UNIVERSIDAD DE VALLADOLID

ESCUELA DE INGENIERIAS INDUSTRIALES

Grado en Ingeniería Química

# Estudio de casos fronterizos en una planta anaerobia de tratamiento de aguas residuales con diferentes escenarios de aguas residuales de baja resistencia.

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TÍTULO: Experimental study of border line cases in an anaerobic wastewater treatment plant with different low-strength wastewater scenarios.

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## Resumen y palabras clave

En este trabajo de fin de grado se han estudiado distintos casos en una planta anaerobia de tratamiento de aguas residuales con diferentes scenarios de aguas residuales de baja resistencia. Las condiciones de trabajo son anaerobias y de una etapa. Los parámetros analizados son la demanda química de oxígeno y la concentración de amonio. Sacarosa y peptona son los sustratos añadidos al agua residual en distintas concentraciones para estudiar 12 casos distintos. Se observó que la adición de 100 gramos de sustrato causaba un incremento de 100 mg/l en los niveles de DQO. La adición de sacarosa causa una degradación mayor de materia orgánica en comparación a la adición de peptona. Con una vision final del estudio se puede decir que la reducción total de DQO excede el 70% pero los límites de DQO de 75 mg/l en agua de servicio no fueron alcanzados con los tiempos de retención establecidos.

**Palabras clave:** Anaerobio, agua, residual, amonio, DQO.

## Abstract and keywords

In this bachelor thesis a decentralized pilot plant is constructed to study border line cases with different low strength wastewater scenarios in order to produce service water. The pilot plant works under anaerobic conditions with one-step process. The parameters that have been taken into account to analyze the treated wastewater are the chemical oxygen demand and the ammonium concentration. Saccharose and peptone are subtracts that were added to the wastewater in different concentrations to study 12 batches. It was observed that each 100 grams of addition of subtract the COD level increased 100 mg/l. The addition of saccharose causes greater degradation of the organic matter in comparison with the addition of peptone. With a final view of the study it can be said that although the total reduction of COD exceeds 70%, the COD limits of 75 mg/l in service water was not reached with the established retention times.

**Keywords:** Anaerobic, digestión, wastewater, ammonium, COD.

# EXPERIMENTAL STUDY OF BORDER LINE CASES IN AN ANAEROBIC WASTEWATER TREATMENT PLANT WITH DIFFERENT LOW-STRENGTH WASTEWATER SCENARIOS

Bachelor Thesis

Friedrich-Alexander University Erlangen-Nuremberg

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## Declaration of authorship

I hereby certify that this thesis has been composed by me and is based on my own work, unless stated otherwise. No other person's work has been used without due acknowledgement in this thesis. All references and verbatim extracts have been quoted, and all sources of information, including graphs and data sheets, have been specifically acknowledged.

Erlangen, 08.08.2017

Andrea Barcenilla Canduela

# Content

# Acknowledgements

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# Abstract

In this bachelor thesis a decentralized pilot plant is constructed in the municipal wastewater treatment plant of Erlangen to study border line cases with different low strength wastewater scenarios in order to produce service water. The pilot plant works under anaerobic conditions with one-step process and it consists of a buffer tank and three consecutive reactors. In this study the parameters that have been taken into account to analyze the treated wastewater are the chemical oxygen demand and the ammonium concentration. Saccharose and peptone are subtracts that were added to the wastewater in different concentrations with the aim of studding a series of 12 batches with different compositions. It was observed that each 100 grams of addition of subtract the COD level increased 100 mg/l. Each batch passed through the different stages of the plant and samples were collected every 24 hours to study the evolution of the digestion process. In all batches, the behaviour was the same. With the addition of subtract, initially there was an increase in the value of COD and $NH_4$ but afterwards these levels decreased gradually. With the obtained results different graphs were studied and it was observed that in reactor one most of the degradation of organic matter took place and reactor two continued with the digestion of the no digested subtract. With a final view of the study it can be said that although the total reduction of COD exceeds 70%, the COD limits of 75 mg/l in service water was not reached with the established retention times. It should also be mention that the addition of saccharose causes greater degradation of the organic matter in comparison with the addition of peptone.

# 1. Introduction

Pure water is essential for all living beings. It is an exhaustible source that has to be recycled. One possibility to recycle water is to produce service water from waste water treating it in a degradation process. For example for irrigation of gardens, it is sufficient to use service water instead of fresh water. Also for clothes washing and toilet flushing service water can be used. One consequence for the use of service water is the implementation of short water transport ways, that means decentralized systems shell be used instead of centralized ones. Anaerobic processes provide several advantages for decentralized wastewater treatment. These include the less aeration, because no oxygen is needed, as well as the space because this process doesn't need a huge amount of space. The production of sludges is much less in comparison to aerobic process and also methane which a valuable product is produced so it can be used as power source to obtain thermal or electrical energy. The most important controlling factors of this process are pH, temperature, HRT (hydraulic retention time), substrate composition and substrate concentration so this control can be reached with different sensors and an automated control. Other advantages are the low operating cost and the small amount of space needed. All this advantages can lead to a satisfactory, simple and useful process which can be installed in many buildings and thus be able to help our planet by saving a lot of pure water. All these advantages are the reason for the intention to use an anaerobic process of domestic wastewater treatment. Nevertheless, there are also some disadvantages of this process which can be: no complete degradation of waste water so it can only be used as service water, an additional step is needed to eliminate nitrogen, and this step is called Anaerobic Ammonium Oxidation (Anammox process). Other disadvantage is that anaerobic wastewater plants are sensitive for changes in concentration, composition and temperature of the water (Lorenzo Acosta, 2005). A particular feature of anaerobic microorganisms is their low rate of growth; therefore, a period of time that may vary between 30 and 180 days depending on the quality and quantity of the inoculum used is required at the start of the process (Weiland & Rozzi, 1991).

Therefore the institute of Fluid Mechanics of Friedrich-Alexander University of Erlangen-Nürnberg constructed a pilot plant for the decentral treatment of domestic wastewater up to service waste quality in the municipal wastewater treatment plant of Erlangen. The process is based on anaerobic biological treatment combined with post treatment. For a decentral application easy to use and space efficient design are important design criteria.

In order to have a good behaviour of the plant, temperature in the reactors has to be controlled under certain levels to prevent the death of the microorganisms inside the reactors.

# 2. Objective

In this Bachelor Thesis, a decentralized pilot plant is presented for the study of the treatment of municipal waste water to produce service water and thus save pure one. With this study it is wanted to make easy to operate and fully automated plants in different buildings in order to produce service water.

The objective is to study border line cases with different low strength wastewater scenarios. It is important to test different scenarios because municipal wastewater has not always the same composition, each day the weather conditions or the wastewater produced in buildings change. For this reason it is needed a huge variety of studied sewage waters and different quantities of peptone and saccharose have to be added to the water to observe the parameter's evolution along the different steps of the pilot plant which are: buffer tank, reactor 1, reactor 2 and reactor 3.

The aim of the analyses consists basically in proving the reduction of both ammonia and COD concentration. In order to study the degradation process in dependency of the wastewater scenarios different parameters such as chemical oxygen demand (COD) and ammonium concentration have to be determined. For these measurements samples have to be taken from each reactor every 24 hours. To older, pH, temperature and oxygen dissolved have to be measured in each collected sample to study the relation between the parameter's concentration and the reactor's behaviour.

# 3. State of the art

## 3.1 Municipal wastewater

Wastewater is a liquid waste collected in the sewage system to send it to the municipal wastewater treatment plant. Municipal wastewater can be classified in three different types according to the collected sewage system (Metcalf, 1991) (Mujeriego, 1990):

- Domestic wastewater: Are the wastewater produced by human activities related to the consumption of drinking water: washing dishes, showers, lavatories, sanitary services… It is produced in houses, public buildings and similar which have used supply sources for different uses.
- Industrial wastewater: Are waters that have been used in industrial processes and that have received contaminating by-products as a result of that use. Their quality is highly variable and a particular study for each industry is practically required.

- Infiltrations and uncontrolled connections: Wastewater that access the sewerage network through the registration wells or other devices, as well as the groundwater that infiltrates the sewerage system through improperly made or deteriorated connections.

The principal compounds in the municipal wastewater are (Espigares García & Pérez López, 1985):

- Proteins (40-60%)
- Carbohydrates (25-50%)
- Fats (10-15%)

In municipal wastewater, urea and ammonia are the major sources of nitrogen, along with proteins. Organic matter can also contribute in sulfur, iron and phosphorus. Most of the amino acids presented in nature can be detected in the wastewater as a result of protein breakdown. Other important compounds are sugars such as glucose, lactose, sucrose, fructose and galactose. Acids such as acetic, propionic, lactic, butyric and citric acids are found in wastewater. Fats are broken down more slowly by bacteria. They float because of their low density and this interferes in the treatment process favoring the anaerobic environment but causes a slower degradation and that is why bad odors are released. Nitrogen is present in wastewater in the form of urea and proteins, but these compounds are easily degradable by bacteria, which transform them into ammonia, and from it produce nitrites and nitrates. Ammonia is the first product of degradation of urea and protein material. Conversions between the various forms of nitrogen are influenced by the pH and temperature of the medium. Nitrites are often considered as indirect indicators of fecal contamination. They are unstable and readily oxidize to nitrates. Their absence in the medium may be due to anoxic conditions, which oblige the microorganisms to use bound oxygen, bringing the hydrogen into smaller forms (Espigares García & Pérez López, 1985).

Typical concentrations and compound in municipal wastewater are summarized in Table 1. The wastewater is divided in columns with the different concentrations (high, medium and low). Some of the main compounds presented in wastewater are solid matter (organic or inorganic), total organic carbon, total nitrogen, phosphorus, chlorides and fats among others. In the case of solid matter it can be presented in the wastewater as suspension or completely dissolved. The concentration of suspension solid matter is at least 2 times less than dissolved solid matter (for example 850 mg/l of dissolved solid matter in high concentration against 350 mg/l of suspension one in high concentration wastewater). The chemical oxygen demand (COD) varies from 250 mg/l in low wastewater concentration until 1000 mg/l in high wastewater concentration. The total nitrogen can be presented in different compounds such as ammonia or organic ones being the concentration of the first ones higher than the second ones. Total phosphorus compounds are also presented in the wastewater but in less concentration (15 mg/l, 8 mg/l and 4 mg/l for high, medium and low concentration wastewater respectively). These data are intended only as a guide and not as a project basis as concentrations vary with the time of day of the week, month of the year and other local conditions.

Table 1. Composition of a typical municipal wastewater (Metcalf, 1991).

| **Compound** | **Concentration (mg/l)** | | |
|---|---|---|---|
| | **High** | **Medium** | **Low** |
| Solid matter | 1200 | 720 | 350 |
| Totally dissolved | 850 | 500 | 250 |
| Inorganic | 525 | 300 | 145 |
| Organic | 325 | 200 | 105 |
| In suspension | 350 | 220 | 100 |
| Inorganic | 75 | 55 | 20 |
| Organic | 275 | 165 | 80 |
| Decantables solids | 20 | 10 | 5 |
| BOD | 400 | 220 | 110 |
| Total organic carbon | 290 | 160 | 80 |
| COD | 1000 | 500 | 250 |
| Total nitrogen | 85 | 40 | 20 |
| organic | 35 | 15 | 8 |
| ammonia | 50 | 25 | 12 |
| nitrites | 0 | 0 | 0 |
| nitrates | 0 | 0 | 0 |
| Total phosphorus | 15 | 8 | 4 |
| organic | 5 | 3 | 1 |
| inorganic | 10 | 5 | 3 |
| Chlorides | 100 | 50 | 30 |
| Alkalinity | 200 | 100 | 50 |
| Fat | 150 | 100 | 50 |

## 3.2 Wastewater treatment

The considered pilot plant is an anaerobic wastewater treatment plant with three successive reactors, one in each step. In the first two steps, the aim is to degrade de organic matter of the wastewater whereas the aim of the third reactor is to remove nitrogen.

### 3.2.1 Degradation process.

Anaerobic digestion can be defined as the process of conversion of organic matter into biogas (carbon dioxide and methane) and sludge by using bacteria without any presence of oxygen. Depending on the digester used and the parameters of its operation, the efficiency and stability of the process can vary significantly. Further important factors of this process are wastewater type, its temperature, retention time (HRT), pH, and bacteria in the reactor. To optimize the process, it is necessary to work in the optimal range of parameters (Karena Ostrem, Millrath, & Nickolas Themelis, 2004)

Wastewater can be seen as a valuable resource to provide energy if it is managed in an appropriate way. Organic matter presented in wastewater, can be decomposed by specialized bacteria to produce biogas, this product consists of 50-60% methane which can be combusted in a unit of cogeneration to produce energy. Also, the volume of excess sludge produced is reduced in compare of the aerobic digestion (S.Siddhart, 2006)

According to the author (S.Siddhart, 2006) the anaerobic process is divided in four successive steps which are describe below: Hydrolysis, acidogenesis or acidification, acetogenesis and methanogenesis. An additional step is needed to degrade the nitrogen of ammonium, Anammox step.

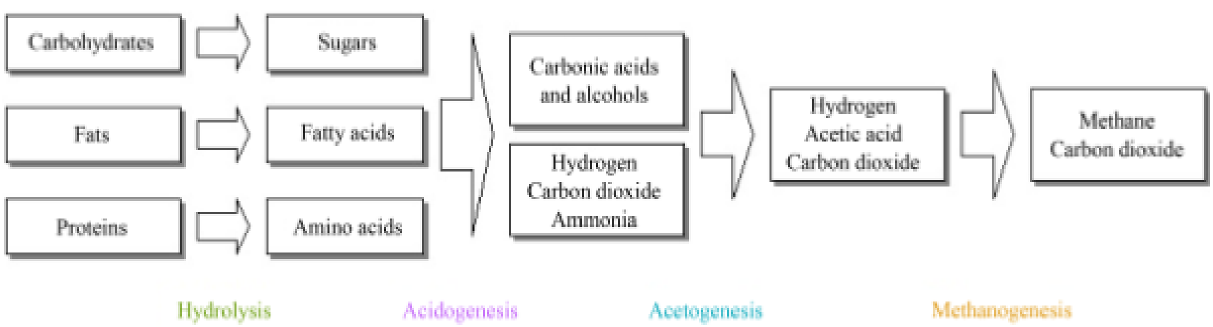



Figure 1. Anaerobic digestion steps (Kayode Feyisetan & Jude Awele, 2015).

Hydrolysis:

In the first step, hydrolysis, long chain molecules are degraded by the excretion of enzymes to soluble monomers, mainly monosaccharides, such as amino acids, fatty acids or simple sugars.

Hydrolysis reaction can be represented by the following:

$$C_6H_{10}O_4 + 2\,H_2O \rightarrow C_6H_{12}O_6 + 2\,H_2 \qquad [1]$$

Acidogenesis:

In this step, the products of the hydrolysis phase are the substrates for this process. Acidogenic bacteria produce simple and short chain organic compounds. Most of these products are fatty acids like butyric acid, propionic acid, lactic acid and acetic acid aside from alcohols such as ethanol and gases like $CO_2$ and $H_2$. In consequence to the formation of these organic acids the pH is decreasing. (Karena Ostrem, Millrath, & Nickolas Themelis, 2004)

Both hydrolysis and acidogenesis are usually made by the same microorganism that's why often these two steps occur simultaneously.

The oxidation reactions of volatile fatty acids of long and short chain produce the most quantity of hydrogen. In this step hydrogen ions ($H^+$) are reduced because they behave as electron acceptors. There is no significant decrease in COD due to that the original organic matter is in other organic compounds. (Lastra Bravo, 2013) (S.Siddhart, 2006).

Typical reactions of this stage are the followings:

$$C_6H_{12}O_6 \leftrightarrow 2\,CH_3CH_2OH + 2\,CO_2 \qquad [2]$$

$$C_6H_{12}O_6 + 2\,H_2 \leftrightarrow 2\,CH_3CH_2COOH + 2\,H_2O \qquad [3]$$

Acetogenesis:

During this phase single acids are formed through carbohydrate fermentation. Methane can be produced easily from acetic, $CO_2$ and $H_2$ using methanogenic bacteria, but ethanol, propionic acid, butyric acid and lactic acid from the phase of acidogenesis have to be converted into acetic acid to produce afterwards methane. For this process acetagenic bacteria are necessary.

Hydrogen plays an important intermediary role in this process, as the reaction will only occur if the hydrogen partial pressure is low enough (maximum $10^{-4}$ bar) to thermodynamically allow the conversion of all the acids. Symbiosis occur due to $pH_2$

(partial pressure of $H_2$) balance, acetogenic bacteria produce $H_2$ and methanogenic bacteria consume it. Thus the hydrogen concentration of a digester is an indicator of its health. (S.Siddhart, 2006).

The result is a combination of $H_2$, $CO_2$ and acetate as main product.

Some important reactions in this step are:

$$CH_3CH_2COO^- + 3\,H_2O \leftrightarrow CH_3COO^- + H^+ + HCO_3{}^- + 3\,H_2 \quad [4]$$

$$C_6H_{12}O_6 + 2\,H_2O \leftrightarrow 2\,CH_3COOH + 2\,CO_2 + 4\,H_2 \quad [5]$$

$$C_6H_{10}O_4 + 2\,H_2O \rightarrow C_6H_{12}O_6 + 2\,H_2 \quad [6]$$

$$CH_3CH_2OH + 2\,H_2O \leftrightarrow CH_3COO^- + H^+ + 2\,H_2 \quad [7]$$

$$2\,HCO_3{}^- + 4\,H_2 \rightarrow CH_3COO^- + 4\,H_2O \quad [8]$$

<u>Methanogenesis</u>:

Also named methane fermentation, in this stage methane is formed from acetic acid and hydrogen through the use of methanogenic bacteria. About two thirds of methane comes from acetate and the other one thirds is because the $CO_2$ reduction by hydrogen.

Acetogenic bacteria and methanogenic bacteria live in symbiosis because the growth of the first ones depends on the removal of the fermentative products. These products are used by methanogenic bacteria to produce methane from hydrogen and acetic acid.

The pH in this process should exceed pH 6 because in a range below methanogenic bacteria cannot survive. (S.Siddhart, 2006).

The reactions that take place in this step are:

$$CH_3COOH \rightarrow CH_4 + CO_2 \quad [9]$$

$$4\,H_2 + CO_2 \rightarrow CH_4 + 2\,H_2O \quad [10]$$

Anammox step:

According to the author (Dapena Mora, 2007), the anaerobic Ammonium Oxidation is a necessary back stage to degrade nitrogen present in some nitrogen-containing compounds of wastewater like ammonium ($NH_4^+$), proteins and urea.

The stoichiometric reaction of the Anammox process is the following one (Strous & Heijnen, 1998):

$$NH_4 + 1{,}31\, NO_2{}^- + 0{,}066\, HCO_3{}^- + 0{,}13\, H^+ \rightarrow N_2 + 0{,}26\, NO_3{}^- + 0{,}066\, CH_2O_{0,5}N_{0,15} + 2\, H_2O \quad [\,11\,]$$

As can be seen in this equation, the molar ratio between nitrite:ammonium has to be 1.31 moles of NO2- per mole of NH4+. To get this, a previous step is necessary where ammonium is partially oxidized into nitrite (partial nitrification) (Dapena Mora, 2007).

In this process, nitrite work as electron acceptor and ammonium is oxidized to nitrogen under anoxic conditions without any emission of nitrous oxide.

The removal of ammonium in wastewater treatment consists of two different processes, the first one nitrification of the half ammonium to nitrite by ammonia oxidizing bacteria. The reaction that takes place is:

$$2\; NH_4{}^+ + 3\, O_2 \rightarrow 2\, NO_2{}^- + 4\, H^+ + 2\, H_2O \quad [\,12\,]$$

In the Anammox process the ammonium that result and the nitrite are converted to nitrogen gas. The following reaction takes place in this step:

$$NH_4{}^+ + NO_2{}^- \rightarrow N_2 + 2\, H_2O \quad [\,13\,]$$

### 3.2.2 Parameters in anaerobic digestion.

Important parameters have inhibitory effects on the wastewater treatment. To prevent microorganism’s death, parameters should be controlled so the process has to be operated in the optimal ranges. (S.Siddhart, 2006).

Influence and control of temperature:

As every chemical reaction, according to the laws of thermodynamics, the rate of reaction increase with temperature. Degradation process is a biological reaction so the decomposition of organic matter is affected by temperature. The issue is that this happens only in certain limits. Usually, microorganisms change their behavior and their metabolism can be affected or inhibited with high temperatures so the digestion decreases at elevated temperature.

Depending on the microorganisms, there are different ranges for optimal temperature. Three different types of microorganisms can be classified according to their optimal temperature: psychrophilic, mesophilic and thermophilic bacteria. The first ones has the optimal temperature below 20ºC, the second one between 20 and 40ºC and the last above 40ºC (Lettinga & Rebac, 2001).

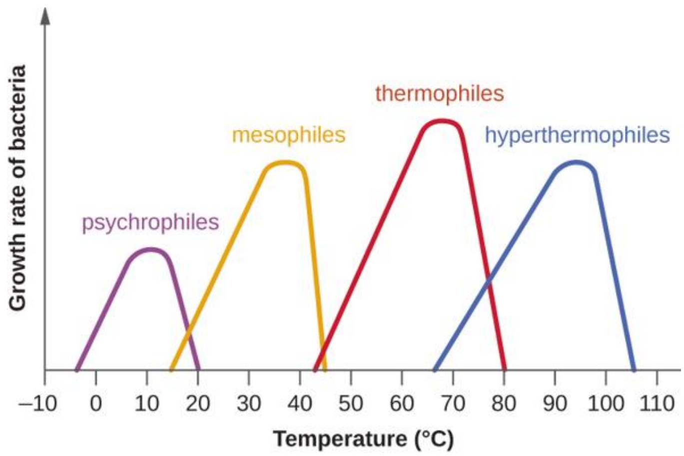



Figure 2. Behaviour of bacteria according to temperature.

The bacteria used in the described plant are mesophilic one. Small deviations from optimal temperature, which is around 35ºC, causes relatively little variations in activity that means a stable process.

Influence and control of pH:

The pH level changes because of biological conversions during the anaerobic process.

Hydrolysis and acidogenesis step have an optimum pH range in between 5.2-6.5 (Solera & Romero, 2001) and acetogenesis and methanogenesis step the optimum pH range is in between 6.7-7.5 because most of the methanogens grow and the production of biogas is favorable at this range of pH (Kayode Feyisetan & Jude Awele, 2015).

In case of reactor three where the Anammox process is carried out, the tolerance range is between 6.7 and 8.3 of pH being 8 the optimal one, but out of this range of tolerance the Anammox bacteria will be inhibited irreversibly. A slight increase in the pH value is caused by the consumption of protons during the process.

Influence of retention time:

Hydraulic retention time (HRT) or residence time is the amount of time that water is in a reactor. Digestion time to digest organic matter determine the retention time of the feedstock. According to (Appels & Baeyens, 2008) in a study on laboratory scale, the obtained relationship between HRT and production of biogas in a semi-continuous stirred tank reactor indicates that volatile fatty acids concentrations are still high at 5-8 days because of the incomplete breakdown of compounds such as lipids. The breakdown curve stabilizes after 10 days when most of the sludge compounds are reduced.

Influence of oxygen in the Anammox process:

Working with high concentrations of dissolved oxygen (DO) cause irreversible inhibition of Anammox process but at low concentration reversible inhibition is possible. Thus, DO should be controlled during the process.

# 4. Engineering process

## 4.1 One and two step processing anaerobic treatment

Anaerobic digestion in two steps:

According to (Pohland & Ghosh, 1971) dividing the microbial population (acid formers and methane formers) in two different reactors, it can be worked with optimum conditions. Each group of microorganisms working in their optimal range of pH for example can increase the stability and control of the process.

In the first reactor hydrolysis and acidogenesis takes place where complex organic compounds are broken and volatile fat acids such as acetic or propionic are produced (Guardia Puebla, 2012). Thanks to the formation of these acids, the pH in this reactor decreases and this it is beneficial to acidogenesis and acetogenesis bacteria that prefer pH ranges of 4.5-5.

In the second step take place acetogenesis and methanogenesis reactions. The volatile fat acids from the acidogenesis step are converted into acetic acid and then into methane (Gerardi, 2003). About two thirds of the methane is produced from acetate conversion and the rest is produce from the reduction of carbon dioxide. Methanogenic bacteria are very sensitive to pH and works well in neutral alkaline medium (6.5-7.2 pH) according (Karena Ostrem, Millrath, & Nickolas Themelis, 2004), (Appels & Baeyens, 2008). Most of the authors that had studied the effect of pH in the separation of these steps ensure that acidogenic bacteria grow more quickly and are less sensible to pH than acetogenic-methanogenic bacteria.

The HRT of this acidogenesis reactor varies between 1.7 and 4 days (Solera & Romero, 2001), whilst the HRT in the second reactor was 4 days as it is shown in Table 2. The most COD reduction is accomplished in the methanogenic reactor, around two times more than in the acidogenic reactor which has around 31% of COD reduction.

Table 2. Performance and operating quantities for the control of the anaerobic process during the period studied (Solera & Romero, 2001).

| Reactor | HRT (t/d) | Organic loading rate( $g\cdot L^{-1}\cdot d^{-1}$) | COD reduction (wt %) |
|---|---|---|---|
| **Acidogenic** | 1.7 | 9.17 | 31.9 |
| **Methanogenic** | 4 | 2.43 | 61.5 |
| **Acidogenic** | 4 | 3.79 | 30.1 |
| **Methanogenic** | 4 | 2.65 | 71.7 |

Anaerobic digestion in one step:

In this way of working there is no control of pH and the four different stages of the anaerobic digestion occur in just one reactor. In this process, the acids produced during acidogenesis are consumed in the next reactions so the decrease in pH is not as big as in the two-step process. During the process the COD decreases in this reaction thanks to the degradation of the organic matter.

# 5. Materials and methods

## 5.1 Configuration and description of the pilot plant.

The studied pilot plant consists of three anaerobic reactors consecutives and a buffer tank equipped with a heating system and a mixer to prepare the wastewater before pumping it to the first reactor. The aim of the first two steps (reactor 1 and 2) is the degradation of organic matter through anaerobic digestion. The last reactor carries out the Anammox process to remove the ammonium present in the wastewater. Figure 3 shows the set up of the plant:

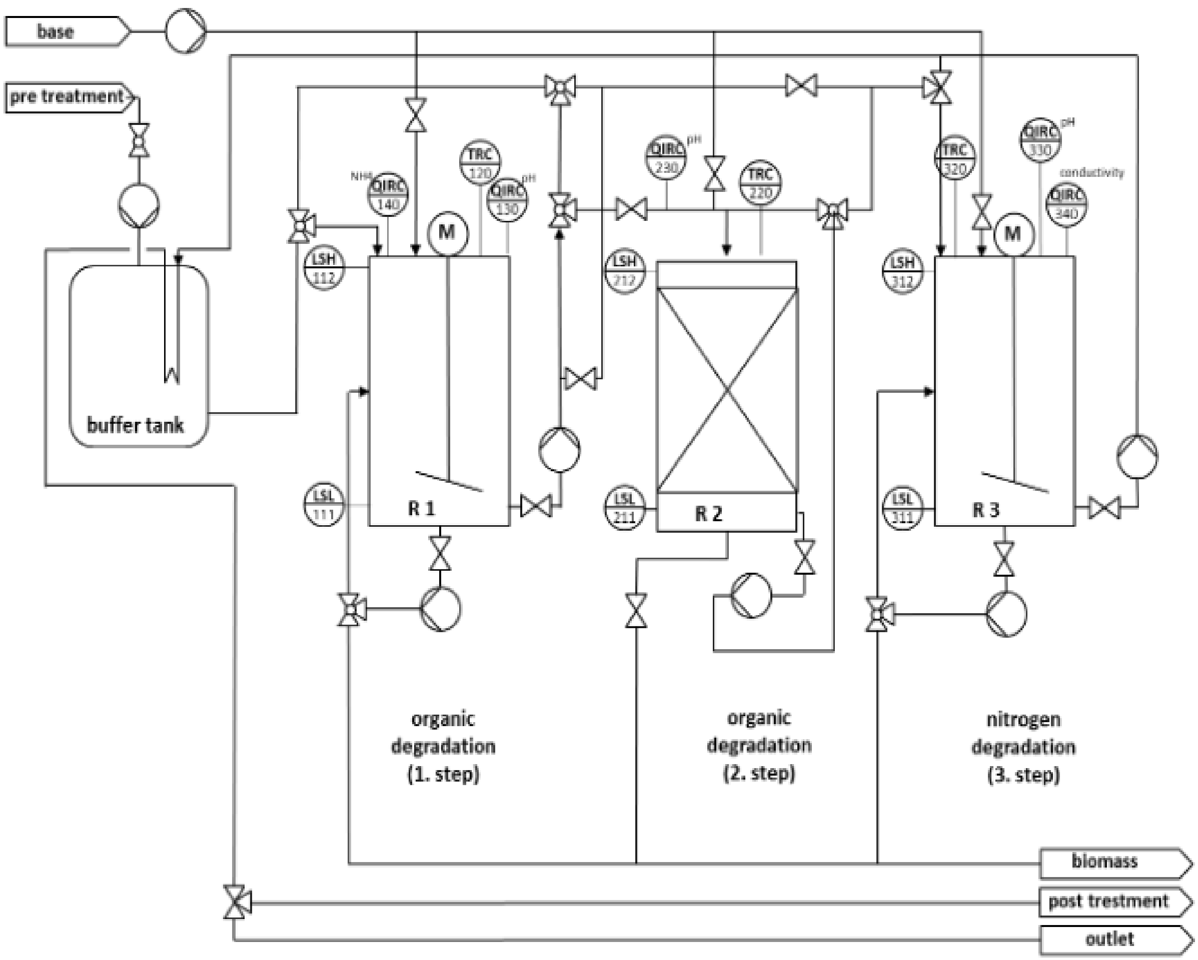



Figure 3. Set up of the wastewater treatment pilot plant.

The pre treatment consists of a buffer tank with a volume of approximately 1000 liters equipped with a heating system to reach the desired temperature of the wastewater and a mixing system to homogenize.

Reactor one (batch stirred tank, BSTR) has 1000 liters of capacity and the two first phases of the anaerobic digestion should predominantly take place. The following pictures show the reactor:

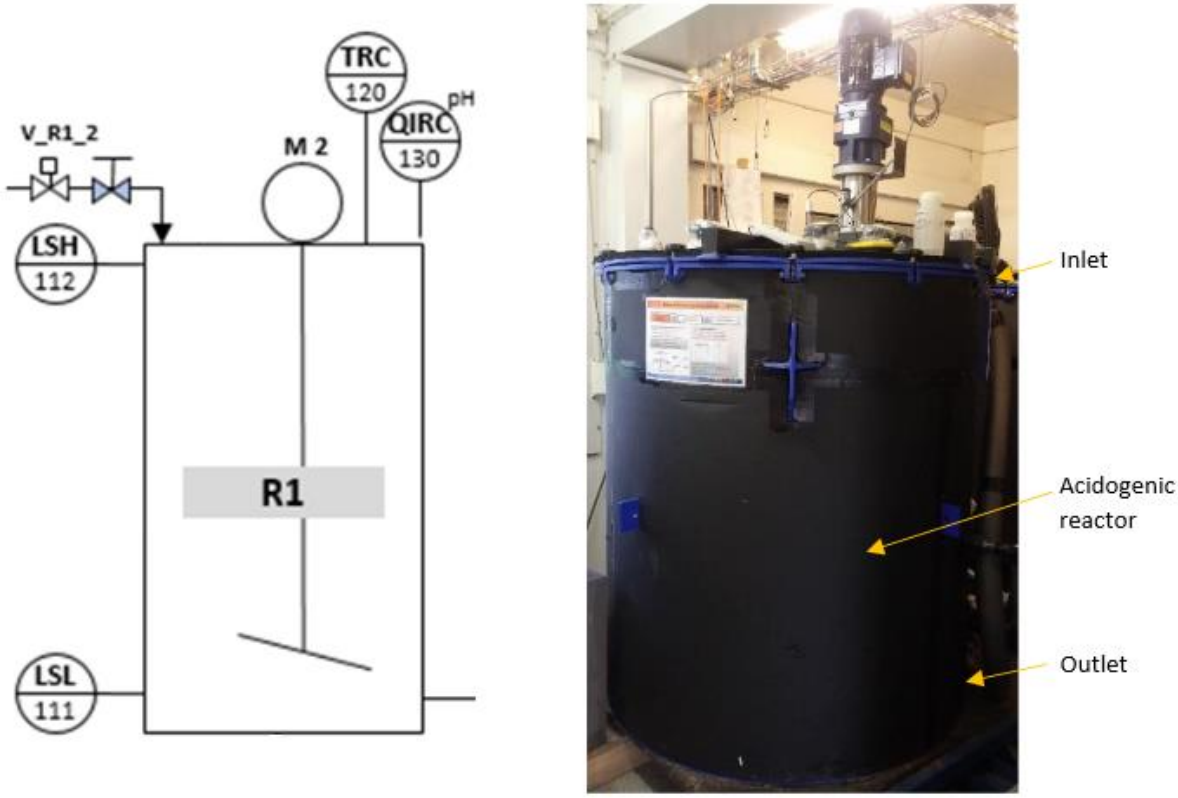
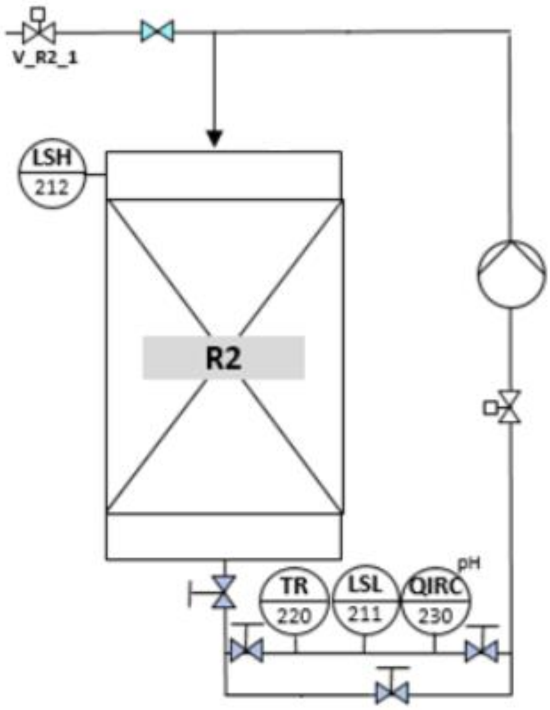



Figure 4. Reactor one.

As the reactor is designed as a batch stirrer tank reactor, it is equipped with a stirrer to homogenize the reactor. In this reactor as the second one the degradation of organic matter takes place.

Reactor two in which acetogenesis and methanogenesis should predominantly take place is designed as a fixed bed reactor (FBR) with 1000 liters approximately; here the degradation process continues. Homogenization of the wastewater inside the reactor is done with a recirculation pump. In the next pictures can be observed the second reactor:



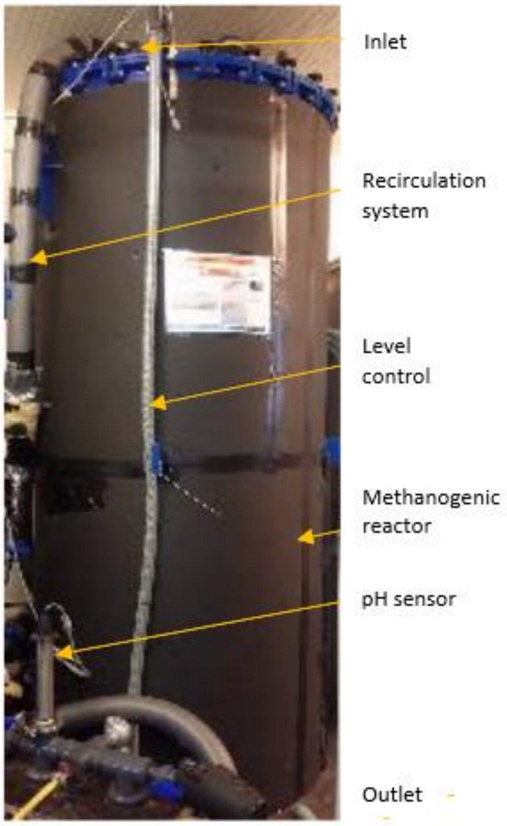




Figure 5. Reactor two.

The third and last reactor is where Anammox process takes place. The volume of this reactor as the others is about 1000 liters. Bacteria inside is Anammox bacteria and the aim is to remove the nitrogen of the wastewater. In this reactor is very important work without any oxygen because it is an inhibitor of the process. In the picture below reactor three is illustrated:

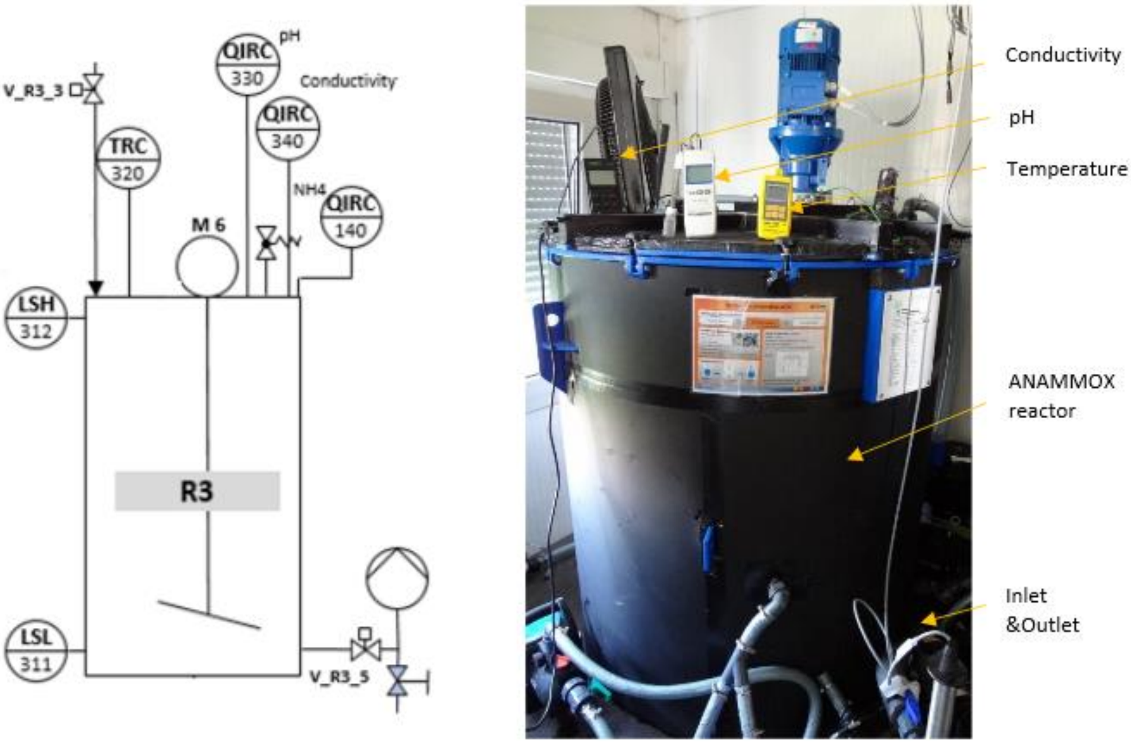



Figure 6. Reactor three.

Buffer tank and reactors are connected through pipelines equipped with different valves and pumps that move the wastewater from one step to the following one. An automatic program is implemented in the plant to control pipes, temperature and other parameters.

## 5.2 Substrate composition for different wastewater scenarios

The aim of this thesis is to analyze artificial wastewater adding different quantities of peptone and saccharose in order to simulate different scenarios of sewage water. In wastewater ammoniacal nitrogen is presented because of the sanitary sewage water of the toilet and basins. Peptone, as proteins is the main source of nitrogen in organic medium; it contains amino acids and short chains of peptides. In the case of saccharose, the common sugar, it is found in many foods and plants. Sugar the same as proteins are presented in our diet and therefore in the wastewater that we produce, that is why peptone and saccharose were chosen as subtract to synthesize different wastewater compositions.

## 5.3 Way of proceed

During this study, different experiments are performed with different concentrations of wastewater to study the behavior of the plant. In Table 3 is shown the different amounts of substrate present in the batches of the wastewater. To study the behavior of peptone, in the first four batches the amount of peptone is increased without any quantity of saccharose. In the next three batches (5 to 7) the amount of saccharose is increased to see the effect in the wastewater without any peptone. And the last four batches with a mixture of both substrates are carried out to study the influence of peptone and saccharose in different quantities in the wastewater.

Table 3. Different amounts of substrate in the wastewater.

| | | | |
|---|---|---|---|
| BATCH 1<br>0 grams Peptone<br>0 grams Saccharose | BATCH 2<br>100 grams Peptone<br>0 grams Saccharose | BATCH 3<br>200 grams Peptone<br>0 grams Saccharose | BATCH 4<br>400 grams Peptone<br>0 grams Saccharose |
| BATCH 5<br>0 grams Peptone<br>200 grams Saccharose | BATCH 6<br>0 grams Peptone<br>400 grams Saccharose | BATCH 7<br>0 grams Peptone<br>600 grams Saccharose | BATCH 8<br>100 grams Peptone<br>100 grams Saccharose |
| BATCH 9<br>100 grams Peptone<br>200 grams Saccharose | BATCH 10<br>100 grams Peptone<br>400 grams Saccharose | BATCH 11<br>200 grams Peptone<br>100 grams Saccharose | BATCH 12<br>400 grams Peptone<br>100 grams Saccharose |

Three days per week (Monday, Wednesday and Friday), water is pumped into the plant from the municipal wastewater treatment plant. The first step is the buffer tank, here the water comes with a temperature around 12-15ºC depending on the weather conditions and is heated to about 24 degrees and homogenised. Measurements of the pH, ammonia level, COD, DO and temperature are taken before and after the addition of the substrate and also samples to analyse in the laboratory. When the substrate is completely dissolved, the water is pumped into reactor one through pipes and the help of a centrifugal pump installed in the outlet of the buffer tank. After 15 minutes of mixing in this reactor, new samples and measurements are taken like in the previous step. The wastewater is processed during the following two days. New samples are taken in 24 hours with the same procedure and also before pumping into the next reactor at 48 hours. When the wastewater is in the second reactor it has to be homogenised by the recirculation pump. The time of waiting until the taken of new samples and measurements is about 30 minutes to reach the whole homogenization. Again, the water is 48 hours in this reactor but samples are taken 24 hours and 48 hours after the pumping. After taking 48 hours samples, the water is pumped to the waste.

Every day, the taken samples are analysed in the laboratory with special kits and devices. The parameters which are analysed are COD and ammonia. For these analyses, the instructions of the kits are followed as indicated.

## 5.4 Devices and test kits used

In this subchapter the devices and the test kits used for the measurements are going to be described.

For the measurement of pH in the plant it is used PH-100ATC sensor device from Voltcraft. The determination of pH consists in measuring the potential that develops through a thin membrane of glass that separates two solutions with different concentration of protons. Consequently the sensitivity and selectivity of the glass membranes during pH are very well known. The sensor is very delicate so it should be saved in a solution of potassium chloride (KCl). (Ebbing & Gammon, 2010).


Figure 7.Typical pH sensor.

The DO sensor is a device used for the measurement of oxygen dissolved in the wastewater and the temperature. The one that is used in these measurements is DO-100 from Voltcraft. The sensor is introduced in the wastewater and for the measurement it is needed to wait until a constant value. Two scales are used, the first one is ppm or parts per million and the second one in percentage saturation (Percentage of oxygen dissolved in 1 liter of water, with respect to the maximum amount of dissolved oxygen that may contain 1 liter of water) (Yaque Sánchez, 2013).


Figure 8. Typical DO sensor

Spectrophotometer Pharo 100 from Merk GmbH was used for the measurements of the samples to obtain the values of COD and $NH_4$-N. It is an instrument used in chemical analysis to measure, as a function of wavelength, the ratio of values of the same photometric magnitude relative to two beams of radiation and the concentration or chemical reactions that are measured in a sample. The samples that are prepared in measuring cells are introduced into the apparatus. The samples are prepared in small cuvettes of methacrylate. This measuring cell has two transparent sides and two opaque. The cuvette has to be put in the spectrophotometer in such a way that the beam passes through the transparent opposites sides. The following pictures show a typical spectrophotometer and small cuvettes. (Hernández Hernández & González Pérez, 2002).

Figure 9. Measuring cell.


Figure 10. Spectophotometer.

Another device used was a thermo reactor. These devices are required to determine COD, total nitrogen and total phosphorus. These instruments guarantee a complete digestion of the sample, as they maintain the high reaction temperature for the entire defined period. The samples are prepared in small tubes that are introduced into the thermo reactor. These tubes and a thermo reactor can be seen in the next pictures. (*Bureau Veritas Colombia)*.


Figure 11. Thermo reactor.


Figure 12. Measuring tubes for thermo reactor.

Ammonium Test Kit of Merck KGaA is used to obtain the value of ammonium present in the taken samples. The kit has a bottle of reagent $NH_4$-1, one bottle of reagent $NH_4$-2 (contains granulate and desiccant capsule) and two Auto Selectors to select the program in the spectrophotometer. The measuring range was 2.0-75.0 mg/l NH4-N and the standard deviation of this method is $\pm$0.48. The picture below shows the package content. (Merck KGaA).


Figure 13. Ammonium Test kit.

For the analyse of COD, two solutions of Merck  KGaA were used, the first one was COD solution B and the second one COD solution A. Depends on the range of COD there were two possibilities for solution B one for the range 10-150 mg/l and the other one 100-1500 mg/l. Both solution are presented in Picture 14. (Merck KGaA).

The chemical oxygen demand is the amount of oxygen needed to oxidize all the organic and oxidizable matter present in a wastewater. It is therefore a representative measure of the organic pollution of an effluent.

Figure 14. Solution A and B for COD analyse.

# 6. Evaluation and discuss of experimental results

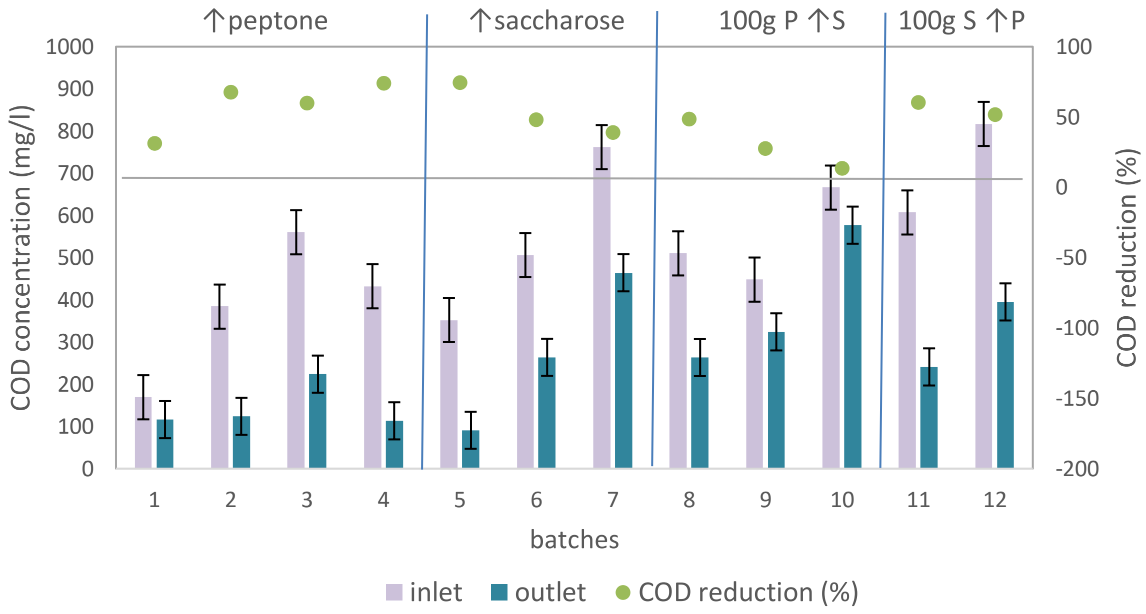
In this chapter is going to be discussed the obtained results with the help of different graphs that show the relation of some analyzed parameters from the first two reactors and buffer tank.

The degradation process starts in the pumping of the wastewater into the buffer tank and continues in the next reactors.

The addition of the substrate is made in the buffer tank during the experiments without pH control.

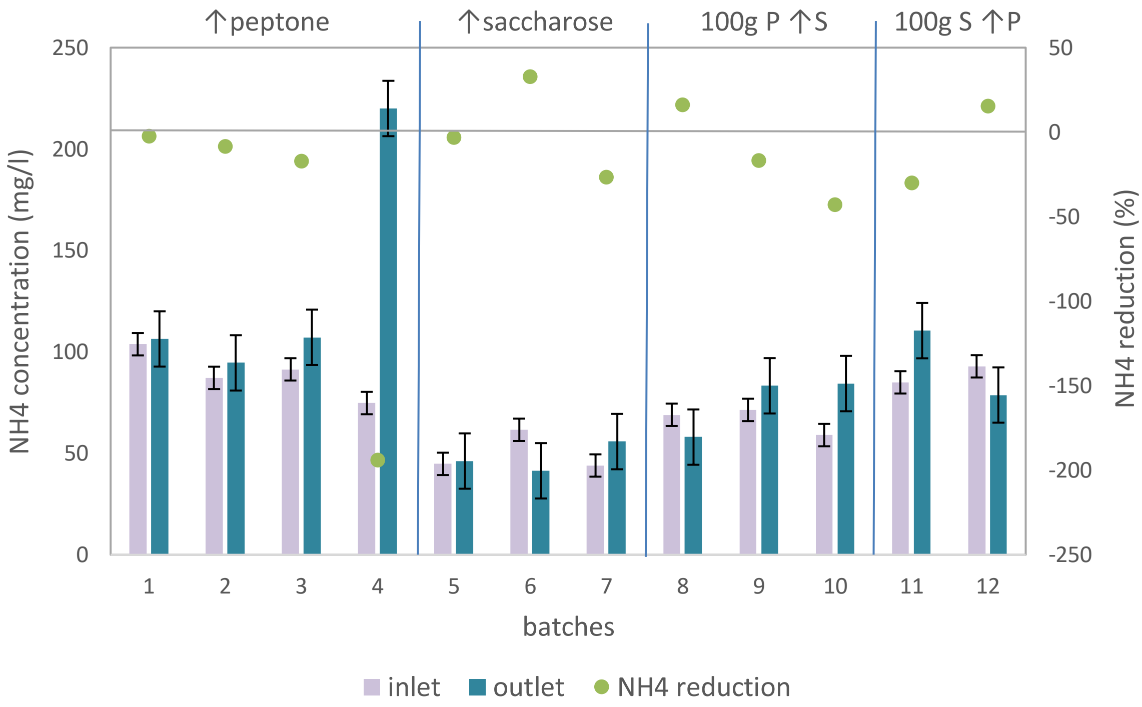
## 6.1 Degradation in reactor 1

In Graph 1 is shown the results obtained in the measurements of chemical oxygen demand for the different studied batches in reactor 1. In the X-axis are represented the 12 batches divided in 4 sections according to the substrate that influences. In the first section, the first 4 batches (just increasing peptone) are represented. The light column represents the inlet COD concentration in mg/l and the dark column the outlet COD concentration after about 48 hours. In the secondary axis, COD reduction in percentage is represented according the inlet and outlet obtained concentration. What is can be observed in this section is the more peptone added the more COD reduction is obtained. The COD reduction of batch 2 is a bit higher than the trend line because this batch was realized after batch 4 (400g of peptone) so some residual substrate could be left in the reactor. The second section shows the influence of saccharose addition, when adding more saccharose a decrease of COD reduction is observed. This can be observed also in the behavior of the next 3 batches (a constant quantity of 100grams of peptone but increasing the amount of saccharose). In the last section, when more peptone is added (with a constant quantity of 100 grams of saccharose) a decrease of COD reduction is observed, this behavior can be explained due to a dissolving problem of the substrate in the reactor. The bars situated in the top of each column represent the error bars with typical error.




Graph 1. COD concentration and reduction in the different batches in reactor 1.

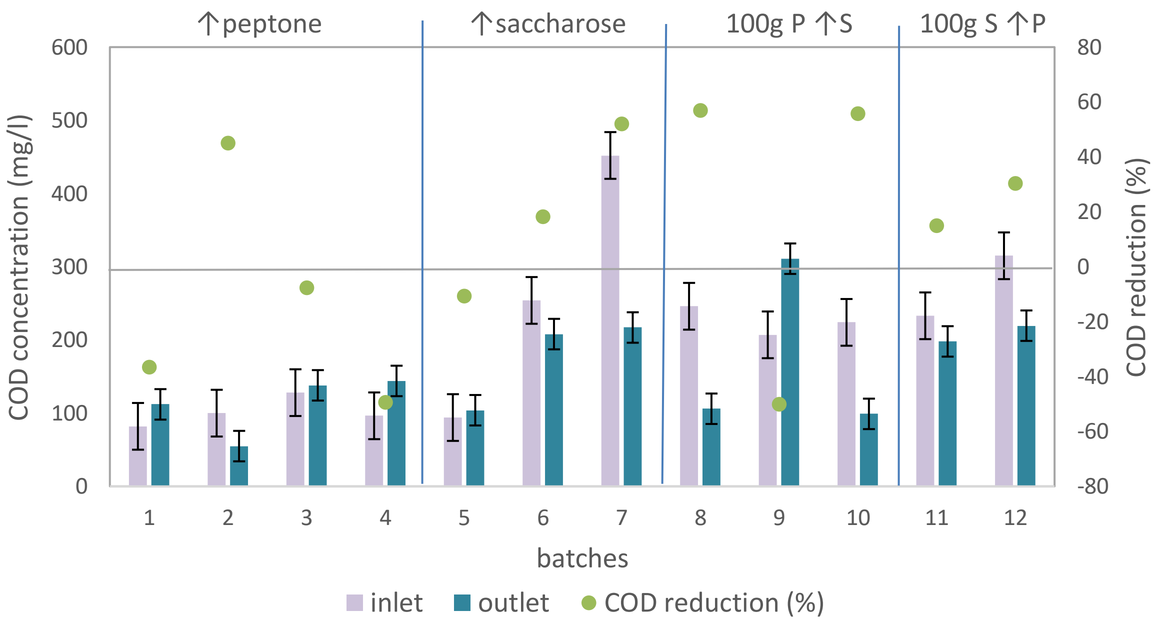
What it can be seen in Graph 2 is the inlet and outlet ammonium concentration in mg/l for the different studied batches in reactor 1. In the secondary axis is represented the ammonium reduction in percentage. A positive percentage means a reduction of $NH_4$ concentration but a negative one means a production. The batches are divided in sections as it was mentioned before. What it can be say in this graph is that more peptone added (without any saccharose) more production of ammonium takes place because the percentage of $NH_4$ reduction is negative. The next sections where more saccharose is added it can be observed a less reduction of ammonium finishing with production of ammonium. And in the last section, as there were problems in dissolving the substrate, the concentration of ammonium was not increased but reduced. In batch 4 it can be observed a huge production of ammonium from 74.8 mg/l in the inlet concentration to 220 mg/l in the outlet one.




Graph 2. NH4 concentration and reduction in the different batches in reactor 1.

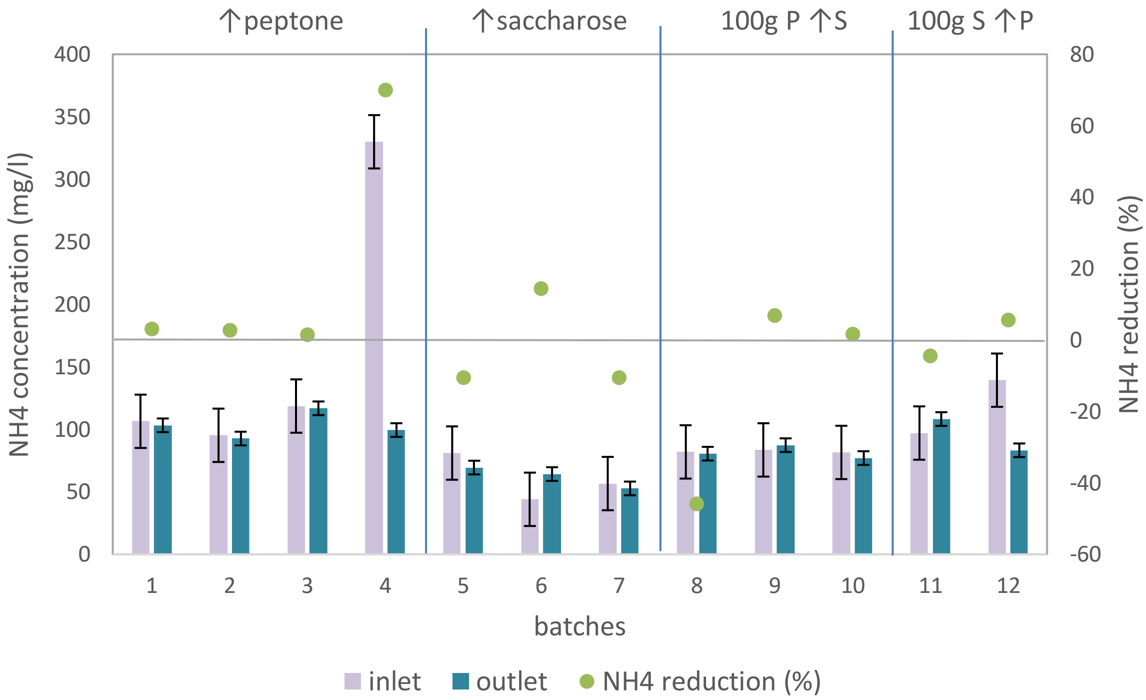
## 6.2 Degradation in reactor 2

Below is represented Graph 3 where COD concentration and reduction of reactor 2 are shown. More peptone added causes an increase of COD concentration whereas more saccharose added causes an increase of COD reduction. In batch 12 the dissolving problems caused a decrease in the reduction of COD in reactor 1 but the residual substrate was dissolved in the second reactor that is why the COD reduction increased in this graph. To finish commenting this graph, batch number 9 has a huge production of COD in the sample taken 48 hours after pumping into reactor 2. This was due to some problems in the mixing of the reactor but at the end of the degradation process of this batch, the COD decrease as it is shown in Graph 9 that shows the evolution of COD along the retention time.




Graph 3. COD concentration and reduction in the different batches in reactor 2.

Graph 4 represents the $NH_4$ inlet and outlet concentration in mg/l and reduction in percentage against the 12 different batches. The ammonium concentration in this reactor remains constant in the first four batches. This happens because most of the ammonium was digested in the first step and almost nothing is digested in reactor 2. What it can be observed is that batch number 4 has a huge reduction of $NH_4$ due to the great formation of this compound in the previous reactor as it was mentioned before. By the contrary, in reactor 8 there is a big production of $NH_4$ which may be due to dissolving problems with the substrate in reactor 1. As it was mentioned before, there were some problems with the mixing in batch 12 that caused in reactor one a no completed dissolution (reducing the concentration of $NH_4$ and behaving contrary to what should happen with more peptone added). This no dissolved subtract was dissolved in the second reactor at first and later digested by the microorganisms. The samples were taken with 4 days between them (not 48 hours) so the results do not reflect what happened in the while but it is known that at the end the ammonium concentration was reduced in this 12$^{th}$ batch (see Graph 4).

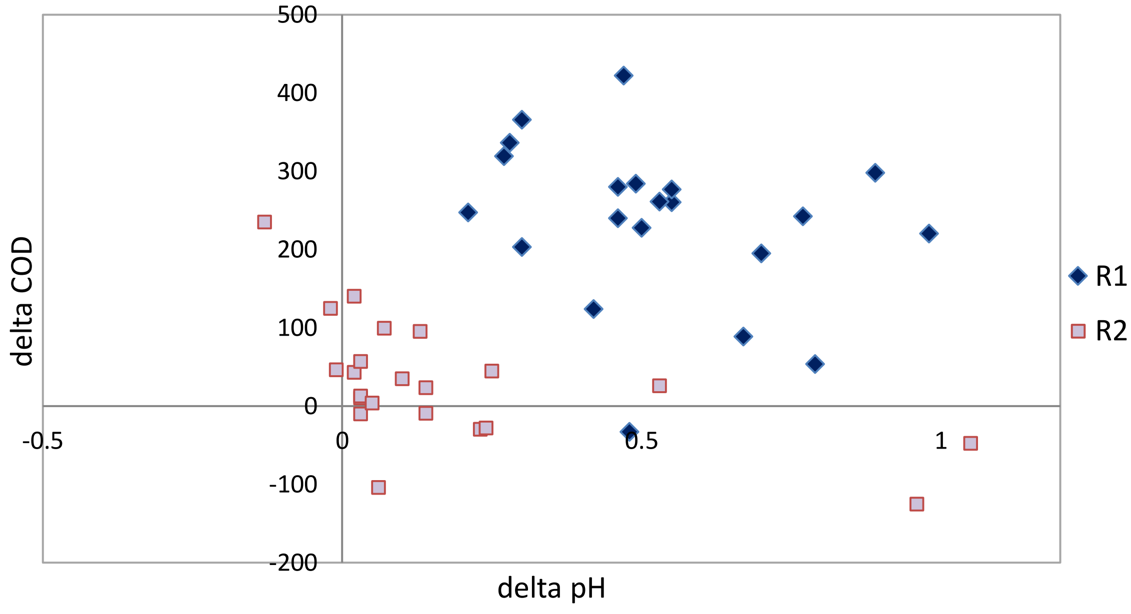



Graph 4. NH4 concentration and reduction in the different batches in reactor 2.

## 6.3 Comparison of reactor 1 and reactor 2.

### 6.3.1 Influence of delta pH in COD concentration in reactor 1 and reactor 2.

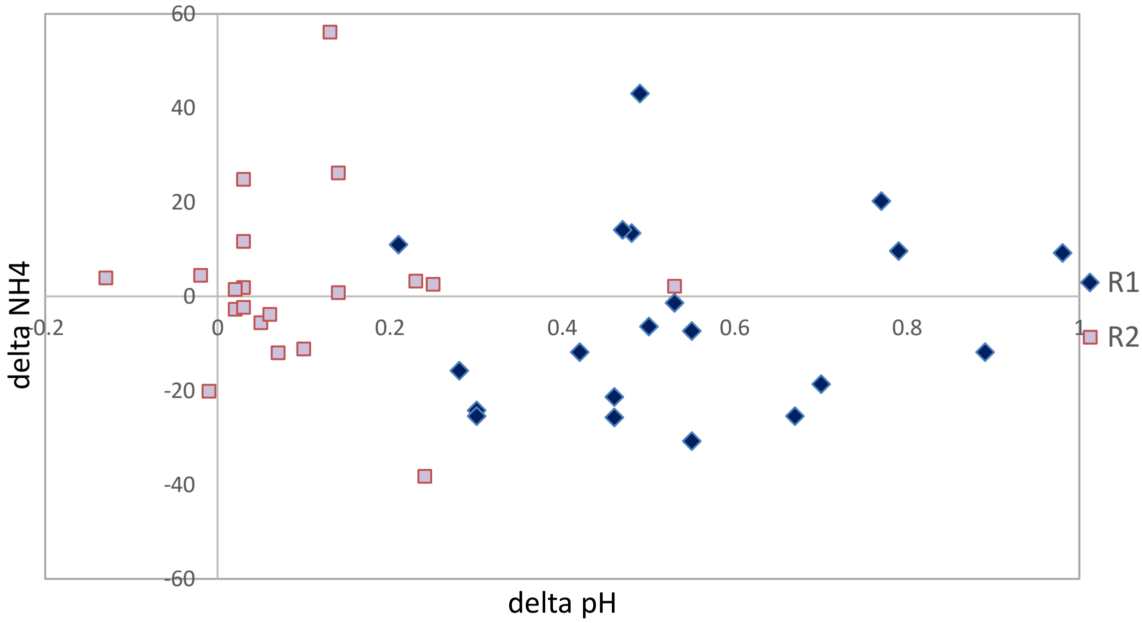
As it can be observed in Graph 5, delta COD is represented against delta pH for both reactors. Delta COD and delta pH is bigger in reactor one than in two. The bigger variation of COD is because most of the digestion process takes place in the first reactor and the rest continues in the second one as it can be seen in the graph (variation of COD in reactor 1 is between 100 and 400 hundred against reactor two that has a variation between around 0 and 100 mg/l). The decrease of the pH variation is because of the formation of acids in the acidification step and most of them are produced in the first step.




Graph 5. Delta COD against delta pH in reactor 1 and reactor 2.

### 6.3.2 Influence of delta pH in ammonia concentration in reactor 1 and reactor 2

In Graph 6, it can be seen that more ammonia is produced in the first reactor than in the second one (negative delta $NH_4$ means production of this compound), this is because the substrate which is added is a source of organic matter and it degrades to amino acids. If there is a good homogenization and dissolving process, most of this substrate is degraded in the first step, if not this takes place in the second one.

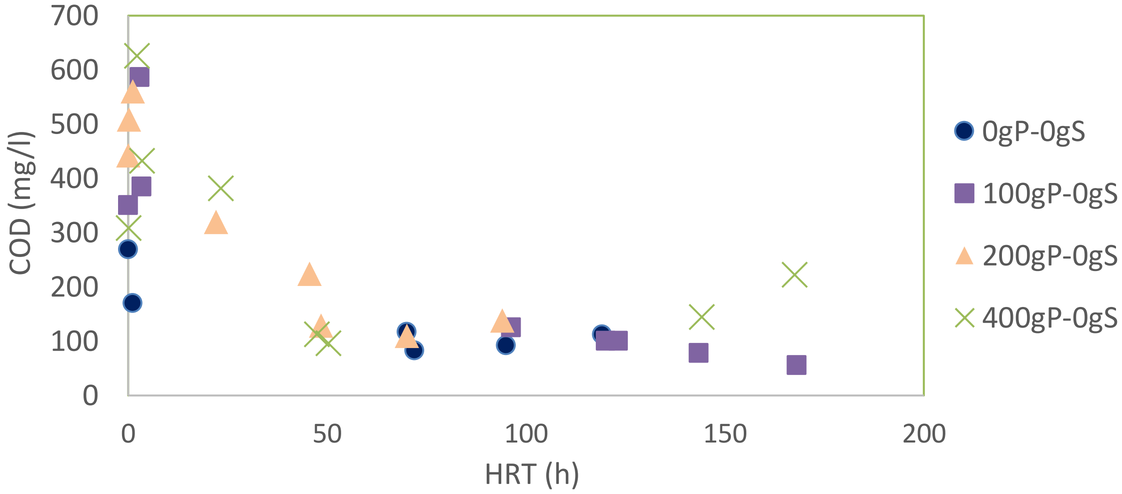



Graph 6. Delta ammonia concentration against delta pH in reactor 1 and reactor 2.

### 6.3.3 Evolution of COD with the retention time.

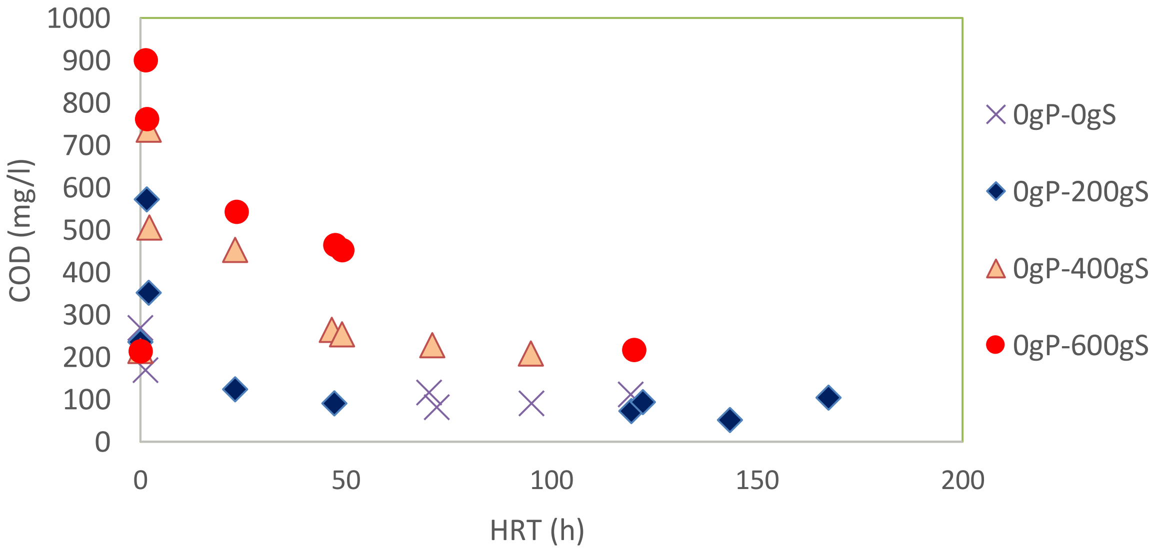
In this subchapter some graphs are going to describe de evolution of the organic matter during the process of degradation of the different batches of wastewater. The batches are divided in three different graphs according the substrate influence. The first one represents batches 1 to 4 (increasing peptone without any saccharose). The second one represent the influence of saccharose and the last one the evolution of the batches with both substrates.

As it can be observed in Graph 7, COD level decrease during the whole process of degradation but taking place a bigger decrease in the first reactor as it was mention previously. In the first 50 hours the COD level for example in batch 4 (200 grams peptone) changed from 500 mg/l until 100 mg/l or in batch 1 (without any substrate) from 200 until 100 mg/l. The initial level of COD increase with the more substrate added. This is normal because more quantity of substrate means more organic matter that should be degraded in the process. The initial value of COD without any substrate is around 200 mg/l and the addition of each 100 grams of peptone makes an increase of 100 units in the initial COD. Another thing to mention is that more substrate added more initial decrease in COD value. This can be observed in the slope from 0 to 50 hour. Batch one decrease 100 units (from around 200 to 100 mg/l), batch 3 decrease 400 units and batch 4 decrease 500. In the case of batch two, it decreases from 600 to 200 but in 100 hours, so in 50 hours it may decrease 200 mg/l.



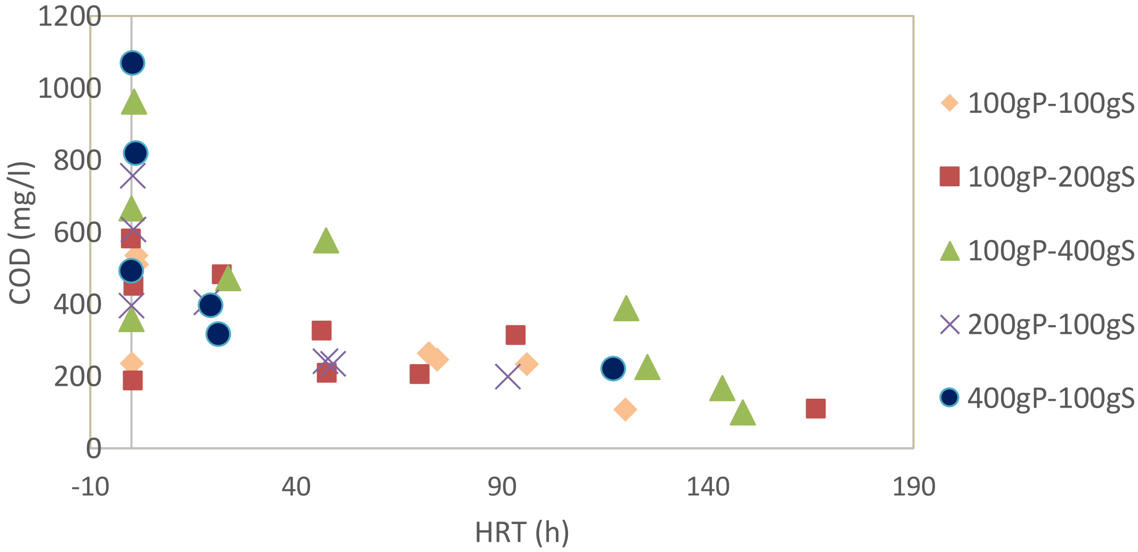

Graph 7. Evolution of COD with HRT in batches 1-4 (influence of peptone).

In Graph 8 COD level from batches 5 to 7 are represented with the time. Batch 1 is also represented as a reference of no substrate. This graph illustrates the decrease of COD level in the batches with addition of saccharose and no peptone. As the graph above, the COD decrease more quickly in the first 50 hours than in the rest of the time. All the batches without addition of any substrate start in COD levels around 200 mg/l (time 0). The trend is the same as with the addition of peptone, around each 200 grams of substrate added, the initial COD increases 200 mg/l. If batch 1 is compared with the rest batches, it can be said that the decrease of COD in this batch is lesser. These curves differ very well the evolution of each batch; more saccharose added the curve moves up to more concentration of COD.

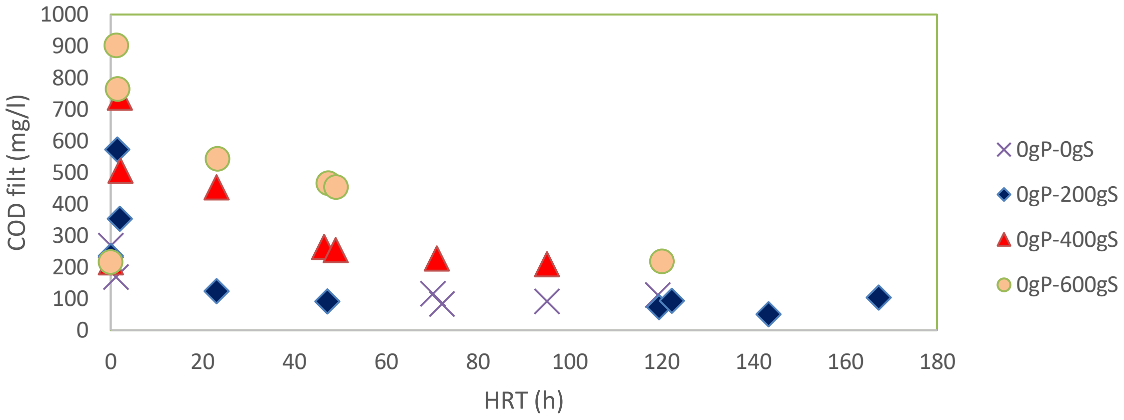



Graph 8. Evolution of COD with HRT in batches 5-7 (influence of saccharose).

The evolution of COD with the time for the batches 8-12 is represented in Graph 9. This graph is not as clear as the others but what it can be seen is COD concentration decreasing with the time. In this graph, apart from the initial increase of COD, it can be observed that for some batches, the COD concentration increases for a while in the middle of the degradation process but later decreases. This happens to batches 9 (100 grams of peptone-200 grams of saccharose) and 10 (100 grams of peptone and 400 grams of saccharose). In the case of batch 9, the evolution of COD begins with the typical increase of COD level from 200 until 600 mg/l because of the addition of substrate. Then it decreases until 207 mg/l 48 hours after the pumping into reactor 1 but another increase (up to 311 mg/l) takes place until 93 hours later. After this increase, the degradation process continues and the last value of COD concentration is 108 mg/l. Batch 10 suffers the same increase from 471 mg/l to 577 mg/l in COD concentration but 40 hours after pumping into reactor one. After that, COD starts to decrease gradually until it finishes in 99 mg /l when 150 hours have passed. These increases are due to the dissolving problems that were mentioned in the subchapters of degradation in reactor 1 and 2.




Graph 9. Evolution of COD with HRT in batches 8-12 (influence of peptone and saccharose).

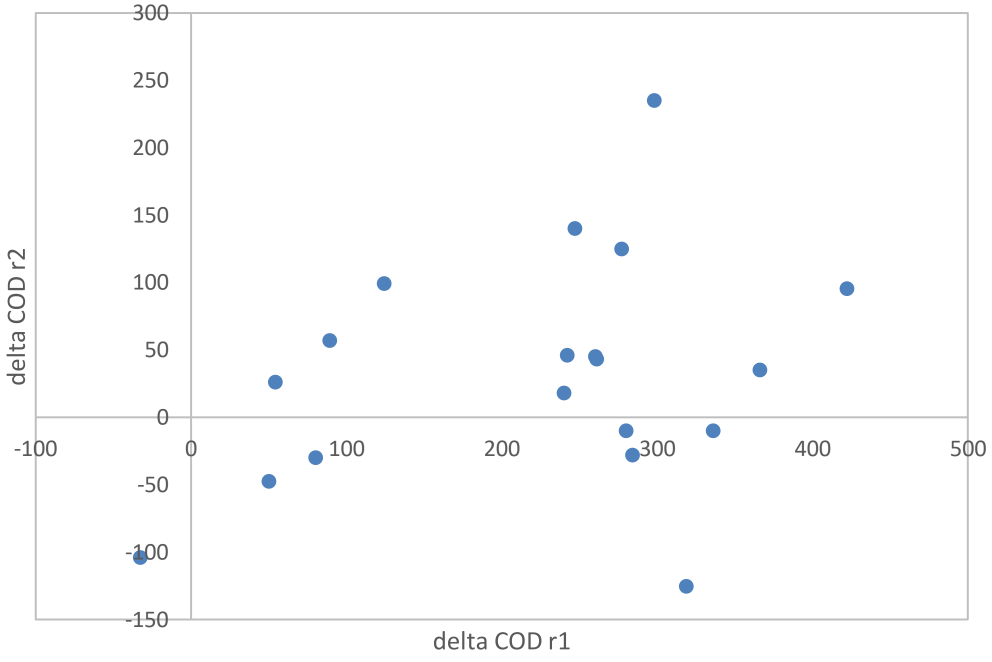



Graph 10. Influence of saccharose in COD of filtrated samples**.**

### 6.3.4 Comparison of degradation process in reactor 1 ad reactor 2.

Comparing the results of the addition of peptone and saccharose in COD in filtrated samples, it can be said that the influence is quite similar, add 200 grams of peptone or 200 grams of saccharose to the wastewater produce an increase of around 400 mg/l in COD level at the beginning and after certain of time the degradation produce a negative slope curve.

Most of the digestion of organic matter is carried out in the first reactor and this can be shown in the following graph. Delta COD in reactor 1 oscillates between 100 and 400 versus 0 and 250 in reactor 2, this means that more or less two times more of digestion occurs in the first reactor.




Graph 11. delta COD in reactor 1 against delta COD in reactor 2.

# 7. Error discussion

This chapter describes that the analyses of the samples carries with them some measurement errors. These errors are due to different causes: errors in the preparation of the sample because of human failure or accuracy in the pipettes, test kits or devices used during the measurements. Also some errors are due to lack of complete homogenization in the reactors or buffer tank, residual substrate of a batch in the following or plant failures. Great care must be taken in the dosage of the solutions using pipettes or spoons because the quantities used are very small and carry to big errors.

In the instructions of each test kit there is a section of quality assurance in which standard deviation, coefficient variation, sensitivity or accuracy of the procedure are specified.

The different graphs have in the top of their columns typical error bars which shows the variation that can be found in the results.

During the measurements, double samples were taken to study the error of measuring. With the results obtained from the double measurement the standard deviation was calculated obtaining a value of 11,314.

# 8. Conclusions

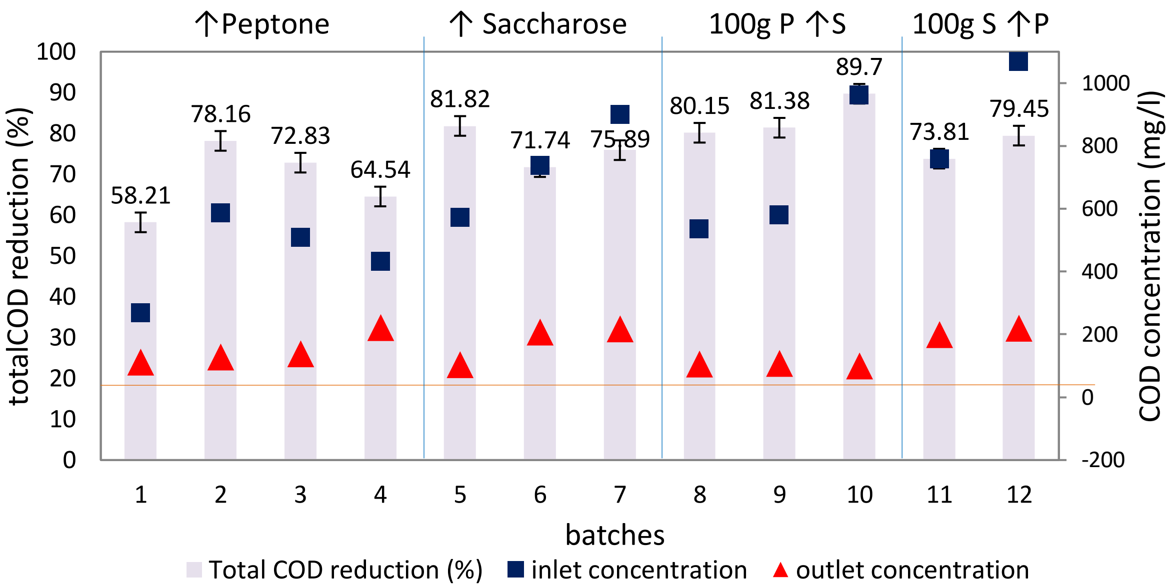
In this chapter some conclusions about the study of these different scenarios of wastewater in an anaerobic degradation process are going to be described.

To begin it has to be mentioned that the process was carried out in one step; this means that the study was not carried out under optimal conditions but nevertheless, the degradation of the organic matter was successful. Graph 12 shows the total COD reduction in percentage according the different studied batches. Most of them have a reduction which exceeds 71%.

The different batches are represented in x-axis separated in the four sections as before while total COD reduction is represented in y-axis. With this graph it can be observed that the addition of saccharose causes more reduction in organic matter than the addition of peptone. This can be explained as follows, if batch 3 (200 grams peptone without any saccharose) is compared with batch 5 (200 grams of saccharose without any peptone), the reduction of COD in batch 3 is 9 units less than in batch 5. The same happens if batch 8 (100 grams of peptone and 200 grams saccharose) is compared with batch 10 (100 grams saccharose with 200 grams peptone), the second batch has 6.34% less of reduction in COD than the first one.

In the secondary axis are represented two series of points that shows the inlet and outlet COD concentrations of the different 12 batches. The horizontal line reflects the limit value of COD in service water that is fixed in 75 mg/l. With this, what it can be said is that although the reduction of COD is quite big, the limit value has not been archived with the retention time that each batch was in the process. Two different options to reach this value are: increment the retention time of the wastewater inside the process, this means microorganisms have more time to degrade the organic matter and there is enough time to carry out all the reactions of the process; or work with optimal conditions, controlling the pH and temperature in the reactors so microorganisms can work in a better way degrading the organic matter of the wastewater.

Another conclusion to consider is that a good mixing system is needed to have totally homogenization inside the reactors. In some batches such as number 12, some dissolving problems with the substrate have taken place. These problems caused an increased of COD in the middle of the degradation process that could have been avoided with a complete dissolution of the substrate thanks to a good mixer.




Graph 12. Total COD reduction (%) and inlet-outlet concentrations in the different batches.

To finish with the conclusions, this study has been made working with one-step process where the four different phases take place in the same reactor. More degradation of organic matter took place in the first reactor than in the second, this can be reflected in Graph 11 or comparing the different graphs of subchapter 6.3.3 where the slope is greater in the first part of the degradation process. The production of $NH_4$ is also bigger in the first reactor because of the addition of substrate. In this reactor begins the degradation of the carbohydrates and proteins from the organic matter of the wastewater producing acids.

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# 10. Appendix

## 10.1 List of tables



## 10.2 List of figures

## 10.3 List of graphs