because of hydrogen short circuit flow into the product gas rather than the kind of introduced acid. When no hydrogen is introduced, the gas production has a “two-steps” behavior: a first step with a strong gas production according to zero-order reaction and a second step with a gas production nearly reduced to zero. When 0.15 atm of hydrogen is introduced, the gas production stays constant all along the dosing experiment. And for intermediary amount of introduced hydrogen, the behavior of the gas production tends to a “two-steps” behavior but less pronounced.

The dosing experiments made with a mixture of raw sludge and acid gave different results from an acid to another. The addition of sludge with acetic acid didn’t change the degradation behavior of acetic acid (no inhibition in R2 was observed). For butyric acid the addition of sludge resulted in an absence of inhibition of the butyric acid degradation in R2. For propionic acid it can be concluded that the degradation rate of R1 was lower than R2 which suggested that it induced an inhibition in R1 that was stronger than in R2, probably caused by too high propionic acid concentrations, which resulted in an inhibition.

The addition of raw sludge impacted the gas production of the reactors as expected and the effects were the same for each acid. The addition of raw sludge resulted for both acids in a constant production of gas for both reactors nearly all along the dosing. R2 had every time a higher production rate than R1. The only exception was the dosing made with no hydrogen where the production rates were much lower than the others. This result is interesting and could be explain by the fact that the introduction of hydrogen influenced the population of micro-organisms in R2. As a consequence, the population of R2 is less efficient to produce gas without addition of hydrogen than the population of R1. Which leads to these different outcomes between R1 and R2 although the parameters were the same for R1 and R2. And for both reactors the total amount of produced gas during the dosing was higher than the amount obtained during the dosing with pure acid which was expected as the sludge was a supplementary source of organic matter.

5. Summary

The biological methanation is a method of transformation of CO_2 and H_2 into CH_4. It is used in the power-to-gas concept to convert fluctuating electricity produced by wind and solar energy into methane. The surpluses are used to produce hydrogen through hydrolysis of water which is then convert into methane gas, which is easier to store and used through the distribution networks of the natural gas.

Anaerobic digestion is a technology of treatment of sewage sludge which uses the microorganism contained in the sludge to degrade the organic matter in it and convert it into biogas. Six different conversion processes were identified by Gujer and Zehnder (1983) during this digestion. Among them there is the conversion process of the organic acids into acetic acid, CO_2 and H_2. There are also the methanogenesis from acetic acid and the methanogenesis from H_2 and CO_2.

Raw sludge from a municipal wastewater treatment plant was used as substrate and source of CO_2. Two reactors of 14 L were used during this thesis. One reactor (R1) served as reference with no hydrogen injection while the other (R2) had hydrogen injection.

The raw sludge that were used during the project contained high amounts of organic acids such as acetic acid, propionic acid and butyric acid. Several authors have reported phenomenon of inhibition of organic acids in digester under high partial pressure (Fukuzaki et al., 1990; Luo et al., 2011; Luo et al., 2013). If they occur, they can lead to instability in the reactor that can result in a shutdown of the methane production. In the case of a full scale methanation plant, it would be disastrous and in the worst case could make the methanation plant inoperable for a certain amount of time. This is why it was chosen to investigate the kinetic behaviour of degradation of acetic acid, propionic acid and butyric acid during in-situ methanation under high concentrations of hydrogen and their effect on the gas production.

The hydraulic retention time was set to 50 days. Every day before dosing or feeding samples were taken to analyze the composition of the sludge inside both reactors. The gas composition, the temperature and the pH were also measured manually. Online measures were performed continuously to monitor the gas composition, the temperature, the pH and the gas production of the reactors. During the dosing probes were regularly made. They were centrifuged and then analyze with a HPLC chromatogram to measure the concentration of organic acid. Acetic acid, butyric acid and propionic acid a were used for the dosing experiments.

Dosing experiments with acetic acid showed no inhibition of the acetic acid degradation under a partial pressure of hydrogen of 0.15 atm. For propionic acid, there was a strong inhibition (around 70%) under a hydrogen partial pressure of 0.15 atm but this inhibition disappeared for a partial pressure inferior to 0.10 atm. For butyric acid there were not enough data to draw a solid conclusion. However, from the few gathered data it can be assumed that a moderate inhibition (around 40%) occurs under a hydrogen partial pressure of 0.15 atm. More data would be needed to verify this hypothesis.

The gas production during the dosing with acetic acid, propionic acid and butyric acid was also monitored. First, it was noticed that the amount of produced gas was consistent with the theoretical values calculated with the stoichiometric formula. It also revealed that each reactor had its own behavior. But this behavior didn't vary from one acid to another. The production of R1 was characterized by a "two-step" production: a first step with a high production rate and a second step with a production rate nearly reduced to zero, when the dosed acid was converted into gas. While the production of R2 was characterized by a constant production rate. The dosing made with only 0.10 atm hydrogen partial pressure highlighted the fact that if the amount of introduced hydrogen is reduced, R2 tends to have an intermediary behavior. It has a "two-step" production like R1 but with a gap of production between the two steps less pronounced.

The dosing experiments made with a mixture of raw sludge and acids gave different results from an acid to another. The addition of sludge with acetic acid didn't change the degradation's behavior of acetic acid (no inhibition was observed). For butyric acid the addition of sludge resulted in an absence of inhibition of the butyric acid degradation. And for the propionic acid the results were that the degradation's rate of R1 were lower than R2 which suggested that it induced an inhibition in R1 that was stronger than in R2. Concerning the gas production, the results were the same for every reactor and for each acid. The gas production was constant. The production rate of R2 was systematically higher than R1 with the exception of the dosing made with no hydrogen where the production rates were much lower than the others. The addition of hydrogen in R2 might explain this systematic higher production rate. And for both reactors, the total amount of produced gas during the dosing was higher than the amount obtained during the dosing with pure acid which was expected as the sludge was a supplementary source of organic matters.

These results obtained for the propionic acid match well with the results reported by Fukuzaki (1990). The expected inhibition of the propionic acid degradation under high hydrogen concentration occurred. And for the dosing with raw sludge, the inhibition in R1 correspond to an inhibition caused by high concentration of propionic acid as described by Fukuzaki. However, the fact that this inhibition was stronger in R1 than in R2 for this dosing was surprising. The results obtained for butyric acid were more unexpected. As Luo et. al (2013a) underlined an inhibition of butyric acid degradation in a batch fed with manure would occur under high concentration of hydrogen (1 atm) only with a high mixing intensity (mixing speed of 300 rpm). While the mixing intensity of R1 and R2 in this study was low (11 rpm) and the hydrogen concentration in R2 was only 0.15 atm, the inhibition of the butyric acid degradation occurred in R2. But as not enough data were collected, it's not possible to be categorical about it. For acetic acid, the obtained results correspond to what was expected: no inhibition occurs even with addition of hydrogen. The part made with dosing with a mixture of raw sludge and acid was unprecedented in this domain. No article or reference were found mentioning dosing with organic acid and raw sludge from waste water treatment plants. The data gathered from these experiments and the gas production monitoring enhanced the knowledge about the degradation mechanism of organic acid in a digester.

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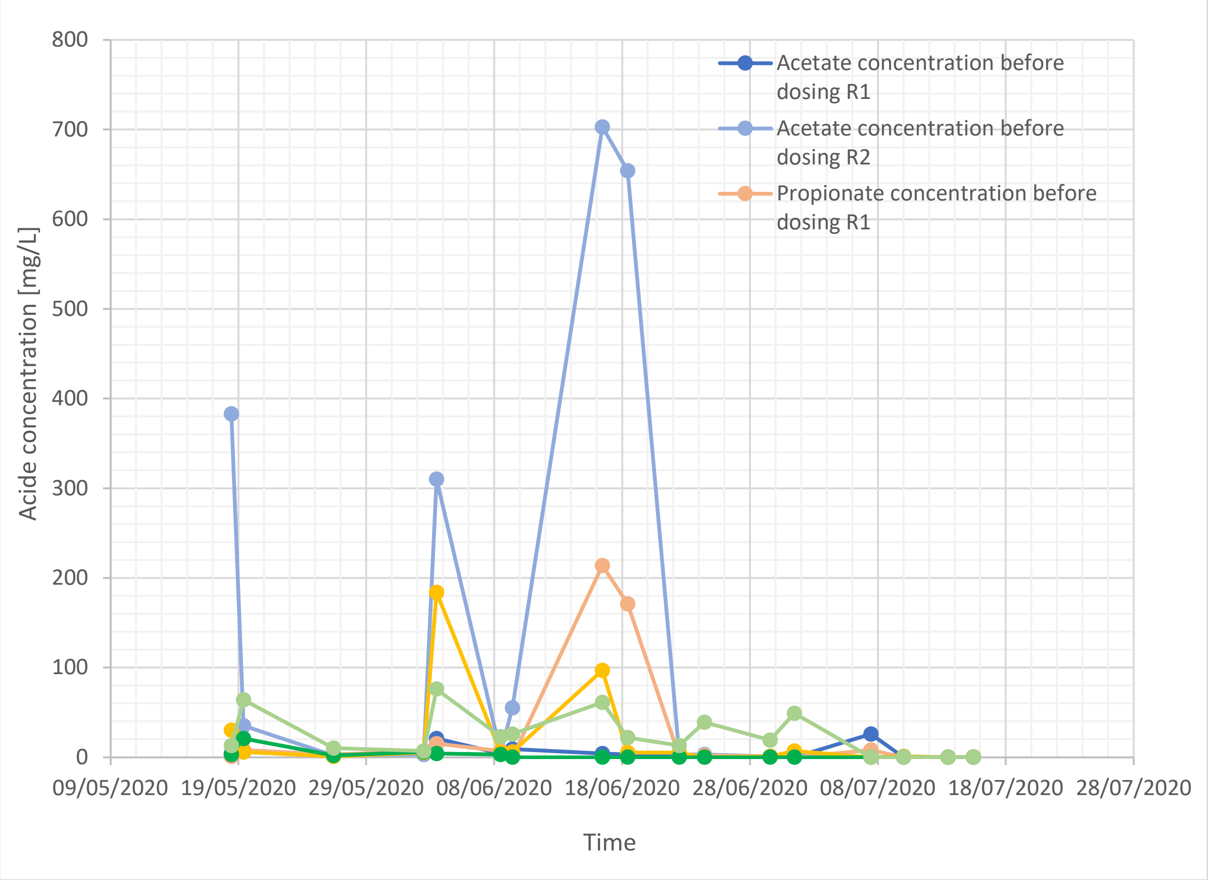
7. Appendix

7.1. Acid concentration in R1 and R2 before dosing



7.2. Acid composition of the two batch of sludge used during the thesis

	Date	Acetic acid	Propionic acid	Butyric acid
		mg/L	mg/L	mg/L
Old raw sludge	**07/07/2020 10:49**	**1309**	**1200**	**478**
New raw sludge	**07/07/2020 10:49**	**774**	**580**	**182**
New raw sludge	**10/07/2020 00:00**	**835**	**632**	**231**

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9. Affirmation

I certify that the master thesis was written by me, not using sources and tools other than quoted and without use of any other illegitimate support.

Furthermore, I confirm that I have not submitted this master thesis either nationally or internationally in any form.

Vienna, the 28th September 2020, Julien Audouin