Bioponics : Recylcing goat and chicken manure into nutrient solution for hydroponics through aerobic and anaerobic digestion

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MARIE KHRONIS

TRAVAIL DE FIN D'ETUDES PRESENTE EN VUE DE L'OBTENTION DU DIPLOME DE MASTER BIOINGENIEUR EN SCIENCES ET TECHNOLOGIES DE L'ENVIRONNEMENT

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CO-PROMOTEURS : HAISSAM JIJAKLI, PIERRE RAULIER

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RESUME

La culture hydroponique a montré, depuis son renouveau dans l'agriculture urbaine, ses nombreux avantages en termes d'espace, de productivité et d'économie d'eau. Malheureusement, elle présente un inconvénient majeur : sa dépendance aux engrais minéraux. La bioponie est une technique innovante qui vise à remplacer les engrais minéraux dans les systèmes hydroponiques par des engrais organiques. Les engrais organiques, à l'opposé des engrais minéraux, peuvent être fabriqués partout dans le monde. Ils trouvent donc une utilité directe dans les pays en voie de développement.

Ce travail a comme objectif d'évaluer les possibilités et l'efficacité d'un système de minéralisation de la matière organique permettant la création d'une solution nutritive à partir d'excréments de chèvres et de poules, élevages communément retrouvés en République Démocratique du Congo. Afin de réaliser cet objectif, plusieurs paramètres ont été étudiés comme la concentration en matière organique, le type de digestion (aérobie, anaérobie), la gestion du pH et le type de tampon utilisé. L'impact de ces paramètres sur le système a été analysé en étudiant le pH, la température, la CE et les concentrations de NPK libérés en solution.

Les résultats de cette thèse de master ont permis la création d'un système de minéralisation fonctionnel de faible technicité qui permet la réplication du processus de digestion. Cependant, trois faiblesses ont également été identifiées : la chute des températures, les pertes de matière organique et la méthode d'échantillonnage. De plus, le processus de minéralisation effectué par le système n'est pas optimal. En effet, la nitrification de l'azote n'a pas eu lieu lors des expériences mises en place. En outre, l'équilibre entre les concentrations d'azote, de phosphore et de potassium n'est pas idéal pour une solution nutritive complète permettant une croissance optimale des plantes. En effet, les concentrations en NH4 et potassium sont respectivement 2 à 25 fois et 5 à 15 fois plus élevées que les valeurs retrouvées dans la littérature. A l'opposé, les concentrations en phosphore sont 5 à 15 fois plus faibles que les valeurs de la littérature.

Cependant, malgré ces lacunes, plusieurs conclusions intéressantes ont été mises en évidence comme l'influence principale du type de matière organique. Le type de digestion et la gestion de pH semble également jouer un rôle mais celui-ci est moins clair que pour la matière organique. Le système créé ne permet donc pas la création d'une solution organique idéale pour la culture hydroponique. Cependant, ce travail fournit de nouvelles connaissances sur les systèmes bioponiques. D'autres recherches sont bien sûr encore à effectuer afin de pouvoir caractériser les processus et créer un système "low tech" de minéralisation de la matière organique performant.

Mots clés : Bioponie ; Minéralisation ; Digestion aérobie ; Digestion anaérobie ; Hydroponie organique ; Solution nutritive ; Fertilisant organique ; Déjections de chèvres ; Déjections de poules

ABSTRACT

Since its revival in urban agriculture, hydroponics has shown its many advantages in terms of space, productivity, and water savings. Unfortunately, it has a major disadvantage: its dependence on mineral fertilisers. Bioponics is an innovative technique that aims to replace mineral fertilisers in hydroponic systems with organic fertilisers. Organic fertilizers, as opposed to mineral fertilizers, can be made anywhere in the world. They are therefore of direct use in developing countries.

The objective of this work is to evaluate the possibilities and effectiveness of a system for mineralising organic matter to create a nutrient solution from the excrement of goats and chickens, which are commonly found in the Democratic Republic of Congo. To achieve this objective, several parameters were studied such as organic matter (OM) concentration, type of digestion (aerobic, anaerobic), pH management and type of buffer used. The impact of these parameters on the system was analysed by studying pH, temperature, EC and NPK concentrations released in solution.

The results of this master thesis allowed the creation of a functional mineralisation system of low technicality that allows the replication of the digestion process. However, three weaknesses were also identified: the drop in temperature, the loss of organic matter and the sampling method. In addition, the mineralisation process carried out by the system is not optimal. Indeed, nitrogen nitrification did not take place during the experiments set up. Furthermore, the balance between the concentrations of nitrogen, phosphorus and potassium is not ideal for a complete nutrient solution for optimal plant growth. Indeed, the concentrations of NH4 and potassium are respectively 2 to 25 times and 5 to 15 times higher than the values found in the literature. Conversely, phosphorus concentrations are 5 to 15 times lower than the values found in the literature.

However, despite these shortcomings, several interesting conclusions have been highlighted as the main influence of the type of organic matter. The type of digestion and pH management also seems to play a role, but this is less clear than for OM. Therefore, the system created does not allow the creation of an ideal organic solution for hydroponics. However, this work does provide new knowledge about bioponic systems. Further research is of course still to be carried out to characterise the processes and create a low-tech system for the mineralisation of OM.

Key words: Bioponics; Mineralisation; Aerobic digestion; Anaerobic digestion; Organic hydroponics; Nutrient solution; Organic fertilizer; Goat droppings; Chicken droppings

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GLOSSARY

EC: Electrical conductivity or Electroconductivity

pH: Potential of hydrogen

N_2: Dinitrogen or molecular nitrogen

NH_4^+: Ammonium

NH_3: Ammonia

NO_2^- : Nitrite

NO_3^-: Nitrate

TAN: Total Ammonia Nitrogen (NH_4^+ and NH_3)

TIN: Total Inorganic Nitrogen ($N\text{-}NH_3$, $N\text{-}NO_2^-$ and $N\text{-}NO_3^-$)

K: Potassium

P: Phosphorus

OM: Organic matter

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1 INTRODUCTION

This master thesis is part of Félicien Mununga katebe's PhD thesis carried out between the University of Liège and the University of Lubumbashi (Democratic Republic of Congo). The objective of the thesis is to produce uncontaminated vegetables in Katanga, where soils present high heavy metals pollution. The techniques developed during the first year of this PhD thesis have made it possible to reduce the concentration of pollutants in vegetables, but not below the acceptable consumption threshold proposed by the FAO. He therefore turned to a hydroponic system to get rid of the polluted soil. Indeed, hydroponics is defined as an off-soil cultivation technique in which plants grow on a substrate and are supplied with a nutrient solution (Tomson, 2019).

However, access to nutrient solutions in Katanga is also a problem. It can be solved by recycling organic matter, such as goat and chicken droppings, from locally available farms, into nutrient solution.

The main objective of this work is therefore to create a nutrient solution for hydroponics from chicken and goat droppings under controlled conditions. Three specific objectives can also be formulated to:

a) Create a state of the art on fertilization in hydroponics.
b) Create and test an experimental system and protocol for the mineralisation of organic matter in aerobic and anaerobic conditions.
c) Carry out exploratory tests to evaluate which parameters allow the best NPK ratio when mineralising goat and hen droppings and produce a nutrient solution that can be used in hydroponic systems.

This work is divided into two parts. The first part meets the first specific objective of creating a state of the art. The second part describes the experiments implemented. It is divided into three sub-parts. Firstly, two preliminary tests meet the specific objectives b and c. Secondly, a first experiment allows to deepen the response to objective c. Finally, the second experiment allows the realisation of a nutritive solution which will be tested, outside the framework of this work, on lettuces in hydroponics.

2 LITERARY REVIEW

2.1 HYDROPONICS

The term hydroponics comes from the Greek words "HYDROS" and "PONOS" meaning water and work, respectively. Literally, hydroponics is therefore translated as the work of water. Subsequently, it is defined as a soil-less cultivation technique in which plants grow in the presence or absence of substrate and are supplied with a nutrient solution containing all the essential elements they need for normal growth and development (Resh, 1978; Tomson, 2019).

Hydroponics is a type of cultivation that was practiced early in human history. Indeed, the gardens of Babylon are already considered the first hydroponic systems. Moreover, some hieroglyphs in Egypt represent a type of cultivation in water. The floating gardens of the Aztecs in Mexico and those in China are also good examples (Resh, 1978). However, the term hydroponics did not appear until 1930 when W.F. Gericke introduced this type of cultivation on an industrial production scale. Little known for many years, this technique spread throughout the world thanks to the automation and development of the plastic industries but also by the awareness of the limits of current agricultural techniques (Jijakli, 2018).

In developed countries, due to population growth, intensive agriculture is favoured to increase agricultural yields. Nevertheless, land use increases the consumption of energy, water, and fertilizers. In addition, intensive farming accelerates soil salinization and soil erosion leading to the loss of natural fertility (Dumitrescu, 2013). In developing countries, one of the major problems is food insecurity and malnutrition (Croft et al., 2017). The latter is exacerbated by increasing urbanization and a lack or shortage of safe water (FAO et OMS, 2003). As a result, traditional farming systems are no longer enough to meet people's needs.

In both situations, hydroponics has many advantages. Indeed, it enables a more efficient use of resources such as water and nutrients and high planting density (Resh, 1978; Uchimura et al., 2014; Foucard et Tocqueville, 2019). In addition, it represents an interesting alternative for freeing oneself from the soil, particularly in uncultivated areas of the world (Resh, 1978; Foucard et Tocqueville, 2019). When land availability is compromised due to soil physicochemical properties unsuitable for cultivation, this technology makes it possible to produce vegetables regardless of pedo-climatic conditions, presence of pests and diseases, weeds, ... (Charoenpakdee, 2014; Ansari et al., 2015). Moreover, hydroponics makes it possible to dispense with conventional practices such as ploughing and weeding. It therefore simplifies cultivation techniques by increasing yield (Foucard et Tocqueville, 2019). These systems thus provide a response to future needs for food security as well as to changes in land use (Dumitrescu, 2013).

Nevertheless, hydroponics also has its limits. On the one hand, the implementation of this technique is constrained by the availability of freshwater but also access to nutrients (Resh, 1978). Indeed, soil-less crops use nutrient solutions rich in mineral fertilizers that are used in large quantities to meet plant needs. Regular draining is practised because nutrient concentrations change according to the rate of absorption of ions by the plants (Foucard et Tocqueville, 2019). Although this drainage water is reused to create new nutrient solutions, this technique generally requires high initial investment costs and advanced knowledge in fertilization (Jijakli, 2018; Foucard et Toucqueville, 2019). In addition, excessive recycling leads to an accumulation of minerals, organic acid and plant exudates, which can ultimately be toxic to plants (Foucard et Tocqueville, 2019). It is therefore essential to regularly check the pH and concentration of the solution in order to prevent it from becoming phytotoxic (Jijakli, 2018). On the other hand, hydroponics is highly dependent on fossil energy sources. The latter are used in the synthesis of simple nitrogenous chemical

fertilizers or other complex mineral salts. In addition, hydroponics makes extensive use of plastics (Foucard et Tocqueville, 2019). Other disadvantages include rapid spread of certain diseases throughout the crop, dependence on pump problems and increased root sensitivity to temperature and salt stress (Resh, 1978; Jijakli, 2018).

To describe hydroponic systems, two characteristics are essential: the type of system used and the fertilization. There are five commonly widespread cropping systems in the world: Drip system, NFT, rafting, Ebb and Flow, and aeroponics (Resh, 1978; Jijakli, 2018). As far as fertilization is concerned, two types of fertilizers can be used: mineral fertilizers and organic fertilizers. The concept of organic fertilizer used in hydroponic systems is called "**BIOPONICS**".

2.1.1 Hydroponic systems types

Different types of production systems, with or without substrate, are used in hydroponics. The five most used systems in the world are: Deep-water culture, NFT, Drip system, Ebb and Flow, and aeroponics (Resh, 1978; Jijakli, 2018).

In NFT (Nutrient Film Technique) systems, the roots grow in gutters, usually made of white plastic, where a thin layer (1 or 2 cm) of nutrient solution flows. This thin layer creates permanent movement of the solution resulting in a particularly good oxygenation of the solution (Resh, 1978; Jijakli, 2018). NFT is one of the most widespread techniques for the production of leafy greens (Figure 1a).

The Deep-water or raft culture consists of a table, regulated by an overflow, filled with nutrient solution. The plants are installed on perforated plates placed on the surface of the table. The roots are thus immerged in nutrient solution (Jijakli, 2018). The water or air pumps are generally used to create water movement and oxygenation of the nutrient solution. This system is not common in European hydroponic farms, but is quite popular in aquaponic farms working with different vegetable species (Figure 1b).

In Drip systems, the substrate serves as a support for the development of the plant. During its intermittent irrigation, the substrate retains the nutrient solution within it. The voids created by the plant by absorption of this solution allow the oxygenation of the latter (Jijakli, 2018). Drip cultivation is the most widespread technique for the production of fruit vegetables (Figure 1c).

The Ebb and Flow system consists of a table, filled with substrate or pots containing the substrate, flooded by the nutrient solution, and drained alternately (Jijakli, 2018). This system is not extensively used in professional hydroponics but is very popular in small and middle scale aquaponics (Figure 1d).

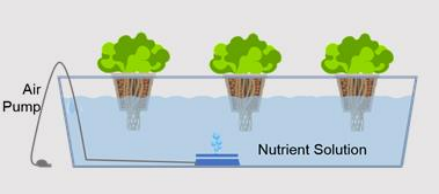
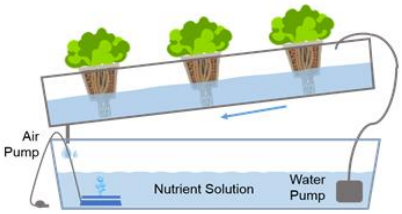
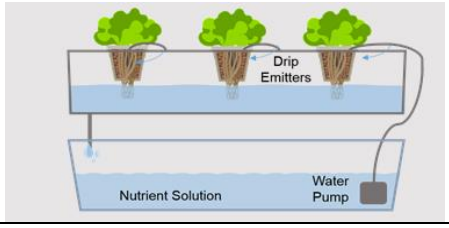
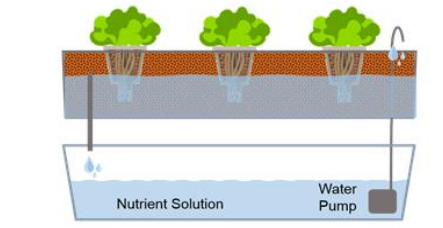
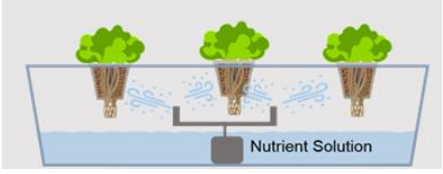
Finally, the term aeroponics is used when the nutrient solution is sprayed onto the roots as a "nutrient mist" (Resh, 1978; Jijakli, 2018) (Figure 1e).

Figure 1: Hydroponics systems; a) NFT systems (Brooke, 2015); b) Deep water systems (Grant, 2018); c) Drip systems (Green and Vibrant, 2019); d) Ebb and Flow systems; e) Aeroponics systems 2020)

Different culture media can also be used, such as water, foam, gravel, rockwool, sand, sawdust, peat, coco coir, perlite, pumice, peanut hulls, polyester matting, or vermiculite (Resh, 2013).

Table 1 lists the advantages and disadvantages of each technique.

Table 1: Comparison between the different systems used in hydroponics (Soilless, 2016)

Hydroponic Systems	Pros	Cons
Deep Water Culture : 	• Cheapest of the active systems • Simple set up • Can work without water pump • Reliable	• Risk of root rot if not cleaned regularly • Slower growth rate • Must top water until roots are long enough to fall into the nutrient solution • Must frequently refill reservoir
Nutrient-Film Technique: 	• Excess nutrient solution recirculates • Plentiful oxygen flow • Space efficient	• Prone to clogging • Technical malfunctions could result in crop loss
Drip Method: 	• Excess nutrient solution recirculates • Enough oxygen flow	• Prone to clogging • Prone to algae growth • Requires regular cleaning
Ebb and Flow: 	• Affordable • Low maintenance • Excess nutrient solution recirculates	• Prone to algae growth • Technical malfunctions could result in crop loss
Aeroponics: 	• Maximum nutrient absorption • Excess nutrient solution recirculates • Plentiful oxygen flow • Space efficient	• Prone to clogging • Technical malfunctions could result in crop loss • High-tech • Time intensive • Poorly suited to thick organic-based nutrients & additives

2.2 FERTILIZATION

2.2.1 Introduction

This chapter will focus on plant fertilizations, which is particularly important in hydroponics (Resh, 1978). It is divided into four main parts. The first part will define the essential elements and describe more particularly nitrogen, phosphorus, and potassium. It will also give an idea of the needs of different types of plants. Finally, it will briefly describe the consequences of a nutrient deficiency. The second part will deal with parameters such as pH and EC that allow good fertilisation management in hydroponic systems. The third part will focus on the origin of nutrients. Finally, the last part will try to list the limits of inorganic fertilization.

2.2.2 Nutrients

2.2.2.1 Essential elements

The plant is composed of 80 to 95% water. More than 90% of the dry weight of the plant matter is made up of three elements: carbon, oxygen, and hydrogen. The remaining 10% represents the other elements. A total of 60 different elements are contained in the different plant species. However, many of them are not considered essential (Resh, 1978).

To be considered essential, an element must meet three criteria. Firstly, the plant cannot complete its life cycle in the absence of such an element. In addition, the action of the element must be specific. No other element can therefore replace it. Finally, the element must be directly involved in the nutrition of the plant, i.e. be an essential component of an essential metabolite. It may also be necessary for the action of an essential enzyme. However, it cannot simply make another element more readily available or counteract the toxic effect of another element (Resh, 1978; Dumitrescu, 2013).

In the end, only 16 elements are considered essential. They are divided into two categories: macroelements and microelements. Macroelements, also called macronutrients, are needed in relatively large quantities. They include carbon, hydrogen, oxygen, nitrogen, phosphorus, sulphur, potassium, magnesium, and calcium. On the other hand, the plant needs only small quantities of the microelements or trace elements. These include iron, chlorine, manganese, boron, zinc, copper and molybdenum (Resh, 1978). Table 2 lists the various essential elements, their plant-available forms, and their percentages in dry plant tissues.

Table 2: Essential elements and their plant-related characteristics (Resh, 2013)

Element	Symbol	Available Form	Atomic Weight	Parts per Million (ppm)	Concentration in Dry Tissue (%)
Macronutrients					
Hydrogen	H	H_2O	1.01	60,000	6
Carbon	C	CO_2	12.01	450,000	45
Oxygen	O	O_2, H_2O	16.00	450,000	45
Nitrogen	N	NO_3^-, NH_4^+	14.01	15,000	1.5
Potassium	K	K^+	39.10	10,000	1.0
Calcium	Ca	Ca^{2+}	40.08	5,000	0.5
Magnesium	Mg	Mg^{2+}	24.32	2,000	0.2
Phosphorus	P	$H_2PO_4^-$, HPO_4^{2-}	30.98	2,000	0.2
Sulfur	S	SO_4^{2-}	32.07	1,000	0.1
Micronutrients					
Chlorine	Cl	Cl^-	35.46	100	0.01
Iron	Fe	Fe^{3+}, Fe^{2+}	55.85	100	0.01
Manganese	Mn	Mn^{2+}	54.94	50	0.005
Boron	B	BO_3^{2-}, $B_4O_7^{2-}$	10.82	20	0.002
Zinc	Zn	Zn^{2+}	65.38	20	0.002
Copper	Cu	Cu^{2+}, Cu^+	63.54	6	0.0006
Molybdenum	Mo	MoO_4^{2-}	95.96	0.1	0.00001

In addition to being grouped into macro and micro elements, essential elements can also be sorted according to their mobility. So-called mobile elements can be transferred from their original site to the region of active growth. When there is a deficiency of these elements, the first symptoms appear on the older leaves on the lower part of the stem. Mobile elements include magnesium, phosphorus, potassium, zinc, nitrogen, and molybdenum. On the other hand, the so-called immobile elements cannot be transferred. They remain on their original site, i.e. the oldest leaves. If one of these elements is missing, symptoms then appear first on the young leaves before spreading throughout the plant. The constituent elements in this category are calcium, iron, sulphur, boron, copper and manganese (Resh, 2013).

The main elements studied in this work are nitrogen, phosphorus, and potassium.

2.2.2.1.1 Nitrogen

Nitrogen is used by the plant for its vegetative and reproductive growth (Cáceres et al., 2015). It is used for the synthesis of many necessary organic compounds such as amino acids, proteins, enzymes, nucleic acids, and vitamins. It is also one of the compounds of chlorophyll. It is therefore involved in the proper functioning of photosynthesis (Resh, 1978; Tremblay et al., 2004). In addition, plant alkaloids are of major social and economic importance since they have enabled the pharmaceutical industry to flourish (Tremblay et al., 2004).

Nitrogen is found in different forms: dinitrogen (N_2), ammonia (NH_3), ammonium (NH_4^+), nitrite (NO_2^-), nitrate (NO_3^-) and amino acids (2.3.2.1 The nitrogen cycle). However, plant roots can absorb nitrogen in the NO_3^-, NH_4^+ or amino acids configuration but NO_3^- is the nitrogen form the more easily absorbed by plants (Tremblay et al., 2004; Shinohara et al., 2011).

The physiological role of nitrogen is no more important than the other essential elements. Nevertheless, the plant needs it in much larger quantities (Tremblay et al., 2004). The forms of nitrogen present in nutrient solutions for hydroponics are (Resh, 1978):

- Ammonia (NH_3)
- Nitrate (NO_3^-)
- Potassium nitrate (KNO_3)
- Calcium nitrate ($Ca(NO_3)_2$)
- Ammonium sulfate ($(NH_4)_2SO_4$)
- Ammonium nitrate (NH_4NO_3)
- Diammonium phosphate ($(NH_4)_2HPO_4$)

2.2.2.1.2 Phosphorus

Phosphorus is a particularly important macroelement for the plant. It constitutes the adenylates which are involved in ADP and ATP providing the energy necessary for the plant's synthesis reactions. It also forms phospholipids, structural components of membrane lipides (Resh, 1978; Lachapelle, 2010; Möller et al., 2012). In addition, it is used by the plant to produce sugar phosphates, nucleic acids but also enzymes involved in photosynthesis and respiration (Resh, 1978; Lachapelle, 2010).

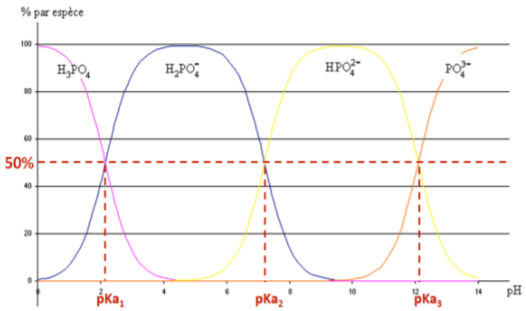


Figure 2: Percentage of the main forms of P as a function of the pH of the solution (Aix-Marseille Université, 2014)

In soil cultivation, phosphorus is often the limiting factor to plant growth. Unlike nitrogen, phosphorus does not have an atmospheric reservoir. In addition, bioavailable phosphorus is present in solution in the form of ions. However, phosphorus is, for the most part, absorbed by soil particles (Beaudin et al., 2008). It is therefore not very mobile and is gradually released by the degradation of soil and rocks (Lachapelle, 2010).

In the soil solution, phosphorus is found in the form of monovalent ($H_2PO_4^-$) and divalent (HPO_4^{2-}) orthophosphate anion. At pH close to neutral (7.2), the proportion between these two forms is 50/50. $H_2PO_4^-$ is the only form present at lower pH (4-6). In contrast, at basic pH (8), $H_2PO_4^-$ is only present at 20% while the remaining 80% is in the HPO_4^{2-} form (Figure 2) (FAO, 2004).

The form of phosphorus preferentially assimilated by the plant is $H_2PO_4^-$. However, other forms of phosphorus are sometimes present in nutrient solutions used in hydroponics (Resh, 1978):

- Phosphoric anhydride (P_2O_5)
- Phosphate (PO_4^{-3})
- Monopotassium phosphate (KH_2PO_4)
- Diammonium phosphate ($(NH_4)_2HPO_4$)
- Phosphoric acid (H_3PO_4)

2.2.2.1.3 Potassium

Unlike nitrogen and phosphorus, potassium is not a constituent of synthetic compounds (Lachapelle, 2010). It therefore does not form a stable structural part of molecules within plant cells (Resh, 1978). On the other hand, it does play a role in osmoregulation and thus in the water status of the plant. It also serves as a proton pump. It is therefore involved in ionic membrane exchanges. In addition, it plays a role in pH balance. Potassium also acts as co-enzymes and activators of many enzymes including those related to respiratory activity. Protein synthesis also requires a high level of potassium (Resh, 1978; Lachapelle, 2010). The form of potassium available to the plant is K^+. In nutrient solutions, it is present in solution in the form of salt (Resh, 1978):

- Potash (K_2O)
- Potassium nitrate (KNO_3)
- Monopotassium phosphate (KH_2PO_4)
- Potassium chloride (KCl)
- Potassium sulfate (K_2SO_4)

2.2.2.2 Unit and notations

The quantity of elements present in nutrient solutions is generally expressed in ppm (parts per million) or ppb (parts per billion) (Resh, 1978). This unit is not a concentration but a ratio. Concerning the notation, a ppm is a fraction of 10^{-6} while a ppb is 10^{-9}. This unit is used to express a mass ratio (mg/kg) or a volume ratio (µL/L). However, in solution, this unit is related to a concentration since 1 litre of water is equivalent to 1 kilogram. In this case, one ppm can then be equivalent to 1 mg/L.

The following notations will henceforth be applied:

- $N\text{-}NH_4^+$, $N\text{-}NH_3$, $N\text{-}NO_2^-$, $N\text{-}NO_3^-$, …: this notation is used to express the concentration of nitrogen present in NH_4^+, NH_3, NO_2^-, NO_3^-, ... The conversions between these parameters are as follows (HACH, 2020):
  - $NH_4^+ = 1.28 \times N\text{-}NH_4^+$
  - $NH_3 = 1.22 \times N\text{-}NH_3$
  - $NO_2^- = 3.28 \times N\text{-}NO_2^-$
  - $NO_3^- = 4.43 \times N\text{-}NO_3^-$
- TIN: Total inorganic nitrogen is the sum of $N\text{-}NH_3$, $N\text{-}NO_2^-$ and $N\text{-}NO_3^-$ (WERF, 2015)
- TAN: Total ammonia nitrogen is the sum of NH_3 and NH_4^+ (WERF, 2015)

2.2.2.3 Plant needs

The ideal formulation for a fertilizer depends on the species but also on the variety, the growth stage and the part of the plant that will be harvested for consumption (leafy greens, aromatic and fruit vegetables) (Resh, 1978; Dumitrescu, 2013). For example, leafy greens have a higher nitrogen demand than fruit plants.

They can tolerate a high level of nitrogen which favours their plant growth. In contrast, fruit plants consume more phosphorus, potassium, and calcium (Resh, 1978).

As the N:P:K ratio varies from one plant to another, optimal conditions for leafy vegetables and fruit vegetables may be different (Table 3).

Table 3: Optimal pH, EC and nutrients for different fruit and leaf vegetables, strawberries and cut flowers (van der Lugt et al., 2016)

		Fruit vegetables and strawberries						Leaf vegetables	
	Type of substrate[2]	Inert substrate						Water	
	Unit	Cucumber	Eggplant	Melon	Strawberries	Sweet pepper	Tomato	Basil	Lettuce
pH	/	**5,2-6,0**	**5,5-6,0**	**5,5-6,0**	**5,5-6,0**	**6**	**5,5-6,0**	**5,5-6,0**	**5,5-6,0**
EC	mS/cm	**3**	**3**	**3**	**1**	**3**	**4**	**4**	**4**
Na	ppm	<184	<184	<184	<184	<184	<184	<184	<138
Cl		<284	<284	<284	<284	<284	<284	<284	<213
HCO3		<6	<6	<6	<6	<6	<6	<6	<6
$N\text{-}NH_4^+$		**<2**	**<2**	**<2**	**<2**	**<2**	**<2**	**<2**	**<2**
K		**313**	**242**	**274**	**196**	**433**	**313**	**274**	**235**
Ca		260	248	280	180	400	400	280	240
Mg		73	109	61	49	109	109	61	49
$N\text{-}NO_3^-$		**252**	**280**	**280**	**168**	**308**	**308**	**280**	**196**
S		112	96	112	80	218	218	112	64
P		**28**	**28**	**25**	**22**	**31**	**31**	**25**	**62**
Fe	ppb	1680	1400	1400	1960	1960	1960	1400	2240
Mn		385	385	275	385	275	275	275	440
Zn		458	458	458	458	458	458	458	523
B		540	864	540	216	540	540	540	540
Cu		95	44	64	44	44	44	64	95
Mo		48	48	48	48	48	48	48	144

2.2.2.4 Nutritional deficiencies

In cases of nutritional deficiency, symptoms can be characterized by chlorosis (yellowing) or necrosis (browning). It is important to identify this deficiency early to react before the symptoms spread to the whole plant causing its death. In addition, the lack of one element affects the plant's ability to accumulate the other elements. There is therefore a simultaneous deficiency of several elements. In this case, it becomes difficult, if not impossible, to visually identify which elements are deficient and responsible for the symptoms (Resh, 1978).

In the case of nutritional deficiencies, the first step is to identify and describe the symptoms as well as the organs that are affected. Table 4 lists the three main NPK elements and associated symptoms in the case of deficiencies and toxicities.

[2] The types of substrates in this table are those most commonly used for each plant species. Fruit vegetables are therefore mainly grown on inert substrates while leaf vegetables are grown in pure hydroponics (only in water).

Table 4: Deficiency and toxicity symptoms for N, P and K (Resh, 1978; FAO, 2004)

	Deficiency Symptoms	Toxicity Symptoms
Nitrogen	Growth is restricted and plants are generally yellow (chlorotic) from lack of chlorophyll, especially older leaves. Younger leaves remain green longer. Stems, petioles and lower leaf surfaces of corn and tomato can turn purple.	Plants usually dark green in colour with abundant foliage but usually with a restricted root system. Potatoes form only small tubers and flowering, and seed production can be retarded
Phosphorus	Plants are stunted and often a dark green colour. Anthocyanin pigments may accumulate. Deficiency symptoms occur first in more mature leaves. Plant maturity often delayed	No primary symptoms yet noted. Sometimes copper and zinc deficiency occur in the presence of excess phosphorus
Potassium	Symptoms first visible on older leaves. In dicots, these leaves are initially chlorotic but soon scattered dark necrotic lesions (dead areas) develop. In many monocots, the tips and margins of the leaves die first. Potassium-deficient corn develops weak stalks an is easily lodged.	Usually not excessively absorbed by plants. Oranges develop coarse fruit at high potassium levels. Excess potassium may lead to magnesium deficiency and possible manganese, zinc or iron deficiency

It is also important to verify that the source of the symptoms is indeed a nutritional imbalance. Indeed, many other problems can affect the health of plants such as the presence of insects or diseases related to pests, damage caused by pesticides or pollution, water stress, lack of light or a non-ideal temperature. In addition, different species have varying degrees of sensitivity to nutritional deficiencies (Resh, 1978).

To prevent nutritional deficiencies or excesses and thus avoid stress that would limit the growth of the plant, it is important to monitor the nutritional level. The ideal method to diagnose nutrient deficiencies is the periodic analysis of plant leaf tissue coupled with the analysis of the nutrient solution. In fact, there is a correlation between these two analyses which allows the identification of deficiencies and thus the adjustment of the nutrient solution if necessary. However, this technique is often too slow and expensive (Resh, 1978). In practice, different tools are therefore used, such as the ECmeter and the pHmeter (Boudreault, 2018).

2.2.3 Nutrient solution management

2.2.3.1 Hydrogen potential (pH)

The pH, or Hydrogen Potential, is the measure of the H^+ concentration. However, it is commonly referred to as a measure of acidity. This value varies between 0 and 14. A high pH, between 7.1 and 14, corresponds to a low H^+ concentration. The pH is then alkaline or basic. Between 0 and 6.9, the H^+ concentration is high, and the pH is said to be acidic. PH 7 corresponds to neutrality (Resh, 1978; Fogliani, 2016; Requasud, 2019).

In fertilisation, the pH plays an important role in the availability of elements. In fact, these elements have a pH range where their availability for plants is at its highest. On the contrary, certain elements are not available in certain pH ranges (Figure 3).

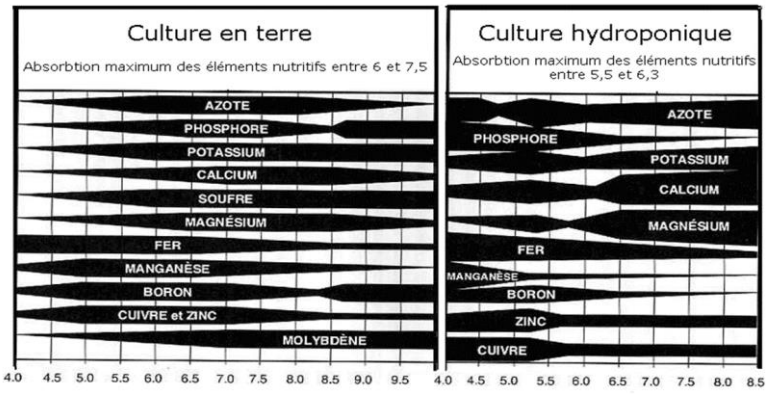


Figure 3: Availability of elements as a function of pH for soil and hydroponic crops (Jijakli, 2018)

In a closed system, the pH changes in response to the degradation of elements by plants and thus affects the balance of anion and cation uptake (Resh, 1978). Indeed, pH is influenced by root activity which releases protons for positive ion uptake (Stalport, 2017). The system therefore has a natural tendency to acidify. To control this change, a pH meter is used and the pH of the nutrient solution is regulated by adding a base (KOH, SiO_2) and an acid (HNO_3, H_3PO_4, H_2SO_4, $C_6H_8O_7$, CH_3COOH) (Resh, 1978; Lambert, 2000; GHE, 2020).

The pH of a soil can vary from 4 to 9. The ideal pH for plant growth varies according to the type of plant and its needs. It is generally between 6 and 7.5 (Resh, 1978; Fogliani, 2016; van der Lugt et al., 2016). In hydroponics, the optimal pH is between 5.5 and 6.3 (van der Lugt et al., 2016; Jijakli, 2018). However, a higher pH, between 6.5 and 8, is observed in aquaponics and does not seem to be a problem (Foucard, 2018). Following these values, there is some doubt about the ideal pH range in aquaponic systems.

2.2.3.2 Solubility and Electroconductivity (EC)

Solubility is a measure of the concentration of salts that remain in solution when dissolved in water. In hydroponics, the solubility of the elements is very crucial because only solubilized elements are available to the plant (Resh, 1978).

Electrical conductivity, or electroconductivity (EC), is the measurement of the ionic concentration of a nutrient solution. It is measured in mS/cm using a conductivity meter. A conductivity of 0 mS/cm means that there are no salts. The higher the conductivity, the more saline the solution becomes and the easier the current is conducted through the solution. The EC therefore determines the rate at which the nutrient solution becomes too concentrated. Conductivity is different for each crop. It varies according to the stage of growth. However, this parameter is generally between 2 and 4 to be optimal for production (Figure 4) (Resh, 1978; Fogliani, 2016). EC is represented by the following formula (Carmassi et al., 2003):

$$EC = 0 + 0.78\,[Na] + 0.28\,[K] + 0.04\,[Mg] + 0.06\,[Ca]$$

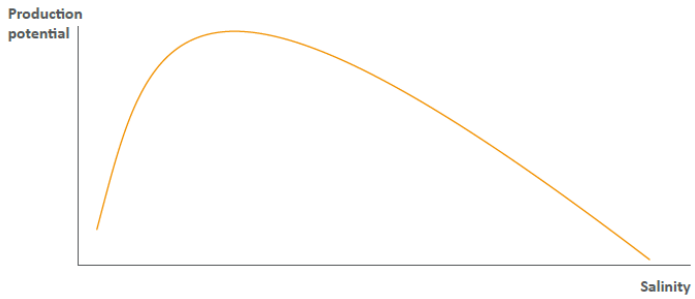


Figure 4: Potential production as a function of solution salinity (van der Lugt et al., 2016)

A low conductivity induces that the water present in solution will enter the roots with the aim of diluting the cellular concentration of ions. The plant is then deficient in certain elements. There are thus severe negative effects on plant growth, health and yield (Vitre, 2003; Peiris et al., 2015; Fogliani, 2016).

On the other hand, a high conductivity will lead to an osmotic pressure which will push the water outside the roots to tend towards a dilution of the external salt concentration. The absorption of water by the roots is then constrained. In addition, the plant is in the presence of an excess of mineral salts that can lead to phytotoxicity (Vitre, 2003; Fogliani, 2016).

Conductivity therefore plays an essential role in a plant's productive potential.

2.2.3.3 Application of mineral fertilizers

In hydroponics, fertilisation is generally composed of 2 or 3 concentrated solutions: one solution A, one solution B and possibly one solution C. These solutions are composed of different salts. The choice of these salts depends on various factors such as their solubility, cost, formulation, etc... (Resh, 1978). The quantity of each solution to be diluted in water depends mainly on the properties of the water and the cultivated species.

It is also important to respect a specific proportion of each solution. There are applications, such as HydroBuddy, which can calculate the ideal proportions of the three solutions according to the salts present to achieve a precise concentration for the different elements (Fernandez, 2020). Some fertiliser manufacturers supply ready-made solution mixtures. They then divide their fertilizer into two categories: a first mixture to be used for flowering and another mixture for fruiting (GHE, 2020).

In all cases, the amount of NPK present in the solutions is presented in the form of three-digit codes. They represent respectively the percentage of nitrogen, phosphorus, and potassium present in the solution (Reece et al., 2012).

2.2.4 Origins of nutrients

Hydroponic systems mainly use fertilizers produced in chemical industries based on minerals and fossil fuels (Dumitrescu, 2013). Many of the nutrients used in these fertilizers come from mineral deposits, but other sources are also possible. Fertilizers consist mainly of 3 elements: nitrogen (N), phosphorus (P) and potassium (K).

The main source of nitrogen on Earth is the atmosphere. However, nitrogen in the gaseous form N_2 cannot be used by plants. The Haber-Bosch process makes it possible to produce NH_3 from N_2 and H_2. However, this process is very energy intensive. Although N_2 is available directly in the atmosphere, H_2 is produced from natural gas, coal, or heavy oils. The Ostwald process produces HNO_3 from NH_3. During this process, only half of the NH_3 is converted into HNO_3 by oxidation. The other half is transformed into NH_4NO_3. The process also produces N_2O and is therefore associated with greenhouse gas emissions (Dumitrescu, 2013).

Phosphorus comes from rock phosphate (RP). RP includes all-natural minerals with a high concentration of phosphate minerals. On the one hand, sedimentary deposits account for about 80-90% of the world's production of RP. They are found mainly in Morocco and other African countries, the United States, the

Near East and China. On the other hand, igneous deposits account for the remaining 10 to 20%. They are found in Russia, Canada, South Africa, Brazil, Finland, Zimbabwe, Uganda, Malawi, Sri Lanka, ... (FAO, 2004). Finally, more than 75% of the world production of rock phosphate is located in 4 countries: China, the United States, Russia and Morocco and Western Sahara (FAO, 2004; U.S Geological Survey, 2020). The distribution of this rock is therefore unequal around the world and poses problems of inequality leading to political instability (Dumitrescu, 2013).

Ultimately, potassium comes from potash that is harvested by conventional well mining or solution mining (Dumitrescu, 2013). Potash is the generic term for all salts extracted with potassium elements in water-soluble form. In agriculture, potash represents all potassium fertilizers including potassium chloride, potassium sulphate, potassium magnesium sulphate or langbeinite. 78% of world potassium production is shared between Canada, Belarus, Russia, China, Germany and Jordan (U.S Geological Survey, 2020).

2.2.5 Limits of inorganic fertilizers

Table 5: Quantification of environmental impacts related to commercial nutrient solution production (Dumitrescu, 2013)

Impact category	Unit	N – $Ca(NO_3)_2$	N – NH_4NO_3	N – KNO_3	K_2O – KNO_3	P_2O_5 – H_3PO_4*
Global warming (GWP100)	kg CO_2 eq/kg	3.836	8.500	15.878	0.865	2.018
Ozone layer depletion (ODP)	kg CFC_{11} eq/kg	6.27 e-07	5.75 e-07	9.15 e-07	1.57 e-07	2.08 e-07
Photochemical oxidation	kg C_2H_4 eq/kg	0.002	0.002	0.002	0.0005	0.002
Acidification	kg SO_2 eq/kg	0.011	0.023	0.040	0.002	0.031
Eutrophication	kg PO_4 eq/kg	0.004	0.010	0.016	0.001	0.045
Non renewable, fossil	MJ eq/kg	63.544	57.950	88.592	15.210	32.190

Source: Ecoinvent 2013

*Note: The values refer to the production of phosphorus pentoxide (P_2O_5), the extracted salt which is then hydrated to form phosphoric acid (H_3PO_4)

Many negative effects are linked to the production of inorganic fertilizers. Indeed, in addition to using energy-intensive processes, the mineral resources that enable the creation of these fertilizers are overexploited and are destined to disappear. In order to illustrate the pollution linked to the production of fertilizers based on the three main macronutrients (NPK), Dumistrescu (2013) carried out a life cycle analysis assessment of commercial solutions including the production and transport processes (Table 5). In this life cycle analysis, the pollution generated by the production of fertilizers is the greatest for nitrogen compounds compared to the pollution generated by the production of potassium and phosphorus. In addition, the proportion of fertilizers not used by plants is difficult to recycle and therefore contributes to environmental pollution and the destruction of ecosystem balance through over-enrichment of the environment leading to eutrophication (Tomson, 2019). These negative effects are mainly attributed to N and P. Indeed, in comparison, micronutrients are in too small concentrations to have a predominant environmental impact (Dumitrescu, 2013).

The use of inorganic fertilizers is therefore not a long-term sustainable solution as it causes pollution and is a threat to health (Quaik et al., 2012).

2.3 Organic Fertilizers

2.3.1 Introduction

In light of the disadvantages of using chemical fertilisers in hydroponics, bioponics is one of the solutions envisaged. According to Fang et al. (2018), bioponics is defined as "A contained and controlled growing system in which plants in growing media derive nutrients from plant-based, animal-based and mineral natural substances which are released by the biological activity of microorganisms".

The use of organic fertilisers is therefore an environmentally sustainable solution. Indeed, faced with the challenge of waste management, organic fertilisers would reduce the waste associated with the manufacture and use of chemical fertilisers (Ansari et al., 2015). Moreover, 1/3 of the world's food production (about 1.3 trillion tonnes) for human consumption is lost or discarded. Recycling it for the manufacture of organic fertilizers is one of the possible solutions for moving towards a circular economy (Stocknes et al., 2016). Many other wastes could be recovered in this way, such as animal excreta or other types of green waste (composts, etc.). Biological treatment is therefore a sustainable waste treatment strategy that allows waste stabilisation and nutrient creation (Abdullahi et al., 2008). Moreover, because of its non-dependence on synthetic fertilizers, bioponics offers its practitioners resilience to supplies of fertilizers.

Second, bioponics has many advantages in terms of the quality of food produced. According to Ansari et al. (2015), organic fertilizers have the property of increasing the quantity and quality of a product, unlike chemical fertilizers which increase quantity at the expense of quality. Indeed, the use of organic fertilizers increases hair growth on roots. The latter allows for better resistance to certain diseases, particularly in lettuce (Fang et al., 2018; Hsieh et al., 2019). They also create space for bacterial growth, resulting in better nutrient uptake (Fang et al., 2018). In addition, many people are increasingly concerned about what they eat. The use of organic fertilizers can therefore address these concerns.

Organic fertilizers also have economic benefits. In the context of resource depletion and crisis economics, they reduce production costs, energy consumption and the production of greenhouse gases (Wang et al., 2018).

Finally, in developing countries, access to inorganic nutrient solutions is limited. Indeed, about one-third of the fertilizers used in developing countries are imported, and foreign exchange shortages, inefficient distribution systems and infrastructure constraints within countries affect their cost and availability. In addition, many countries that use a lot of chemical fertilizers are aware of the environmental problems associated with intensive fertilizer use. Organic fertilization is therefore of great interest to developing countries.

Despite these many advantages, bioponics also has constraints. Indeed, organic fertilizers, unlike inorganic fertilizers, need the presence of bacteria to be properly absorbed by plants but also to avoid being phytotoxic to the plant (Shinohara et al., 2011). This use of living organisms makes the system more fragile and therefore more demanding in terms of maintenance and management time. In addition, it requires effort and knowledge to optimise it. Indeed, one of the drawbacks of this technique is the lack of certain essential elements for the plant (Dumitrescu, 2013). For example, in aquaponics, a particular bioponics system, potassium deficiencies are quite frequent (Harlaut, 2015).

This chapter will therefore focus on bioponics. Organic fertilisers have been available as commercial products for a long time, but more and more people around the world are working on recycling organic matter into nutrient solutions. Many processes, such as aquaponics, are being implemented and many different types of organic matter are being used. So, it is not a case of “one size fits all”. This chapter will therefore present the biological cycles of major elements (NPK). It will then describe the nutritional characteristics of several organic materials that can be used in bioponics and give some examples of processes for the mineralisation of organic matter. Finally, examples of known bioponics will be described.

2.3.2 The biological cycles of major elements

2.3.2.1 The nitrogen cycle

Nitrogen is present in large quantities in the atmosphere in the form of gaseous N_2. However, in this form it is inaccessible to most organisms. To be available to primary producers such as plants, nitrogen is converted to NH_3. In addition to these two forms, nitrogen is present in many other forms, including inorganic (NH_3, NO_3^-, ...) but also organic forms (amino acids and nucleic acids). To change form, nitrogen undergoes numerous transformations (Figure 5) (Bernhard, 2010):

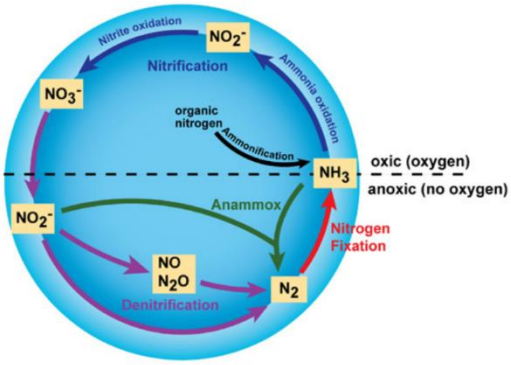


Figure 5: The nitrogen cycle (Bernhard, 2010)

- Nitrogen fixation
- Nitrification
- Anammox
- Denitrification
- Ammonification

These different transformations, based on oxidation reactions (elimination of electrons) and reduction reactions (addition of electrons) form the nitrogen cycle, which depends on the activity of the micro-organisms (Delwiche, 1970).

2.3.2.1.1 Nitrogen fixation

Nitrogen fixation is the conversion of N_2 into biologically available nitrogen. The nitrogen atoms contained in the N_2 are linked by a triple bond which makes the compound very stable. To break this bond, 8 electrons and 16 ATP molecules are required. Therefore, only a small group of prokaryotes can carry out this energetically demanding process. The physiological and phylogenetic diversity among the organisms that fix nitrogen is important: aerobic or anaerobic, phototrophic, or chemotrophic, ... However, nitrogenase, an enzyme complex, is present in all these organisms. It catalyses the reduction of N_2 to NH_3 ((Bernhard, 2010).

$$N_2 + 8H^+ + 8e^- \rightarrow 2NH_3 + H_2$$

2.3.2.1.2 Nitrification

Nitrification is a process that transforms NH_3 into NO_2^- and then into NO_3^-. It is carried out aerobically and is catalysed by autotrophic microorganisms. Two distinct stages can be identified and are carried out by different types of microorganisms. Firstly, ammonia oxidizing bacteria (AOB) convert NH_3 into NO_2^- via the intermediate hydroxylamine, a process that requires two different enzymes, ammonia monooxygenase and hydroxylamine oxidoreductase:

$$1)\ NH_3 + O_2 + 2e^- \rightarrow NH_2OH + H_2O$$
$$2)\ NH_2OH + H_2O \rightarrow NO_2^- + 5H^+ + 4e^-$$

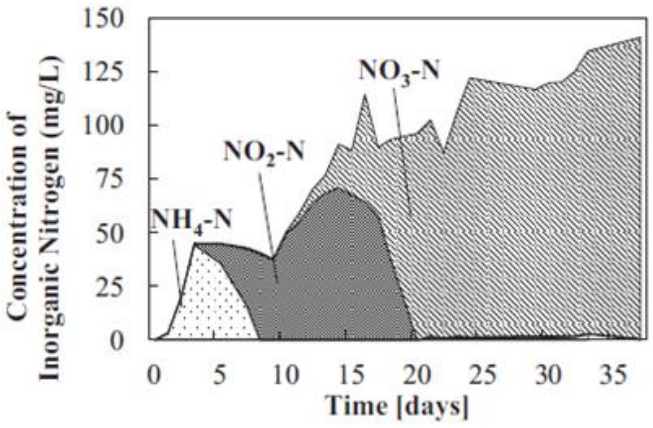


Figure 6: Diagram of the nitrification process (Uchimura et al., 2014)

This phase is called nitrosation. Unlike nitrogen fixation, which is carried out by many different types of microbes, nitrosation is less widely distributed among prokaryotes. Only bacteria of the types Nitrosomonas, Nitrosospira and Nitrosococcus perform this process (Van Bochove, 1993; Bernhard, 2010; Cáceres et al., 2015; Eck et al., 2019).

The NO_2^- is then oxidized to NO_3^- during the nitration phase (Van Bochove, 1993; Cáceres et al., 2015; Eck et al., 2019). This step is carried out by a separate group of prokaryotes, known as nitrite-oxidizing bacteria including Nitrospira, Nitrobacter, Nitrococcus and Nitrospina (Bernhard, 2010):

$$NO_2^- + \frac{1}{2}O_2 \rightarrow NO_3^-$$

For complete nitrification, oxidation of NH_3 and NO_2^- must take place (Figure 6). The efficiency of these reactions can be as high as 97.6% (Shinohara et al., 2011). These processes require the correct temperature and pH, a sufficient supply of dissolved oxygen (DO) and a good contact ratio between substrate and biomass. Nitrifying microorganisms use inorganic C as a source of carbon. The nitrification reaction therefore consumes alkalinity. The pH therefore decreases during the reaction (Cáceres et al., 2015). The nitrification process is best carried out at a pH between 6.6 and 8 and at temperatures between 30 and 35°C.

2.3.2.1.3 Anammox

Anammox is a new type of ammonia oxidation occurring under anoxic conditions. In the past, only the process of nitrification under aerobic conditions was known. Anammox is carried out by Plactomycetes bacteria which oxidize NH_4^+ using NO_2^- as an electron acceptor to produce nitrogen gas (Bernhard, 2010):

$$NH_4^+ + NO_2^- \rightarrow N_2 + 2H_2O$$

2.3.2.1.4 Denitrification

Denitrification is a process that removes bioavailable nitrogen and returns it to the atmosphere. It reduces NO_2^- and NO_3^- to gaseous components such as N_2, the final product of denitrification, or nitrous oxide (N_2O), an intermediate gaseous form of nitrogen. Nitrogen is considered a greenhouse gas and therefore contributes to air pollution. Denitrification is an anaerobic process carried out by a diverse group of prokaryotes (Bacillus, Paracoccus and Pseudomonas) which are chemo-organotrophic. They must therefore be supplied with organic carbon (Bernhard, 2010):

$$1)\ NO_3^- \rightarrow NO_2^- \rightarrow NO + N_2O \rightarrow N_2$$
$$2)\ 2NO_3^- + 10e^- + 12H^+ \rightarrow N_2 + 6H_2O$$

2.3.2.1.5 Ammonification

Ammonification is the mineralization or degradation of organic nitrogen into NH_3 (Delwiche, 1970). Plant and animal remains, in the form of organic nitrogen (amino acids, DNA, etc.), are broken down by various fungi and prokaryotes into NH_3. This process is called ammonification. The NH_3 produced by this process then becomes available to plants. The production of NH_4^+ or NH_3 depends on the ratio between ammonification and the immobilization of NH_4^+ in the form of microbial nitrogen (amino acids) (Van Bochove, 1993). The equilibrium equation between NH_4^+ and NH_3 is as follows:

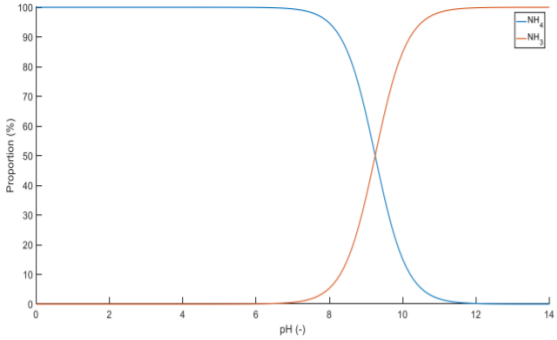


Figure 7: Percentage of NH_3 and NH_4^+ as a function of the pH of the solution (Stalport, 2017)

$$NH_3 + H_2O \overset{K_b}{\leftrightarrow} NH_4^+ + OH^-$$

Equilibrium is therefore influenced by pH, NH_3 and NH_4^+ concentrations, and temperature (Figure 7) (Van Bochove, 1993; Eck et al., 2019). The creation of NH_4^+ therefore leads to an increase in pH. During this stage, nitrogen losses are possible through volatilization of NH_3 and volatile organic nitrogen compounds (Van Bochove, 1993; Cáceres et al., 2015). Total ammoniacal nitrogen (TAN) is all nitrogen in the form of NH_3 and NH_4^+ (Eck et al., 2019).

2.3.2.2 The phosphorus cycle

Phosphorus is one of the most important elements for life since it plays a role in energy transfer and in the passage of genetic information via DNA. However, it is one of the rarest and therefore most limiting elements in an ecosystem. Indeed, the phosphorus cycle is an awfully slow process. Indeed, phosphorus is only present on earth, so the atmosphere plays no role in this cycle. Moreover, it is governed by various meteorological but also biological processes (Science Learning Hub, 2013; The Environmental Literacy Council, 2015; Biology dictonary, 2017; Britannica, 2020; Khan Academy, 2020). The different stages of the phosphorus cycle are (Figure 8):

- Weathering
- Absorption by plants and animals
- Return to the environment through decomposition

2.3.2.2.1 Weathering

Phosphorus is present in abundance in sedimentary rocks. As a result of weathering, leaching, or mining, phosphate salts slowly seep into surface water and soils. Volcanic ash, aerosols and mineral dust can also be important sources of phosphate. Phosphorus has no true gas phase, unlike other elements such as carbon, nitrogen and sulphur (Science Learning Hub, 2013; The Environmental Literacy Council, 2015; Biology dictonary, 2017; Britannica, 2020; Khan Academy, 2020).

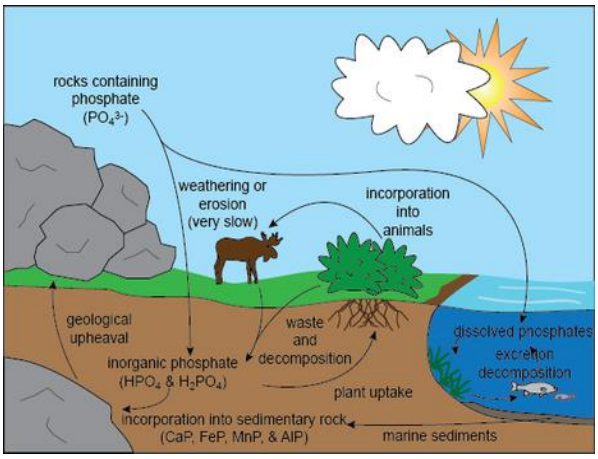


Figure 8: The phosphorus cycle (Raman et al., 2017)

2.3.2.2.2 Absorption by plants and animals

Due to their high reactivity, phosphorus is present in combined form with other elements. Soluble phosphates are produced from insoluble phosphorus by the acids of microorganisms. The phosphate salts dissolved in water are then absorbed by plants. Animals consume the plants or other plant-eating animals and thus absorb the phosphorus. The rate of phosphorus cycling is faster in plants and animals than in rocks (Science Learning Hub, 2013; The Environmental Literacy Council, 2015; Biology dictonary, 2017; Britannica, 2020; Khan Academy, 2020).

2.3.2.2.3 Return to the environment through decomposition

When plants and animals excrete waste or die, they are decomposed by detritus feeders, which convert organic phosphate into inorganic phosphate. In aquatic ecosystems, phosphorus from bodies or waste can also sink and form a new sedimentary layer. Over a long period of time, this layer can be moved towards the earth by a geological process called uplift. The phosphorus thus finds its way into sediments and rocks and can be released again through weathering. In this way, the phosphorus cycle starts all over again (Science Learning Hub, 2013; The Environmental Literacy Council, 2015; Biology dictonary, 2017; Britannica, 2020; Khan Academy, 2020).

2.3.2.3 The potassium cycle

The potassium cycle is divided into three parts (Figure 9):

- The soil components
- The contributions
- The losses

2.3.2.3.1 The soil components

Potassium, exclusively in mineral form, is found in the soil in 4 different components. Firstly, potassium enters the constitution of the mother rock, which can be mica or potassium feldspar. This first form represents 90 to 98% of the potassium present in the soil. The potassium is released into the soil solution through rock weathering and is then made available to plants. This process can take hundreds of years. Secondly, potassium can be included between the leaves of the clays. In this case, it becomes poorly exchangeable. Thirdly, it can be absorbed from the surface of the clay and humus particles. These exchange sites between the potassium ions and the clay and humus particles are called cation exchange complexes (CEC). The potassium is then in equilibrium with the soil solution and composes the samples taken by the roots from the soil solution. These movements with the soil solution are called absorption, the movement of potassium ions from the parent solution to the CEC, and desorption, the movement of the absorbed potassium out of the absorption sites. Ultimately, potassium is also present in the soil solution. Relatively immobile, it reaches plant roots mainly by diffusion into the liquid phase of the soil (wordpress, 2017; UNIFA, 2019; eKonomics, 2020).

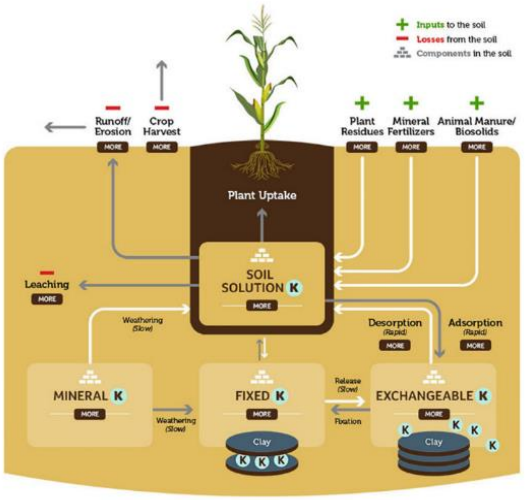


Figure 9: The potassium cycle (eKonomics, 2020)

2.3.2.3.2 The contributions

The potassium is returned to the soil through plant residues. Unlike nitrogen and phosphorus, potassium has no organic form in the tissues and does not need to be mineralized by microorganisms to be available to plants. A second potassium input is the application of fertilizers containing potash (KCl), potassium sulphate (K2SO4) or potassium nitrate (KNO_3). Potassium can also be supplied by animal manure (wordpress, 2017; UNIFA, 2019; eKonomics, 2020).

2.3.2.3.3 The losses

Potassium losses are mainly due to three different phenomena: agricultural exports, runoff/erosion, and leaching. When crops are harvested, the potassium contained in the grains or in the harvested part of the plant is removed from the potassium cycle. In addition, water runoff and erosion due to rain, irrigation, wind, or ice cause potassium losses. Ultimately, potassium can be moved from one area of soil to another by water leaching of nutrients (wordpress, 2017; UNIFA, 2019; eKonomics, 2020).

2.3.3 Sources of organic matter

Organic fertilizers can be made from different sources of organic matter, animal or vegetal. In general, the feedstock biodegradability is different for animal and plant waste (Shinohara et al., 2011). Table 6 gives an overview of the diversity of organic fertilizers and their NPK content.

Table 6: Organic fertilizers and NPK content (Rosen et al., 2005)

Table 7. *Approximate nutrient composition[1] of various organic fertilizers.*

Organic Materials	N	P_2O_5	K_2O
		% dry weight basis	
Manures			
Beef	1.2	2.0	2.1
Dairy	2.1	3.2	3.0
Bat guano	6.0	5.0	3.0
Horse	2.1	3.2	2.0
Poultry	3.0	5.0	2.0
Sheep	1.6	1.2	2.0
Swine	2.5	2.1	1.0
Alfalfa hay	2.5	0.5	2.5
Blood meal	13.0	2.0	1.0
Bone meal, raw	3.0	22.0	0
Bone meal, steamed	1.0	15.0	0
Castor bean meal	5.5	2.0	1.0
Cotton seed meal	6.0	3.0	1.5
Fish meal	10.0	6.0	4.9
Kelp/seaweed	1.5	1.0	0
Peanut meal	7.0	1.5	1.2
Soybean meal	7.0	1.2	1.5
Tankage[2]	7.0	10.2	1.5

[1] *These are total concentrations and only slowly available over weeks, months, or years. Many materials will vary in composition due to moisture content and methods of handling.*
[2] *May contain high levels of chromium.*

As far as animal raw material is concerned, it comes from the organic waste produced during the exploitation of livestock. The management of this waste is essential and its use as a fertilizer is one of the solutions studied (Cáceres et al., 2015). The elements present in the manure depend strongly on the type of animal (sheep, cattle, goats, pigs, ...). Indeed, while cows are mainly herbivores, pigs have a more varied diet. However, the diet also depends on the type of farming and its location in the world. The efficiency of animal manure is therefore difficult to predict due to the multitude of factors involved (size of the animal, feeding, management of the farm, ...) (Charoenpakdee, 2014). In general, chicken and bat droppings are rich in phosphorus and NO_3^-. On the other hand, cow and sheep droppings have lower nutrient concentrations than other manures. Pig manure has the lowest potassium concentration.

As with the use of animal excrement, the nutrient concentrations present in an organic fertiliser made from plant waste depend mainly on the type of plants used.

2.3.4 Techniques to produce organic fertilizers

In soil-based crops, organic fertilizers are directly incorporated into the soil and are degraded by soil micro-organisms that generate NO_3^- through ammonification and nitrification. If the microbial community in hydroponic systems were able to degrade organic fertilizers, it would be possible to add the organic fertilizers directly to the hydroponic solution, but this is not the case (Shinohara et al., 2011). Thus, organic fertilizers have a phytotoxic effect due to the concentration of organic matter and NH_3 present (Uchimura et al., 2014). It is therefore necessary to generate a culture of microorganisms capable of degrading organic matter to release the trapped nutrients in soluble form, potentially usable by plants (Shinohara et al., 2011). This phenomenon is called nutrient mineralization and can be carried out under anaerobic or aerobic conditions (Tomson, 2019).

2.3.4.1 Anaerobic digestion

Anaerobic digestion, also known as methanation, is the degradation of organic matter by micro-organisms in an oxygen-free environment. The main sources for anaerobic digestion in biogas plants are stable manure, crop residues, food industry waste, municipal waste, and dedicated energy crops (Möller et al., 2012). The products of this degradation are methane (CH4), carbon dioxide (CO2), NH_4^+, hydrogen sulphide (H2S) and water (H2O) (Tomson, 2019). Anaerobic digestion produces biogas but also a liquid effluent, called "digestate", which contains all the water, nutrients, and minerals of the organic matter (DeBruyn et al., 2015). The energy produced in this process can also be recovered. Anaerobic digestion is a complex process and can be summarised in 4 phases (Figure 10):

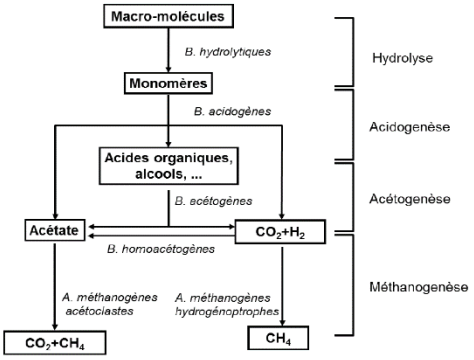


Figure 10: Anaerobic digestion (Bernet, 2015)

- Hydrolysis
- Acidogenesis
- Acetogenesis
- Methanogenesis

Hydrolysis breaks down organic macromolecules (polysaccharides, proteins, lipids) into monomers (simple sugars, amino acids, fatty acids, etc.). The monomers thus obtained are then fermented by acidogenic bacteria during the acidogenesis stage into organic acids, alcohols, hydrogen, and carbon dioxide. Acetogenesis is a key stage which converts the molecules produced previously into precursors of methanogenesis: acetate, hydrogen, and carbon dioxide. Finally, methanogenesis produces biogas and "digestate". Two groups of strict anaerobic micro-organisms belonging to the Archaea domain are involved: acetoclastic methanogenic bacteria and hydrogenotrophic methanogenic bacteria. The first group produces methane and CO2 from the acetate, while the second group uses hydrogen to reduce CO2 to methane. The "digestate" includes all the elements that have not been used to produce methane (Trably, 2002; Bernet, 2015; Orellana et al., 2019).

2.3.4.2 Aerobic digestion

Aerobic digestion, or liquid composting, is a fermentation process in which organic matter is in constant contact with oxygen used for the respiration of micro-organisms such as bacteria, moulds and yeasts. The latter consume the organic matter to extract the energy and elements necessary for their development (anabolism) but also to synthesize new living cells (catabolism). This degradation of organic matter releases the elements contained in it in solution. However, unlike anaerobic treatments, aerobic digestion does not recover energy. Like any biological process, aerobic digestion is influenced by the temperature and duration of digestion (Perez Fabiel, n.d.; Abdullahi et al., 2008; Bouaissa, 2015; Tomson, 2019).

2.3.5 Examples of known bioponics

2.3.5.1 Aquaponics

Aquaponics is one of the most widespread bioponic systems in the world. Aquaponics is the combination of aquaculture (fish farming) and hydroponics. It is based on the symbiosis of fish or, more rarely, other aquatic species, plants, and bacteria. Indeed, the waste generated by the fish is used as a source of nutrients for the plants after undergoing a nitrification process carried out by bacteria. By using the nutrient-rich waste, plants clean the water and thus make it viable for fish (Bodlovich et al., 2014; Shafahi et al., 2014; Kyaw et al., 2017).

The key parameter in this system is therefore the water. It contains the essential nutrients for the plants and oxygen for the fish. Its characteristics in terms of temperature, pH and dissolved oxygen must be a balance between the needs of plants, fish, and bacteria. Water quality monitoring is therefore essential for the proper growth of fish and plants (Shafeena, 2016).

Aquaponics therefore has two main advantages for nutrient cycles. Firstly, it reuses aquaculture effluents enriched with nitrogen and phosphorus, thus preventing their discharge into groundwater. Secondly, it avoids the use of chemical fertilizers by fertilizing above-ground crops with organic fertilizers (Eck et al., 2019; Tomson, 2019).

Table 7 gives an idea of the NPK content of fish droppings.

Table 7: Nutrient content of fish droppings (Peterhans, 2015)

Substance	Faeces content	Reference
N	28.30 g/kg DW	Naylor et al., 1999
K	1.00 g/kg DW	Naylor et al., 1999
Ca	6.53 g/kg DW	Köprücü and Özdemir, 2005
Mg	5.30 g/kg DW	Naylor et al., 1999
P	6.69 g/kg DW	Köprücü and Özdemir, 2005

2.3.5.2 Anthroponics

Anthroponics, also known as “urineponics” or “peeponics”, is the use of human bio-waste (urine and faeces) as a fertiliser. It is based on the concept of aquaponics but replaces the use of fish faeces as fertiliser with that of human urine. Moreover, the objective of anthroponics is different from that of aquaponics. Indeed, it aims to be a wastewater treatment and nutrient recovery system rather than a constructed ecosystem. It therefore combines aspects of aquaponics, organic hydroponics, and wastewater treatment (Sánchez, 2015; Adejumo et al., 2019).

Urine has a NPK fertilizer value of 18:2:5. Everyone produces 1 to 1.5l of urine per day. The composition of the urine depends on eating habits, physical activity, body size, the amount of water drunk and other environmental factors that may affect the individual (Pradhan et al., 2007). However, urine is in all cases an aqueous solution secreted by the kidneys and consists mainly of water. Other elements are also present such as urea and dissolved ions (chloride, sodium, potassium, and creatinine). It can be used as a source of nitrogen for plants through the volatilisation process of ammonia. In this process, the hydrolysis of urea is catalysed by urease into unstable carbamic acid. This acid then rapidly decomposes into ammonia and carbon dioxide. This process can be represented by the following equation:

$$(NH_2)_2CO + H_2O \xrightarrow{urease} NH_3 + H_2NCOOH \rightarrow 2NH_3(gas) + CO_2(gas)$$

The ammonia produced reacts with water to form ammonium. The nitrogen cycle can then take place (Sanchez, 2014).

The advantage of urine is its high nitrogen content. However, this rate is particularly important. It is therefore necessary to dilute the elements P, K and Ca, making them insufficient (Dumitrescu, 2013) . In addition, for health reasons, urine can only be used in hydroponic systems if it comes from a healthy individual, free of disease and infection and without any type of medication. Urine also needs to be sterilised before use because it contains micro-organisms such as faecal coliforms, clostridia, enterococci and coliphages that can cause health problems. The degradation of the urine by the volatilization process allows this fertilization to take place. In fact, the process causes the pH to rise to 9, which leads to bacterial reduction (Sánchez, 2015).

2.3.5.3 Digeponic

During the biogas production process, a by-product, in the form of slurry, is created. This by-product is called "digestate". The digestate produced is rich in NH_4^+. It can therefore be used as an organic fertiliser (Möller et al., 2012). In addition, it contains bioactive substances such as phytohormones, nucleic acids, monosaccharides, free amino acids, vitamins, fulvic acids, ... These phytohormones improve plant growth and increase the plant's tolerance to biotic and abiotic stress (Möller et al., 2012). However, the digestate can be phytotoxic due to the high amount of NH_4^+ and the high concentration of organic acid (Wang et al., 2018). In addition, it is important to control the generally high EC and to supplement the digestate with the missing nutrients (Dumitrescu, 2013; Stocknes et al., 2016; Wang et al., 2018). A second treatment is sometimes necessary to use the digestate. This second treatment has to be carried out aerobically. The water content, odour, carbon, and pathogens are then reduced (Abdullahi et al., 2008).

The digestate produced by biogas plants can replace up to 50% of the conventional mineral solutions found on the market. “Digeponic” is the integration of the products of anaerobic digestion, carbon dioxide and digestate, into greenhouse vegetable cultivation (Stocknes et al., 2016). The use of biogas slurry as a nutrient solution is therefore not only important in terms of recycling but also of reducing the use of mineral fertilisers. It therefore promotes the development of sustainable agriculture (Wang et al., 2018).

2.3.5.4 Use of Vermiwash

Vermiwash is a by-product obtained by leaching vermicompost during vermicomposting. Vermicomposting is the bioconversion of organic waste into biofertiliser due to the activity of earthworms. The vermicomposting process is optimal at pH 5.5-7.7, temperature 19°C-25°C and humidity 27.68-52.41%. Vermicomposting is therefore a mesophilic process (Manyuchi, 2013).

In addition to being an organic fertilizer, vermiwash has the property of protecting plants. In fact, the secretions from the earthworms improve the physico-chemical and biological properties of the plants. Moreover, vermiwash mainly increases pH, EC and potassium (Ansari et al., 2015).

2.3.5.5 Use of compost tea

Composting is defined as "a biological process that breaks down the organic constituents of by-products and waste into a stable organic product rich in humic compounds" (Van Bochove, 1993). One of the by-products of composting is compost tea. Compost tea consists of the liquid phase obtained during the composting process. The nutrient composition of compost tea depends on the type of waste being composted.

2.3.5.6 Use of ashes

The use of ashes in hydroponic systems helps to make up for deficiencies in potassium, magnesium, calcium and sulphur. Ash also allows the pH to increase, which stops the acidification of the water due to nitrification. It is important to note that the effect of ashes depends on the wood species used and the treatment that the ashes have undergone (Sanchez, 2015).

3 OBJECTIVES

As a reminder, the main objective of this work is the creation of a nutritive solution for hydroponics from goat and hen droppings under controlled conditions. Three sub-objectives were also presented:

- The creation of a state of the art on fertilization in hydroponics
- The creation and testing of an experimental system and a protocol for the mineralisation of organic matter (OM) in aerobic and anaerobic conditions.
- The implementation of exploratory tests to evaluate which parameters influence NPK ratios during mineralisation of goat and chicken droppings and the production of a nutrient solution that can be used in hydroponic systems.

The above literature review helps to meet the first objective. To help in the realisation of the following two objectives, this master thesis is based on the work of Thomas Tomson: "Innovative valorisation of fish farming effluents by the aquaponic method - final scientific report" (2019). In this work, the mineralisation experiment is carried out on fish sludge produced by aquaculture.

4 MATERIALS ET METHODS

Several steps were necessary to set up the experiments carried out within the framework of this master thesis. First, the mineralisation system was created and set up. Then, preliminary tests were carried out to test the system and analyse the mineralisation process. Finally, two experiments were carried out:

- Experiment 1: Aerobic and anaerobic mineralisation of goat and chicken droppings
- Experiment 2: Creation of a nutrient solution from mineralisation

These steps allowed the study of 3 different parameters. Firstly, two types of OM were studied: goat and chicken droppings. The impact of the percentage of OM, used when loading the system, on mineralisation was also evaluated. The choice of these organic materials was made according to the livestock farms in the Democratic Republic of Congo, the country in which Félicien Munungakatebe did his PhD thesis. Secondly, the impact of several pH values on the mineralisation of these OM was also studied. The reference pH is equal to 6. The choice of the studied pH values and the reference pH was based on the work of Thomas Tomson (2019). Two buffers are compared: phosphate buffer and citric acid buffer. The phosphate buffer is chosen for its frequent use in hydroponic systems. Thomas Tomson's experiment (2019) is notably carried out with this buffer. The citric acid buffer is chosen based on two criteria: its no-content of the NPK elements and its pKa (6.4) close to the desired pH (Ruzin, 1999). Finally, the two types of digestion, aerobic and anaerobic, allowing the degradation of OM into nutrients assimilable by plants were applied and their influence on mineralisation was quantified.

These experiments were carried out at a controlled temperature (35°C). This temperature is considered by Thomas Tomson (2019) as optimal for the release of ammonia nitrogen in solution.

During these experiments, pH, temperature, and EC are measured to control their evolution. The different NPK concentrations were also evaluated to estimate the impact of the three parameters studied (OM, pH, and digestion) on the mineralisation process.

The experiments were carried out in the Integrated and Urban Phytopathology Unit of the Faculty of Gembloux Agro-Bio Tech of the University of Liège between 5 May 2020 and 4 October 2020.

4.1 ASSESSMENT OF DRY MATTER CONTENT

The goat droppings are collected at the goat farm "La chèvre et le chou" in Eghezée (Belgium). The chicken droppings come from a breeding in barn in Corroy-le-chateau (Belgium). Once collected, the fresh droppings are put in the oven for 7 days at 40°C before being ground into a fine powder.

Three samples of each OM are taken and weighed with a METTLER PM6400 scale. They are then dried in the oven at 105°C for 24 hours and weighed again to determine their dry weight. The average dry matter percentage of the three samples is then calculated (Appendix 1) and is used to load the reactors.

4.2 REACTOR SET-UP

For the preliminary tests and experiment 1, the experimental structure consists of a shelf containing two watertight tanks that serve as a water bath (Figure 11). These two tanks are heated to 35°C by two 200W resistors. Each tank can contain up to 15 reactors. These reactors consist of a bucket (5L) filled up to 3L because the digestion of the excrements produces a foam which could cause the bucket to overflow (Figure 12a). For aerobic modalities, an air flow from an air pump (AquaForte AP-45) is injected through an air

distributor and distributed between the different buckets (1 bubbler per bucket) (Figure 12b). For anaerobic modalities, no air flow is injected into the buckets and the buckets are closed with a lid. Following the results obtained in preliminary test 1, improvements are made to the system implemented. In preliminary test 2 and experiment 1, tarpaulins were added to the water baths. In addition, 40 biofilters are added to each bucket to accelerate mineralisation.

Figure 11: Overview of the digestion system for preliminary tests and experiment 1

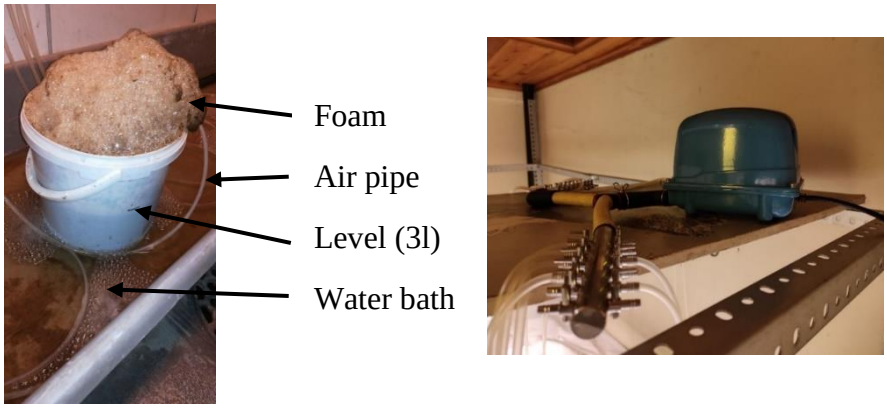


Figure 12: a) Reactor preliminary tests and experience 1; b) Air pump and air distributor

For the second experiment, four 200-litre buckets are used as reactors. Each bucket is only filled up to 100L (Figure 13). The temperature is no longer controlled, but the system of air flow injected into the buckets remains the same as in experiment 1. As all the reactors are in aerobic conditions, five bubblers are placed in each bucket. For modalities where the pH is controlled, an IKS keeps the pH constant (Figure 14). In addition, in each bucket, 150 g of biofilters are added to accelerate mineralisation.

Figure 13: Overview of the digestion system for experiment 2

Figure 14: IKS pH control system

4.3 PRELIMINARY TEST

4.3.1 Preliminary test 1

4.3.1.1 Objectives

The objectives of the first preliminary test were:

- the improvement of the digestion system in place by testing the proper functioning of the water bath and its ability to maintain the bucket at temperature
- the creation of a sampling protocol.
- the study of the buffering capacity of citric acid compared to a phosphate buffer
- Identify the relationship between the amount of TAN and the percentage of dry matter
- Follow the evolution of the elements (NPK) at pH 6 (reference pH in the experiment of Thomas Tomson (2019)).

4.3.1.2 Modalities

For this preliminary test, 24 modalities (numbered from 1 to 24) were tested without repetition. The parameters studied were:

- The OM: goat and chicken
- The percentage of dry matter: 0.2%, 0.4%, 0.6% and 0.8%.
- The pH: 5.5, 6, 6.5, 7 and free pH (control pH)
- The buffer: citric acid and phosphate

For each OM, pH 5.5, 6, 6.5 and 7 were studied for the two buffers with a percentage of dry matter of 0.2%. The free pH was also studied with 0.2% dry matter. The different percentages of dry matter were analysed only at pH 6 with the phosphate buffer.

4.3.1.3 pH control

4.3.1.3.1 Phosphate Buffer Protocol

To create the phosphate buffer, two solutions are used:

- A: Sodium phosphate dibasic dihytrate (Na2HPO4.2H2O), mw=178.05; 0.2M solution contains 35.61 g/l
- B: Sodium phosphate monobasic monahydrate (NaH2PO4.H2O); mw=138.01; 0.2M solution contains 27.6 g/l

Depending on the desired pH value, solutions A and B were mixed according to the proportions in Table 8 and diluted with water to reach 3000 ml of buffer solution (Promega Corporation, 2012; Merck, 2020).

Table 8: Quantity of solution A and solution B for pH 5.5, 6, 6.5 and 7 (phosphate buffer)

pH, 25°C	x ml de 0,2M-Na2HPO4.2H2O	y ml de 0,2M-NaH2PO4.H2O
5.5	90	1410
6	184.5	1315.5
6.5	480	1020
7	915	585

4.3.1.3.2 Citric acid buffer protocol

To create the citric acid buffer, two solutions are used:

- A: Citric acid monohydrate (C6H8O7), mw=210.14; 0.1M solution contains 21.01 g/l
- B: Trisodium citrate dihydrate (C6H5O7Na3.2H2O); mw=294.12; 0.1M solution contains 29.4 g/l

Depending on the desired pH value, solutions A and B were mixed according to the proportions in Table 9 and diluted with water to reach 3000 ml of buffer solution (Promega Corporation, 2012; Merck, 2020).

Table 9: Quantity of solution A and solution B for pH 5.5, 6, 6.5 and 7 (citric buffer)

pH, 25°C	x ml de 0,1M-C6H8O7	y ml de 0,1M-$C_6H_5O_7Na_3.2H_2O$
5.5	695.5	2302.5
6	345	2655
6.5	102	2898
7	30	2970

4.3.1.3.3 Free pH

The free pH reactors were loaded with demineralised water instead of buffer solutions.

4.3.1.4 Experimental follow-up

Three different experimental follow-ups were carried out on the different modalities. Each modality did not have the same follow-up. In fact, the buckets with pH 6 (reference pH) underwent more extensive experimental monitoring. To identify the monitoring to be carried out, a letter was written on the buckets:

- Letter A: every two days the temperature, the pH and EC of the bucket is checked.
- Letter B: after 15 days, the TIN, P (for no buffer) and K content is evaluated using a spectrophotometer.
- Letter C: the first two days and then every two days, the TIN, P (for no buffer) and K content is evaluated using the spectrophotometer for the modalities at pH 6 and free pH (reference pH and control pH in the experiment of Thomas Tomson (2019)).

Table 10 shows the experimental monitoring carried out for each modality.

Table 10: Table of modalities of preliminary test 1 and follow-ups carried out

Samples	Organic matter	pH	Buffer	% dry matter	Analysis		
1	goat	5.5	citric acid	0.2	A	B	
2	goat	5.5	phosphate	0.2	A	B	
3	goat	6	citric acid	0.2	A	B	C
4	goat	6	phosphate	0.2	A	B	C
5	goat	6	phosphate	0.4	A	B	
6	goat	6	phosphate	0.6	A	B	
7	goat	6	phosphate	0.8	A	B	
8	goat	6.5	citric acid	0.2	A	B	
9	goat	6.5	phosphate	0.2	A	B	
10	goat	7	citric acid	0.2	A	B	
11	goat	7	phosphate	0.2	A	B	
12	goat	No buffer	No buffer	0.2	A	B	C
13	chicken	5.5	citric acid	0.2	A	B	
14	chicken	5.5	phosphate	0.2	A	B	
15	chicken	6	citric acid	0.2	A	B	C
16	chicken	6	phosphate	0.2	A	B	C
17	chicken	6	phosphate	0.4	A	B	
18	chicken	6	phosphate	0.6	A	B	
19	chicken	6	phosphate	0.8	A	B	
20	chicken	6.5	citric acid	0.2	A	B	
21	chicken	6.5	phosphate	0.2	A	B	
22	chicken	7	citric acid	0.2	A	B	
23	chicken	7	phosphate	0.2	A	B	
24	chicken	No buffer	No buffer	0.2	A	B	C

Experimental monitoring is carried out in 3 stages:

- Re-levelling the buckets with water. Due to evaporation, the quantity of water decreases each day. It is essential to fill the buckets before sampling.
- Sampling the solution in the buckets. The samples taken for the first crash test were 50 ml Falcon™.
- Temperature, pH and EC measurement of buckets with a multimeter (Hach HQ40d)

It is important to carry out the sampling before the temperature is taken so that the water bath has had time to warm up the water added during the re-levelling.

After sampling, the samples were centrifuged (SIGMA 4-16KS) at maximum speed (9000 min^{-1} or 13131*g) for 10 min at room temperature (27°C) in order to remove the particles present in the samples. They were then stored at -20°C for analysis.

The analyses were carried out with spectrophotometers (Hanna HI 83200) and were:

- Ammonia:
  - Nessler method, yellow coloration
  - Range: 0.00 > 10 mg/l
  - Resolution: 0.01 mg/l
  - Accuracy: ± 0.05 mg/l

- Nitrate:
    - Cadmium reduction method, amber coloration
    - Range: 0.00 > 30 mg/l
    - Resolution: 0.1 mg/l
    - Accuracy: ± 0.5 mg/l
- Nitrite:
    - EPA diazotization method, purple coloration
    - Range: 0.00 > 0.35 mg/l
    - Resolution: 0.01 mg/l
    - Accuracy: ± 0.02 mg/l
- Phosphate:
    - "Standard methods for the examination of water and wastewater 18th edition" by amino acid, blue coloration
    - Range: 0 > 30 mg/l
    - Resolution: 0.1 mg/l
    - Accuracy: ± 1 mg/l
- Potassium:
    - Turbidity or tetraphenylborate method
    - Range: 10 > 100mg/l
    - Resolution: 2.5 mg/l
    - Accuracy: ± 15 mg/l

4.3.2 Preliminary test 2

4.3.2.1 Objectives

The objectives of pre-test 2 were almost identical to pre-test 1. This test was mainly used to verify that the improvements made to the digestion system as a result of the first pre-test work. The objectives of the second test were therefore:

- Test the proper functioning of the water bath and its ability to maintain the bucket at temperature
- Test the effect of aerobic and anaerobic digestion
- Identify the relationship between the amount of TAN and the percentage of dry matter
- Follow the evolution of the elements (NPK) at pH 6 (reference pH in the experiment of Thomas Tomson (2019)).

4.3.2.2 Modalities

For this preliminary test, 26 modalities (numbered from 1 to 26) were tested without repetition. The parameters studied were:

- The OM: goat and chicken
- The percentage of dry matter: 2.5%, 5%, 7.5% and 10%.
- The pH: 5.5, 6, 6.5, 7 (buffer phosphate) and free pH
- The digestion type: aerobic and anaerobic

For each OM, pH 5.5, 6, 6.5, 7 and free pH were studied with a percentage of dry matter of 2.5% in aerobic conditions. The different percentages of dry matter were also studied under aerobic conditions only for pH

6 and free pH. Anaerobic digestion was carried out on a bucket at pH 6 and a bucket at free pH with a dry matter percentage of 2.5%.

4.3.2.3 pH control

The protocols for the manufacture of the phosphate buffer have been validated during preliminary test 1 and therefore they have been applied during preliminary test 2. The citric acid buffer is no longer used due to its reduced efficiency in keeping the pH of the solutions stable during the first preliminary test.

4.3.2.4 Experimental follow-up

The three experimental follow-ups carried out for preliminary test 1 remain the same for preliminary test 2. The letters with the different follow-ups are written again on the buckets. Further follow-up is carried out on the modalities with a percentage of dry matter of 2.5% for pH6 and free pH in aerobic and anaerobic conditions. Table 11 shows the monitoring carried out for each modality.

Table 11: Table of modalities of preliminary test 2 and follow-ups carried out

Samples	Organic matter	Digestion	pH	% dry matter	Analysis		
1	goat	aerobic	5.5	2.5	A	B	
2	goat	aerobic	6	2.5	A	B	C
3	goat	aerobic	6	5	A	B	
4	goat	aerobic	6	7.5	A	B	
5	goat	aerobic	6	10	A	B	
6	goat	aerobic	6.5	2.5	A	B	
7	goat	aerobic	7	2.5	A	B	
8	goat	aerobic	No buffer	2.5	A	B	C
9	goat	aerobic	No buffer	5	A	B	
10	goat	aerobic	No buffer	7.5	A	B	
11	goat	aerobic	No buffer	10	A	B	
12	goat	anaerobic	6	2.5	A	B	C
13	goat	anaerobic	No buffer	2.5	A	B	C
14	chicken	aerobic	5.5	2.5	A	B	
15	chicken	aerobic	6	2.5	A	B	C
16	chicken	aerobic	6	5	A	B	
17	chicken	aerobic	6	7.5	A	B	
18	chicken	aerobic	6	10	A	B	
19	chicken	aerobic	6.5	2.5	A	B	
20	chicken	aerobic	7	2.5	A	B	
21	chicken	aerobic	No buffer	2.5	A	B	C
22	chicken	aerobic	No buffer	5	A	B	
23	chicken	aerobic	No buffer	7.5	A	B	
24	chicken	aerobic	No buffer	10	A	B	
25	chicken	anaerobic	6	2.5	A	B	C
26	chicken	anaerobic	No buffer	2.5	A	B	C

Experimental monitoring is carried out in the same three stages as for preliminary test 1, however, sampling is carried out with 15 ml Falcon instead of 50 ml.

Sample processing and analysis remain the same.

4.4 EXPERIMENT 1: AEROBIC AND ANAEROBIC MINERALIZATION OF GOAT AND CHICKEN DROPPINGS

4.4.1 Objectives

The objective of experiment 1 is to continue the exploratory tests carried out during the preliminary tests. The objective of this experiment is therefore:

- Test the effect of:
  - Aerobic and anaerobic digestion
  - Goat and chicken droppings
  - pH control

  on the release of N-P-K in the nutrient solution.

4.4.2 Modalities

The first experiment lasts 30 days. Eight different modalities are tested (Table 12):

- OM (2): goat and chicken
- Buffer (2): No buffer and Phosphate buffer
- Digestion (2): Aerobic and Anaerobic

Due to lack of time, only modalities with 2.5% OM at pH 6.5 are studied. PH 6.5 is chosen because it optimises the concentrations of TAN present in solution and has a better buffering capacity than pH 6 because it is closer to the pKa of the buffer.

Table 12: Modalities of experiment 1

Samples	Organic matter	pH	Digestion
1	goat	6.5	Aerobic
2	goat	No buffer	Aerobic
3	goat	6.5	Anaerobic
4	goat	No buffer	Anaerobic
5	chicken	6.5	Aerobic
6	chicken	No buffer	Aerobic
7	chicken	6.5	Anaerobic
8	chicken	No buffer	Anaerobic

To be able to produce statistics, each modality is repeated 3 times (repetition A, B and C). A total of 24 buckets/reactors are set up. As the conditions in the water baths are not perfectly homogeneous, the buckets are divided into 3 blocks each corresponding to one repetition. Within the blocks, the modalities are positioned randomly (Figure 15).

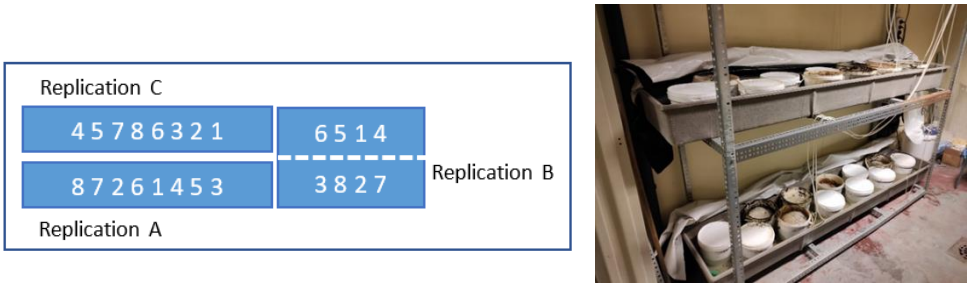


Figure 15: a) diagram of the positioning of the buckets, b) photo of the setting up of experiment 1

4.4.3 pH control

The protocols for the manufacture of the phosphate buffer have been validated during preliminary test 1 and therefore they have been applied during experiment 1.

4.4.4 Experimental follow-up

Every three days, the temperature, pH, and EC of each bucket are checked (Hach HQ40d). The TIN, P (for no buffer) and K content are also evaluated with the spectrophotometer. To perform this analysis, 5 ml are sampled in each bucket. Samples of the same modality (3 replicates) are mixed to have 8 samples of 15 ml each. This type of sampling is carried out due to the lack of time to study the contents of the three repetitions separately. These 15 ml samples are considered to represent the average of the three replicates.

After 15 days and at the end of the experiment (day 30), the TIN, P (for no buffer) and K content is evaluated using a spectrophotometer for each bucket. These data are used to study the variance between replicates.

The analyses carried out on the samples remain the same as for the preliminary test 1.

4.4.5 Statistics

The statistics for this study are produced using Rstudio software (R Core Team, 2018). Several tests were carried out according to the parameter studied.

The ANOVA is performed on the following parameters:

- OM: goat and chicken
- pH: pH 6.5 and free pH
- Digestion: aerobic and anaerobic
- Days: for EC, temperature and pH, 10 days are studied (days 0, 3, 6, 9, 12, 15, 18, 21, 24, 27 and 30), for NPK concentrations only days 15 and 30 are taken into account

The ANOVAs carried out therefore include the study of each of the parameters and their interactions.

The conditions for carrying out an ANOVA are normality and equality of variances. These conditions have been verified posteriori on the ANOVA residues using the Shapiro-Wilk test for normality (H0= ANOVA residues follow a normal population) and the Bartlett test for equality of variances (H0= the variances of the ANOVA residues are identical). If these two conditions are not met, the results of the ANOVA should be taken with caution. However, if the p-values of the parameters (OM, pH, digestion and days) or interactions are very low (p-value<0.001), it is highly probable that they have an influence on the parameter studied (pH, temperature, EC or concentration of N, P or K).

Although there are interactions between the parameters, the ANOVAs carried out have not been subdivided to study the parameters separately. Indeed, the objective of this study is to know the differences present between the different modalities and not the influence of each parameter on the other ones. To achieve this objective, a Tukey's HSD post hoc test was carried out to create groups according to the OM-pH-Digestion interaction from the ANOVAs carried out on the fourth parameters. This interaction corresponds to the eight modalities studied. The Tukey's HSD post hoc test therefore makes it possible to group the different modalities together if their averages are equal.

Concerning the statistical study of ammoniacal nitrogen, only the TAN concentrations were analysed. Indeed, the daily evolution of NH_4^+, NH_3 and TAN concentrations are similar. In addition, the concentrations of NH_3 and NH_4^+ are close. Therefore, only TAN is presented in the results. The results for NH_4^+ and NH_3 separately are presented in Appendix 2 and Appendix 3.

4.4.6 Outside temperature

The outside temperature during the month of September was studied posteriori with data from the observatory in Uccle (Figure 16). These data were used to analyse the temperature data from the different modalities for Experiment 1.

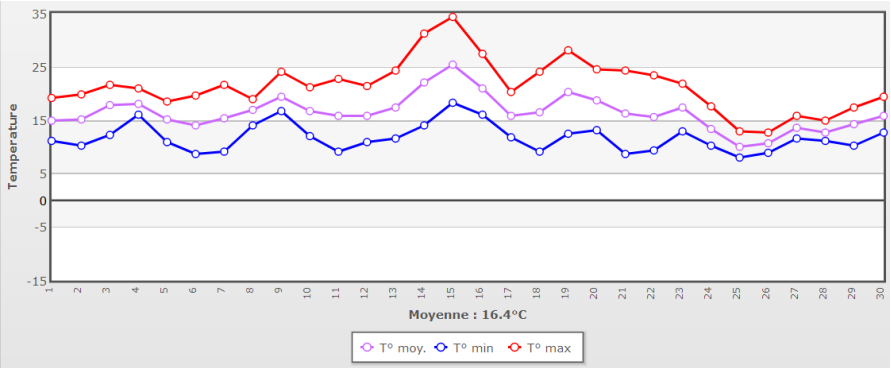


Figure 16: Value of monthly temperatures in Uccle for September 2020 (Mievis, 2020)

4.5 EXPERIMENT 2: CREATION OF A NUTRITIVE SOLUTION FROM MINERALIZATION

4.5.1 Objectives

The objective of the second experiment is to create a nutrient solution in large quantities in order to be able to test it later on the growth of lettuce in hydroponics and to estimate, from its NPK ratio, its efficiency.

4.5.2 Modalities

The modalities are:

- Solution A: goat and water
- Solution B: goat and buffer
- Solution C: chicken and buffer
- Solution D: chicken and water

The experiment set up consists of the creation of 4 different nutrient solutions of 100 litres. The solutions are created in 200 l tanks without temperature control. The percentage of OM is 2.5%. Each tank is placed in an aerobic condition thanks to an air flow from an air pump.

4.5.3 pH control

The buffer used in tanks B and C is phosphate buffer at pH 6.5. The manufacturing protocol is the same as in preliminary test 1 (4.3.1.3.1 Phosphate Buffer Protocol).

In addition to the phosphate buffer at pH 6.5, the pH is controlled by the IKS. Every 5 minutes, the IKS measures the pH. If it is higher than 6.5, the machine adds acid (5% H2SO4) for 30 seconds.

4.5.4 Experimental follow-up

As Experiment 2 is set up in parallel with Experiment 1, the experimental monitoring is carried out in the same way. Fifteen millilitre samples are taken every 3 days, as well as measuring temperature, pH and EC. These are then centrifuged and frozen at -20°C before analysis with a spectrophotometer. The analyses carried out are identical to the preliminary test 1 (4.3.1.3.3 Free pH

The free pH reactors were loaded with demineralised water instead of buffer solutions.

Experimental follow-up).

4.6 LEGEND OF THE GRAPHS

To facilitate the reading of the graphs, a common legend has been applied to all results (Table 13).

In this legend, the different parameters studied are represented by different symbols:

- OM: the modalities containing goat droppings are shown in blue, while those containing chicken droppings are shown in orange.
- Digestion: the aerobic modalities are represented with square bullets while the anaerobic modalities are represented with triangular bullets.
- pH: the modalities at pH 6.5 (noted "buffer") have hollow bullets while the modalities at free pH (noted "water") have full bullets.

The letters symbolizing the groups of the HSD test are noted in the legend of the graphs.

Table 13: Common legend of the graphs

Modalites		Legend
Organic matter	Goat	
	Chicken	
Digestion	Aerobic	
	Anaerobic	
pH	Water	
	Buffer	

Samples	Modalities	Légend
1	Goat/buffer/aerobic	
2	Goat/water/aerobic	
3	Goat/buffer/anaerobic	
4	Goat/water/anaerobic	
5	Chicken/buffer/aerobic	
6	Chicken/water/aerobic	
7	Chicken/buffer/anaerobic	
8	Chicken/water/anaerobic	

5 RESULTS

5.1 PRELIMINARY TEST

5.1.1 Preliminary test 1

Preliminary test 1 led to several conclusions. Firstly, the digestion system in place can be improved as it has significant heat and water losses. In addition, the system did not release N-P-K in solution. The percentage of dry matter used is therefore too low. Afterwards, the sampling protocol was approved. It allows an efficient and as homogeneous sampling as possible. Finally, citric acid does not allow the pH of the buckets to be buffered properly. Results are better with a phosphate buffer.

As far as the improvement of the system is concerned, several points of attention are noted. First, the evaporation of water in the buckets and the water bath is important. To reduce this water loss, a tarpaulin is installed above the water bath during the second preliminary test. This tarp allows for greater water saturation of the surrounding environment. In addition, it allows better control of the temperature, which varies greatly between buckets but also during the test. This second parameter is also improved by the installation of a pipe allowing the creation of a current in the water bath. Secondly, the concentrations of OM used are too low. The mineralisation could not be carried out correctly and the NPK concentrations after 15 days of experiments are insufficient. To increase the mineralisation in the second preliminary test, the concentration of OM is multiplied by ten. In addition, biofilter beads are added to the buckets. Finally, the bubblers used in the first preliminary test do not allow a homogeneous bubbling. The use of these bubblers therefore had an impact on the mineralisation rate of the buckets. To solve this problem, new bubblers were used in the second preliminary test, allowing the oxygenation of the different buckets to be standardised.

The sampling protocol used during the first test allowed for homogeneous sampling. However, several points of attention are highlighted. First, when taking the samples, the water level must be brought back to 3L before sampling. Indeed, the measurements taken on the samples are concentration measurements. The volume must therefore be similar in each bucket to be able to compare these concentrations with each other.

Finally, the study of the efficiency of the citric acid and phosphate buffer has shown that the citric acid buffer is not ideal for this experiment. Indeed, the pH of the samples made with this buffer increases sharply to reach values close to 9.5. It is therefore no longer used in the other experiments carried out.

5.1.2 Preliminary test 2

The second preliminary test confirmed the protocols and system in place. All the improvements proposed at the end of the first test were approved and retained. Regarding NPK values, these are higher and allowed a more in-depth study of the differences between modalities. However, the conclusions drawn from this second preliminary test are based on the results obtained after analysis of a single sample. No statistics are therefore produced to confirm these conclusions. The results of preliminary test 2 are presented in Appendix 4 and Appendix 5.

On the one hand, TAN and K concentrations seem to be proportional to the percentage of dry matter present in the different modalities. On the other hand, the concentration of P does not seem to be related to the concentration of OM. Moreover, nitrogen is only present in ammoniacal form. Indeed, NO_2^- and NO_3^- are not found in solution. Important differences exist between treatments under aerobic and anaerobic

conditions and are also present between types of OM. Indeed, chicken droppings seem to provide more elements in solution.

Following these encouraging conclusions, the objective of experiment 1 is to study the modalities of preliminary test 2 and to carry out repetitions to be able to quantify the different effects highlighted. However, the different concentrations of OM were not retained in experiment 1 due to lack of time. Experiment 1 was carried out with a percentage of OM of 2.5%.

5.2 EXPERIMENT 1: AEROBIC AND ANAEROBIC MINERALIZATION OF GOAT AND CHICKEN DROPPINGS

5.2.1 pH

In general, for all modalities, the pH increases over time. However, the results of the pH analysis clearly show a difference between modalities in aerobic and anaerobic conditions. In aerobic conditions, the modalities have a higher average pH than in anaerobic conditions. Different groups can therefore be distinguished according to digestion, but also according to OM and pH management.

For the pH, the conditions for carrying out the ANOVA are not respected since the residues do not follow a normal population and the variances of the residues are not equal. However, the p-values of the four parameters (OM, pH, digestion, days) are extremely low (p-value<0.001). An impact of these parameters on pH is therefore expected.

The HSD test divides the modalities into 6 groups (Figure 17):

- A: Chicken/water/aerobic (mean=8.05) and Goat/water/aerobic (mean=7.87)
- B: Chicken/buffer/aerobic (mean=7.17) and Goat/buffer/aerobic (mean=6.93)
- C: Chicken/water/anaerobic (mean=6.58)
- CD: Chicken/buffer/anaerobic (mean=6.38)
- DE: Goat/buffer/anaerobic (mean=6.10)
- E: Goat/water/anaerobic (mean=5.90)

As a reminder, the letters in the graph legend correspond to the groups created during the Tukey's HSD post hoc test.

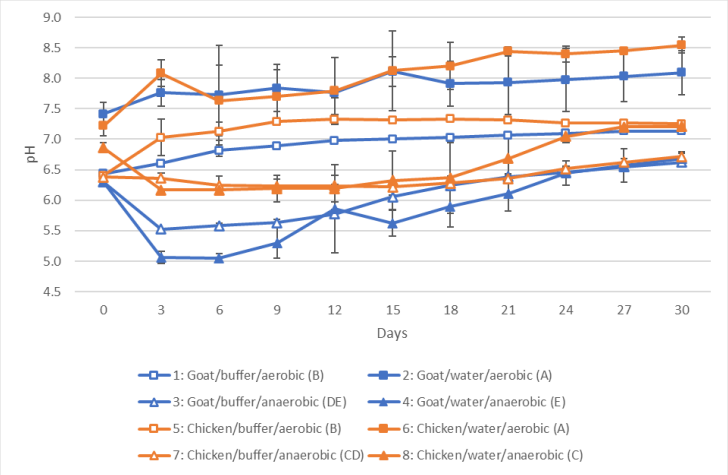


Figure 17: Daily evolution of the pH

The HSD test shows the effect of digestion on pH. In fact, the two groups with the highest averages (A and B) are in aerobic conditions, while the other four groups (C, CD, DE and E) are in anaerobic conditions. On the one hand, for aerobic digestion, pH management also influences the results. In fact, group A includes the free pH modalities while group B includes the controlled pH modalities. Within these groups, even if the averages are not considered significantly different, the chicken has a higher average than the goat. On the other hand, for anaerobic digestion, it is more difficult to draw conclusions. Indeed, OM and pH management seem to have a combined influence on the pH results.

5.2.2 Temperature

The temperature of the buckets generally decreases during the experiment. The calendar day has an influence on the temperature, which means that temperature control was not optimal. The temperature between the different buckets was therefore not homogenous and the bucket with the highest temperature is not always identical. In addition, the type of digestion, both aerobic and anaerobic, also seems to be influenced (Figure 18).

The equality of ANOVA variances is respected but the residuals do not follow a normal population. The ANOVA results show that "digestion" as well as "days" has a significant effect on temperature (p-values<0.001). On the other hand, "OM" does not seem to influence temperature (p-value>0.5). Concerning pH management, no conclusions can be drawn because the p-value is too close to 0.05 (p-value=0.0165).

Tukey's HSD test divides the modalities into 4 groups:

- A: Goat/buffer/anaerobic (mean=33.54°C) and Goat/water/anaerobic (mean=33.46°C)
- AB: Chicken/buffer/anaerobic (mean=33.42°C), Goat/water/aerobic (mean=33.37°C), Chicken/water/anaerobic (mean=33.29°C) and Chicken/water/aerobic (mean=33.25°C)
- BC: Chicken/buffer/aerobic (mean=33.02°C)
- C: Goat/buffer/aerobic (mean=32.73°C)

Tukey'HSD test does not, in this case, give more information than the ANOVA. On the one hand, digestion seems to be influenced by temperature. Indeed, within the same OM, the temperature is on average higher under anaerobic conditions. However, the averages are not, in all cases, significantly different. On the other hand, no conclusions can be drawn for OM and pH management from the results of the HSD test.

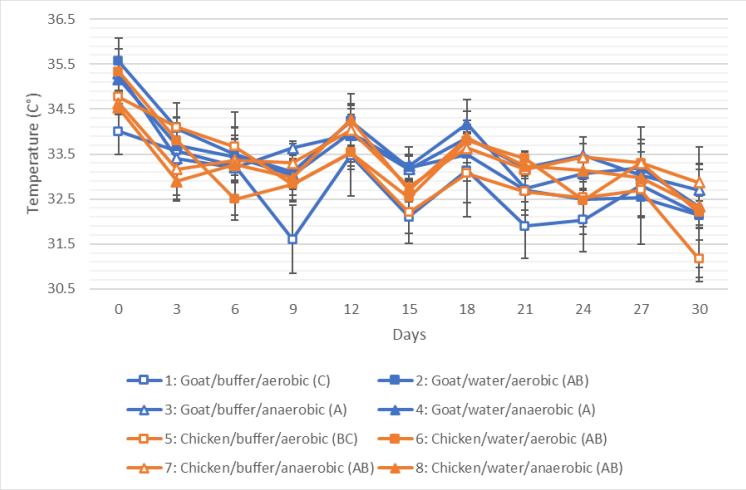


Figure 18: Daily evolution of the temperature

5.2.3 EC

The EC seems to be influenced mainly by pH management but also by the type of OM and digestion. Indeed, pH-controlled modalities have a higher EC than those with free pH. In addition to pH management, chicken droppings seem to be able to release more salts in solution since their EC is higher than that of goat droppings. Finally, within the same pH management and droppings type, anaerobic digestion seems to have a higher EC (Figure 19).

The conditions of normality and equality of variance for ANVOVA residues are not met. However, the p-values for OM, digestion and pH management are low (p-value<0.001). These parameters therefore influence the EC value. No conclusions can be drawn for the day because its p-value is too close to 0.05 (p-value=0.0039).

Tukey's HSD test creates 7 different groups:

- A: Chicken/buffer/anaerobic (mean=12.71 mS/cm)
- B: Chicken/buffer/aerobic (mean=11.88 mS/cm)
- C: Goat/buffer/anaerobic (mean=9.66 mS/cm)
- D: Goat/buffer/aerobic (mean=8.98 mS/cm)
- E: Chicken/water/anaerobic (mean=6.68 mS/cm)
- F: Goat/water/anaerobic (mean=2.96 mS/cm) and Chicken/water/aerobic (mean=2.74 mS/cm)
- G: Goat/water/aerobic (mean=1.43 mS/cm)

Tukey's HSD test has shown that pH management has a major influence on EC. Indeed, buffer-containing modalities have a higher EC than free pH modalities. On the one hand, for pH-controlled modalities, OM appears to have a greater influence than digestion. Anaerobic and aerobic chicken droppings modalities have a higher EC than goat droppings. On the other hand, for free pH modalities, the influence of digestion seems to be stronger than OM. In fact, anaerobic modalities containing chicken and goat droppings have a higher EC than those in aerobic conditions. However, for free pH modalities, the influence of OM or digestion seems difficult to prioritize since the Goat/water/anaerobic and Chicken/water/aerobic modalities do not have significantly different averages.

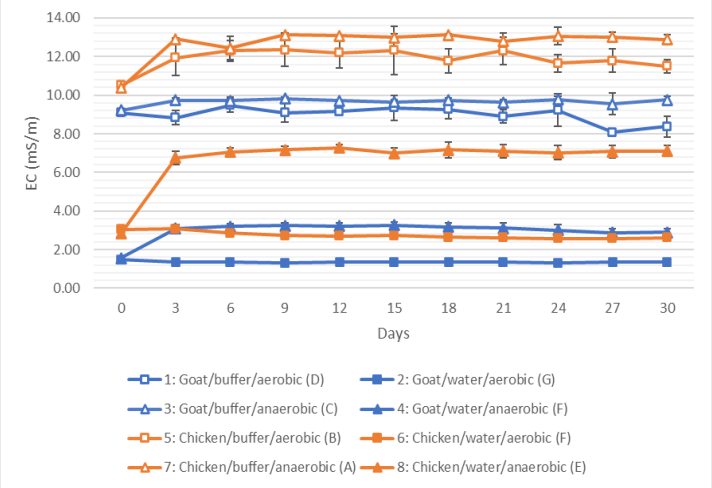


Figure 19: Daily evolution of the electroconductivity

5.2.4 NPK concentrations

5.2.4.1 Correlation between NPK concentration results for pool samples and average replications for days 15 and 30

The pool sampling method can be evaluated by studying the correlation between the pool samples and the averages obtained during the analyses of the replicates for days 15 and 30 of the experiment. If these two values are exactly equivalent, the trend curve is a straight line with a slope equal to 1 and passing through the origin of the axes. In addition, the coefficient of determination is equal to 1, which means that all the points on the correlation graph lie exactly on the trend line.

In general, the correlation between the two types of sampling is good. The pool sampling method can therefore be considered as representative of the average of replications. However, a few points of attention are highlighted.

Concerning the correlation between pool samples and the average of the replicates, the coefficient of determination is close to 1 for all the analyses carried out on days 15 and 30. However, as the NO_2^- analyses give very low results for pool samples but also for the averages, the study of the correlation is not relevant (Figure 22 and Figure 23). The following conclusions are therefore not applicable for this element. In addition, the coefficient of determination for NO_3^- on day 15 is also lower (R^2=0.5909). The trend line is therefore not representative of points on the graph (Figure 24 and Figure 25). Its characteristics are therefore not investigated.

On the one hand, the average of the repetitions for day 15 is higher than the value of the pool sample. Indeed, the slopes of the correlation graphs between these two values are less than 1 for all the nutritional elements studied. This systematic error of unknown origin is between 10 and 17%, depending on the test carried out. On the other hand, for day 30, the slope of the trend lines is very close to 1 for all the analyses (between 1.0041 and 1.0242), except for the correlation for phosphorus, whose slope of the trend line is equal to 0.6721 (Figure 27).

The pool sample is therefore a reliable representation of the replicate averages. However, special attention must be paid to the test results for NO_3^-. Indeed, for this test, the correlation is the pool samples, and the averages of the replicates are less accurate.

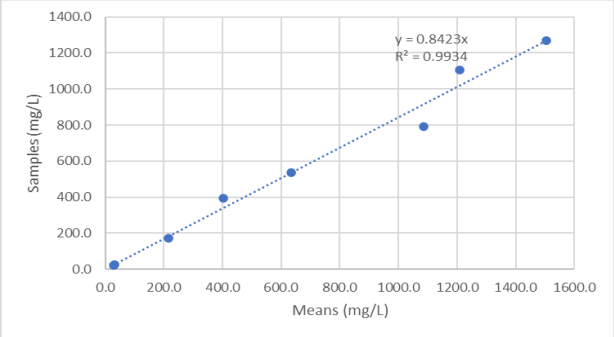


Figure 20: Correlation between the total ammonia nitrogen values of the pool samples and the average for day 15

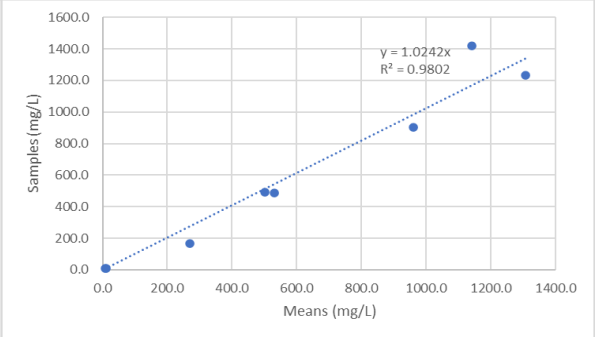


Figure 21: Correlation between the total ammonia nitrogen values of the pool samples and the average for day 30

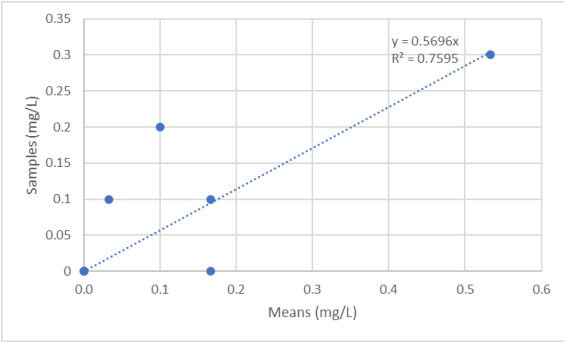


Figure 22: Correlation between the nitrite values of the pool samples and the average for day 15

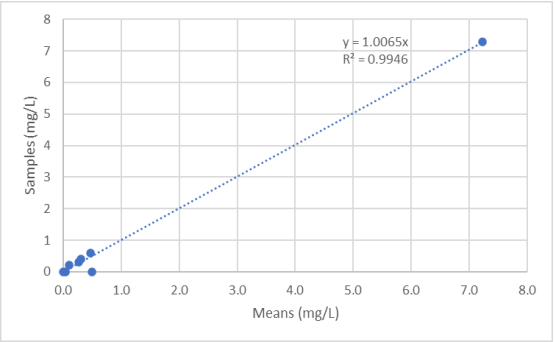


Figure 23: Correlation between the nitrite values of the pool samples and the average for day 30

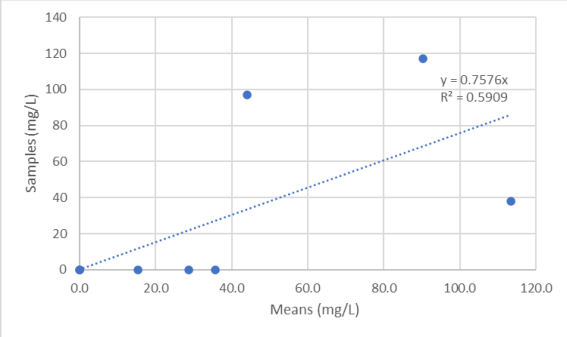


Figure 24: Correlation between the nitrate values of the pool samples and the average for day 15

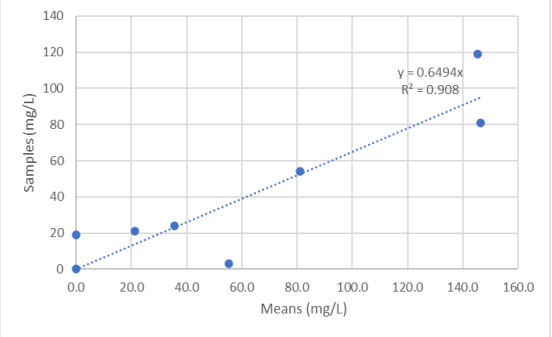


Figure 25: Correlation between the nitrate values of the pool samples and the average for day 30

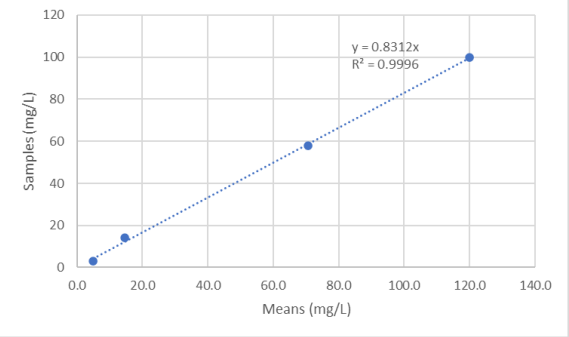


Figure 26: Correlation between the phosphorus values of the pool samples and the average for day 15

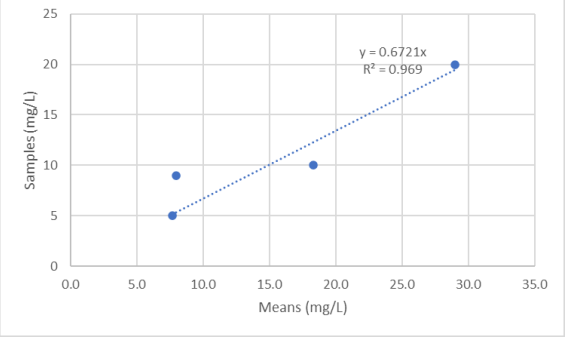


Figure 27: Correlation between the phosphorus values of the pool samples and the average for day 30

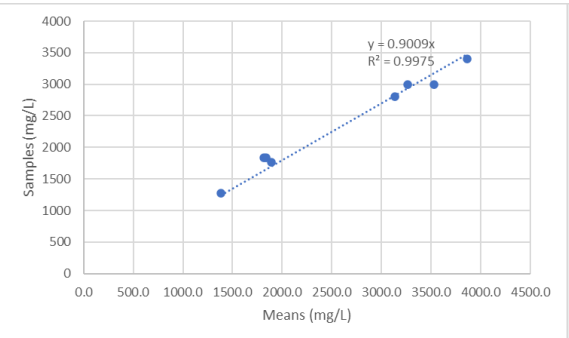


Figure 28: Correlation between the potassium values of the pool samples and the average for day 15

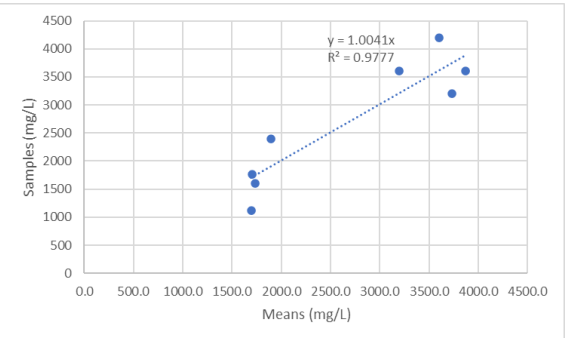


Figure 29: Correlation between the potassium values of the pool samples and the average for day 30

5.2.4.2 Nitrogen

5.2.4.2.1 Total ammonia nitrogen

The concentration of TAN is largely influenced by the type of OM used. Indeed, using chicken droppings releases more TAN in solution than using goat droppings. Moreover, the modalities in anaerobic conditions with free pH do not seem to release TAN in solution. Their value is in fact close to zero (Figure 30 and Figure 31).

For TAN concentrations, the ANOVA conditions are not met. However, OM, type of digestion and pH management all seem to influence the concentration of TAN found in solution. In fact, their p-value is well below 0.001.

Seven different groups emerge from the HSD test:

- A: Chicken/buffer/anaerobic (mean=1321.67 mg/l)
- AB: Chicken/water/anaerobic (mean=1257.33 mg/l)
- B: Chicken/phosphate/aerobic (mean=1022.50 mg/l)
- C: Goat/phosphate/aerobic (mean=581.63 mg/l)
- CD: Goat/phosphate/anaerobic (mean=451.93 mg/l)
- DE: Goat/water/anaerobic (mean=242.33 mg/l)
- E: Goat/water/aerobic (mean=19.68 mg/l) and Chicken/water/aerobic (mean=19.10 mg/l)

Tuckey's HSD test shows the influence of OM on the concentration of TAN released in solution. Indeed, modalities containing chicken droppings have a higher concentration of TAN than modalities containing goat droppings. Furthermore, it confirms that the modalities in aerobic conditions with free pH release almost no TAN in solution. Concerning pH and digestion, their influence is more difficult to describe. Indeed, three groups with hybrid behaviour emerge from the HSD test.

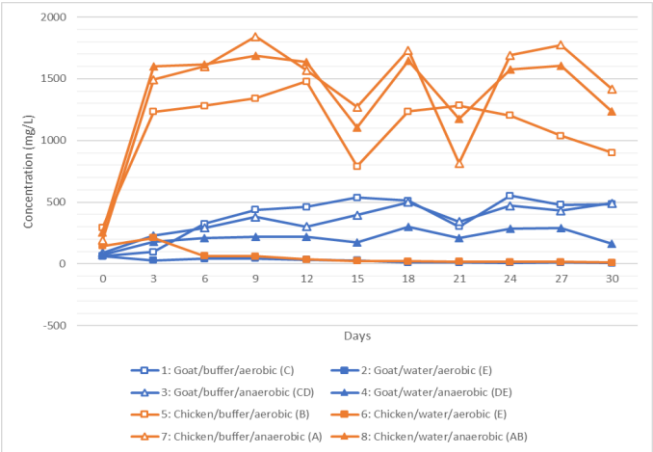


Figure 30: Daily evolution of total ammonia nitrogen for pool samples

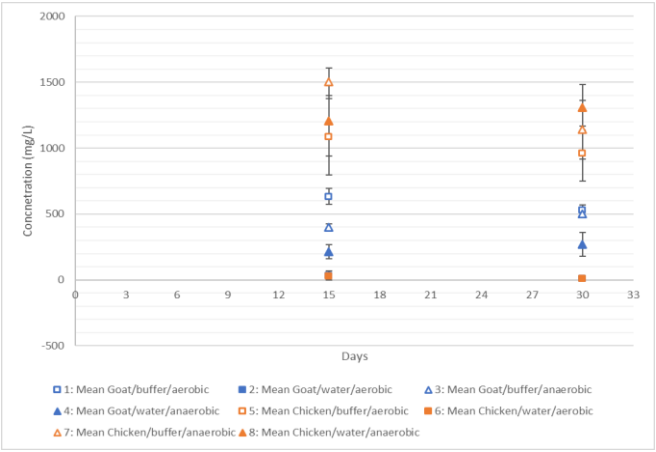


Figure 31: Average values of total ammonia nitrogen replications for days 15 and 30

5.2.4.2.2 Nitrite

The NO_2^- concentration remains extremely low throughout the experiment. The goat/water/aerobic modality increases from day 15 onwards and then has much higher values than the other modalities (Figure 32 and Figure 33).

The ANOVA results do not show any influence of OM, pH, digestion, or days. Indeed, their p-values are all close to 0.05 and the conditions of the ANOVA tested on residues are not respected.

Tukey's HSD test highlights the difference in nitrite concentration for the Goat/water/aerobic modality compared to other modalities. In fact, the latter is in group A while all the other modalities have been placed in group B.

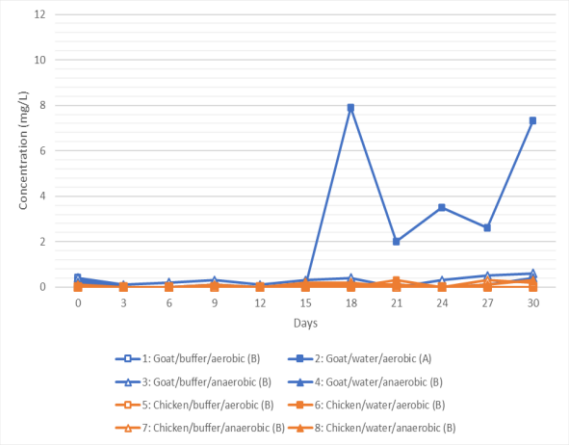


Figure 32: Daily evolution of nitrite for pool samples

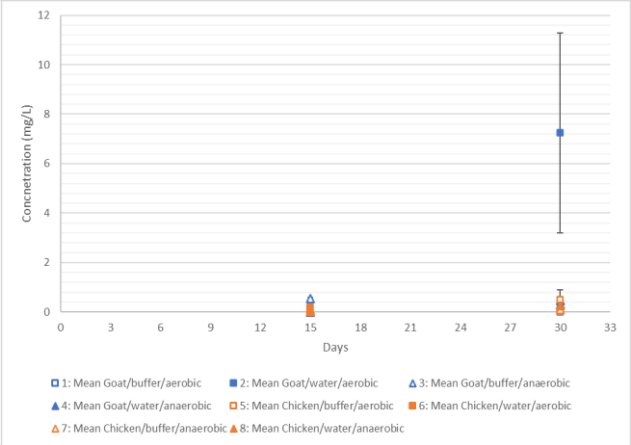


Figure 33: Average values of nitrite replications for days 15 and 30

5.2.4.2.3 Nitrate

The NO_3^- concentration is stable throughout the experiment, except for the chicken/water/anaerobic modality where the NO_3^- concentration increases sharply on day 27 and then suddenly decreases on day 30. As the analysis of replicates was not carried out for day 27, it is not possible to determine whether this peak is due to the increase of a single replicate, in which case it can be considered as an error, or generalised to the three replicates. In the latter situation, no valid scientific explanation could be found. The peak that appeared for the Chicken/water/anaerobic modality is not considered in the statistics since only days 15 and 30 were analysed (Figure 34 and Figure 35).

The ANOVA residuals respect the condition of normality of the population but not that of equality of variances. The influence of OM and pH management on nitrate concentration is highly probable since their p-value is less than 0.001. Concerning digestion and days, no conclusions can be drawn from the ANOVA because the p-values are too close to 0.05 and the ANOVA application conditions are not respected.

Tukey's HSD test separates the modalities into four groups:

- A: Goat/buffer/anaerobic (mean=129.83 mg/l)
- AB: Goat/buffer/aerobic (mean=94.67 mg/l)
- ABC: Chicken/buffer/anaerobic (mean=63.00 mg/l)
- BC: Chicken/buffer/aerobic (mean=45.50 mg/l) and Goat/water/anaerobic (mean=40.50 mg/l)
- C: Goat/water/aerobic (mean=18.33 mg/l), Chicken/water/anaerobic (mean=14.33 mg/l), and Chicken/water/aerobic (mean=0.00 mg/l)

The HSD test shows the influence of pH management. Indeed, pH-controlled modalities have higher concentrations than free pH modalities. Furthermore, the type of OM also plays a role since goat droppings for the same pH management have higher concentrations than hen droppings. Finally, the influence of digestion is also highlighted within modalities with the same pH management and type of OM. In fact, anaerobic modalities have higher nitrate concentrations than aerobic repetitions. However, these conclusions are drawn from the value of the averages given by the HSD test. Yet the HSD test does not consider the averages of the 5 groups to be significantly different since it presents three out of five hybrid groups. The latter therefore have significantly similar averages to certain other groups.

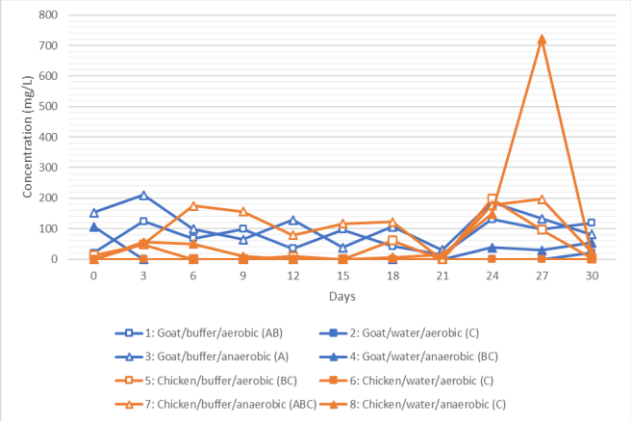


Figure 34: Daily evolution of nitrate for pool samples

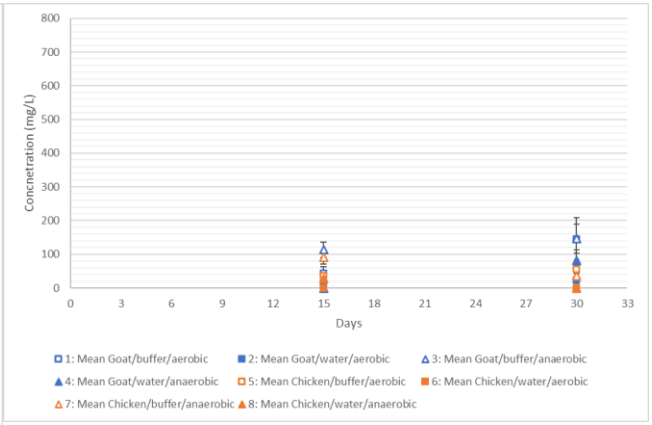


Figure 35: Average values of nitrate replications for days 15 and 30

5.2.4.3 Phosphorus

The analyses for phosphorus were only carried out on samples with a free pH. This is because samples containing phosphate buffer are saturated with phosphorus.

Phosphorus concentrations tend to decrease between the beginning and the end of the experiment.

There are two different groups (Figure 36 and Figure 37):

- Anaerobic modalities: phosphorus concentrations for anaerobic samples increase at the beginning of the experiment and then oscillate downwards.
- Aerobic modalities: the concentrations for aerobic samples decrease during the first three days and then stabilise until the end of the experiment.

The application conditions of the ANOVA are not complied with. However, the results of the ANOVA tend to confirm the presence of two digestion-dependent groups since the p-value of this parameter is less than 0.001. Furthermore, the day also influences the phosphorus concentration.

Regarding the HSD test, three groups can be distinguished:

- A: Goat/anaerobic (mean=74.50 mg/l)
- AB: Chicken/anaerobic (mean=44.50 mg/l)
- B: Chicken/aerobic (mean=11.33 mg/l) and Goat/aerobic (mean=6.33 mg/l)

The HSD test therefore offers a more nuanced view of the two groups (anaerobic and aerobic) proposed by the analysis of Figure 36. Indeed, the average for the Chicken/anaerobic modality is not significantly different from the averages for the aerobic modalities. However, the average values analysis still shows that, in general, phosphorus concentrations are higher in anaerobic conditions.

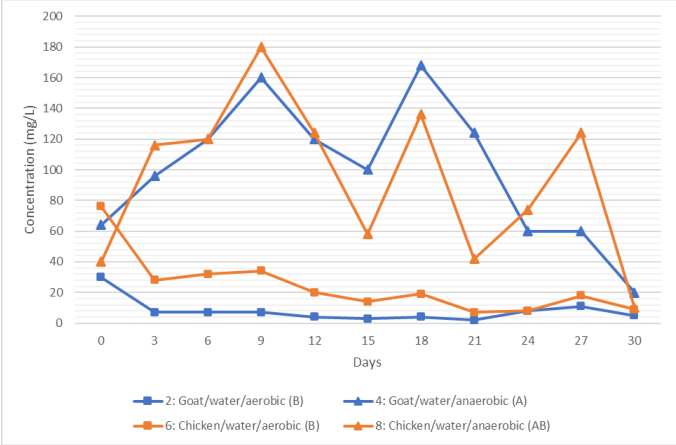


Figure 36: Daily evolution of phosphorus for pool samples

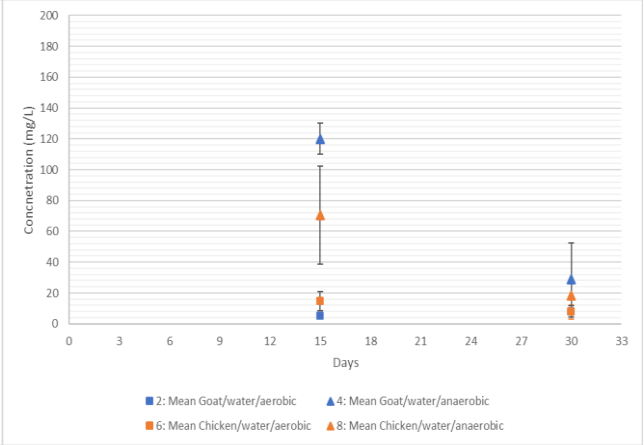


Figure 37: Average values of phosphorus replications for days 15 and 30

5.2.4.4 Potassium

Potassium concentrations increase for the first three days and are then stable. Two groups differ (Figure 38):

- Methods containing chicken droppings: their average concentrations for days 15 and 30 range from 3366.67 to 3700.00 mg/L.
- Methods containing goat droppings: their average concentrations for days 15 and 30 range from 1540.00 to 1866.67 mg/L.

The statistics carried out for this parameter confirm the presence of these two groups. In fact, even if the normality of the ANOVA residues is not respected, the OM is the only parameter with a p-value lower than 0.001. The influence of the OM is therefore confirmed by the ANOVA. In addition, Tukey's HSD test also divides the 8 modalities into two groups. The first group (A) includes all modalities containing chicken droppings, while the second group (B) includes modalities containing goat droppings.

No other influence is evident from the statistics in this study.

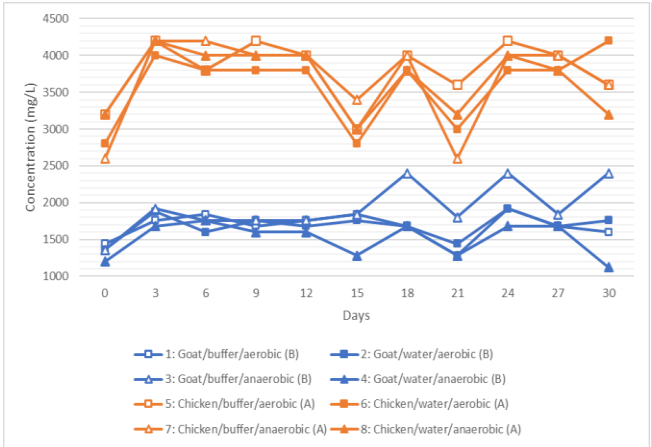


Figure 38: Daily evolution of potassium for pool samples

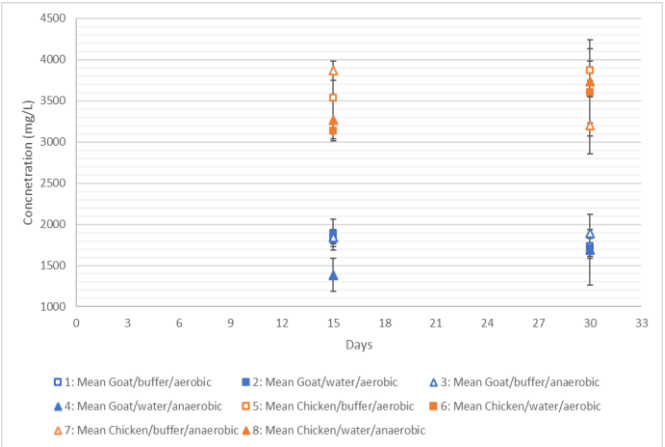


Figure 39: Average values of potassium replications for days 15 and 30

5.2.4.5 Synthesis NPK

To conclude, the statistical analyses have, on the one hand, highlighted the preponderant influence of the type of OM used on the TAN and K concentrations. On the other hand, they showed the importance of digestion on phosphorus concentrations. Concerning nitrite and nitrate concentrations, the analysis of the results was made difficult by the low values obtained during experiment 1 and by the results of one modality whose results were strongly different from those of the other modalities (Goat/water/aerobic for nitrite and Chicken/water/anaerobic for nitrate).

The NPK concentrations for the 8 modalities on day 30 are shown in Table 14.

Table 14: Final NPK value for the 8 modalities of experiment 1

	TAN	NO_2^-	NO_3^-	P	K
1	529.6	0.0	145.3		1733.3
2	7.8	7.2	21.3	7.7	1706.7
3	502.1	0.5	146.3		1893.3
4	269.7	0.3	81.0	29	1693.3
5	959.7	0.5	55.3		3866.7
6	10.5	0.0	0.0	8	3600.0
7	1140.3	0.1	35.7		3200.0
8	1307	0.3	0.0	18.3	3733.3

5.3 EXPERIMENT 2: CREATION OF A NUTRITIVE SOLUTION FROM MINERALIZATION

The objective of the second experiment is to create a nutritive solution from the mineralisation of chicken and goat droppings in large quantities to test its effectiveness on the growth of lettuce in hydroponics. In this experiment, the temperature is not controlled, and the pH of the buffered modalities is controlled by an IKS. The trends analysed in Experiment 1 were also found in Experiment 2. The graphs of the daily evolution of pH, temperature, EC and NPK concentrations for the four modalities can be found in Appendix 6.

The results for the 4 modalities are presented in Table 15.

Table 15: Final NPK value for the second experiment

	Settings	TAN	NO2	NO3	P	K
A	Goat/water	104.5	0.3	56	13	1600
B	Goat/buffer	264.6	0.1	0		1360
C	Chicken/buffer	1257	0	0		3600
D	Chicken/water	262	0.1	0	3	3600

6 DISCUSSION

As a reminder, the main objective of this work was to create a nutritive solution for a hydroponics system from goat and chicken droppings under controlled conditions. Several sub-objectives were formulated. The first of these, the achievement of a state of the art on fertilization, was dealt with in the "Literary review". The results of the remaining three sub-objectives will be discussed in this chapter.

This chapter is therefore divided into four parts. The first part will discuss the performance of the digestion system set up as well as the buffering and sampling protocols. It will try to make suggestions for improvement to make the system more efficient while keeping the objective of using low tech materials.

The second part of this chapter will focus on the mineralization process itself and will try to explain the different results obtained concerning the NPK concentrations, pH and EC during preliminary test 2 and both experiments. A comparison will be made between these results and those obtained by Thomas Tomson on the digestion of fish farm sludge, but also with the results obtained by Félicien Munungakatebe on the digestion of goat and chicken droppings in the Democratic Republic of Congo.

The third part will then evaluate the fertilizing characteristics of the nutrient solutions obtained and the impact they could have on the growth of plants such as lettuce.

Finally, the last part will present the different perspectives being considered for this work. It will describe the experiments that will be carried out as a continuation of this study. In addition, it will endeavour to present new studies that could complement current knowledge on bioponics.

6.1 DIGESTION SYSTEM

6.1.1 Performance

The performance of the system is evaluated solely on the observations made during the experiment but also on the temperature measurements in the buckets. The observations allow the identification of three main imperfections in the system.

Firstly, despite the installation of a flow in the water baths and of a tarpaulin covering the buckets, the temperature within the different buckets is not homogeneous. Indeed, the ANOVA shows the main influence of the type of digestion (aerobic/anaerobic) but also of the day on temperature fluctuations. Concerning the first parameter, the difference in temperature is explained by the presence of a cover on the buckets in anaerobic conditions, as opposed to the buckets in aerobic conditions. The heat loss is therefore lower under anaerobic conditions and the nutrient solution has a slightly higher temperature.

The cause of the day's influence is more debatable. Indeed, the average temperature of the buckets decreases as the days go by. There are two possible explanations for this drop in temperature. Firstly, this heat loss could be due to the poor insulation of the water baths. The room temperature would therefore have had an impact on the temperature of the buckets. This explanation is reinforced by the study of outside temperatures measured at the Uccle weather station during the month of September 2020 (Figure 16). Indeed, the temperature peaks of 15/09 and 20/09 seem to correspond to the temperature peaks observed on days 12 (16/09) and 18 (22/09) of experiment 1 (Figure 18). Moreover, the temperature drops of 17/09 also seem to correspond to the temperature drop observed on day 15 (19/09). The impact of outside temperature could thus be observed posteriori on the temperature of the modalities of experiment 1 with a two-day lag. To confirm this hypothesis, the temperature in Gembloux and the temperature of the room

where the mineralisation system is located could be measured every three days (at the same time as the experimental monitoring). The second explanation would be a loss of power of the resistors during the experiment due to a technical problem. This hypothesis should only be considered if the first explanation is refuted.

The second imperfection observed was the deposits of OM remaining on the walls of the aerobic buckets due to the production of foams during the mineralization process. In some cases, the foam even overflowed the bucket and OM ended up in the water bath. These losses of raw material certainly influenced the mineralization process.

Lastly, an improvement can also be made in the sampling protocol. Indeed, two problems have appeared. Firstly, the levelling of the buckets before sampling is carried out according to a line drawn on the buckets corresponding to the 3-litre level. It is likely that a deviation was made during the tracing process. In addition, as the buckets were only slightly transparent, the levelling was not accurate. Secondly, as presented in the equipment and method section, the sampling in Experiment 1 was carried out by mixing repetitions for the same modality because time did not allow all the repetitions to be studied separately. As already presented in the results (5.2.4.1 Correlation between NPK concentration results for pool samples and average replications for days 15 and 30), the precision of this sampling technique is not perfect and presents small deviations mainly when the measured concentrations were low. For higher concentrations, this technique proved to be representative of the average of the three replicates for each modality.

6.1.2 Improvement

Based on these observations, several avenues for improvement can be explored.

Firstly, it would be interesting to improve the insulation of the system to better control the temperature. The shelf supporting the water baths could be enclosed in an insulated cabinet. Two possibilities can be considered. The cheapest but also the least effective would be to cover the entire shelf with a tarpaulin (not just the water baths). The second solution is to use insulation sheets to completely enclose the water baths. In addition, one of the observations made is that buckets in anaerobic conditions, being closed with a lid, generally have a higher temperature than buckets in aerobic conditions. It would therefore be potentially interesting to also seal the aerobic buckets, either with a plastic film or with a lid with a hole in the middle to allow the tubes to pass through, which allows the oxygenation of the buckets (Tomson, 2019).

Another problem with the system used is the loss of raw material due to the formation of foam. This could be limited by closing the buckets in aerobic conditions to avoid overflows. Another solution would be to reduce the volume of solutions in the bucket or to increase the size of the buckets. Concerning the particles of OM that remain stuck to the walls, they can be limited by cleaning the walls daily with a scraper.

Finally, regarding sampling, the buckets could be levelled more precisely if they were perfectly transparent and graduated. Furthermore, it would be interesting to carry out the experiment by analysing each repetition for each day. Indeed, by analysing each replicate, the results obtained would be more representative of the real nutrient concentrations and the point errors in the measurements could be more easily identified, unlike in a pool sampling. However, the analyses are very time-consuming. Indeed, the analysis of a sample for a single nutrient (NH_4^+, NO_2^-, NO_3^-, P or K) takes an average of 6 to 7 minutes.

6.2 MINERALIZATION PROCESS

The performance of the mineralisation process is evaluated by comparing the results of pH, EC and NPK concentrations obtained in Preliminary Test 2 and the two experiments under aerobic and anaerobic conditions. The figures showing these results are in Appendix 5, in Results and in Appendix 6 for Preliminary Test 2, Experiment 1 and Experiment 2 respectively. The comparison is made only on the general shape of the curves. In fact, the modalities between the test and the experiments are not identical since only the modalities at pH 6 are selected for preliminary test 2, whereas the pH is 6.5 for the experiments. Moreover, experiment 2 is carried out on larger volumes (100L instead of 3L) without temperature control.

In general, the results obtained from the test and the experiments are similar in terms of physiognomy but also in terms of values. The conclusions obtained from the analysis of the results of experiment 1 can therefore be generalised to pre-test 2 and experiment 2. The system put in place therefore allows for a repeatability of the mineralisation process between the different experiments. However, in view of the results obtained, particularly concerning nitrogen, the mineralisation process is not optimal. The results of experiment 1 will therefore be discussed and suggestions for improvement and further research will be put forward.

6.2.1 pH

The study of pH has enabled us to distinguish two main groups: aerobic and anaerobic modalities. On the one hand, the group of aerobic modalities has a higher average pH and slightly increased during the experiment. On the other hand, the anaerobic modality group had a generally lower pH, decreasing during the 3rd day before slowly increasing until the end of the experiment. The general tendency to increase for both types of digestion could be explained by the evolution of the pH within the nitrogen cycle. Indeed, the transformation of organic nitrogen into ammonium in solution causes the pH to increase (Van Bochove, 1993). Concerning anaerobic digestion, the decrease in pH at the beginning of the experiment could be explained by the acidogenesis phase which, by producing organic acids and hydrogen, decreases the pH (Aoun et al., 2015).

6.2.2 EC

PH management has shown its predominant influence on the EC value. The pH-controlled modalities (pH 6 or 6.5) were carried out with a phosphate buffer that has a specific amount of salt in solution (4.3.1.3.1 Phosphate Buffer Protocol). In contrast, the free pH modalities contained demineralised water. As its name indicates, the latter has no minerals in solution. The influence of pH management on EC is perfectly understandable since EC increases with the concentration of salts in solution. Then, within the modalities with the same pH management, the type of organic matter used also plays a key role in the evolution of EC. In fact, modalities containing hen droppings generally have a higher EC than modalities containing goat droppings. This conclusion is in line with the literature, as chicken droppings are richer in nutrients than goat droppings (Rosen et al., 2005).

6.2.3 Nitrogen

The evolution of the different forms of nitrogen is analysed. Firstly, OM seems to have an important influence on the concentration of TAN found in solution. Indeed, TAN concentrations are higher for modalities containing chicken droppings than for those containing goat droppings. This observation is in line with the results obtained for EC and confirms the nitrogen richness of chicken droppings compared to goat droppings. In addition, digestion also seems to play a role in the release of nitrogen in solution.

According to the literature, anaerobic digestion results in a high decomposition rate of OM which leads to a high release of nutrients in solution (Tomson, 2019). However, the results obtained during the experiments do not allow such a clear-cut conclusion to be reached. Indeed, statistical analyses of TAN have highlighted the effect of digestion but do not allow a precise description of its impact on TAN concentrations. Further statistical analyses would allow this description.

Concerning the oxidised forms of nitrogen, the nitrite concentrations measured are extremely low for all the analyses carried out. Nitrate concentrations, although slightly higher than nitrite concentrations, are also low. Furthermore, the nitrate evolution during the test and experiments has a high variability. These observations are due to a low mineralisation of ammoniacal nitrogen to nitrate. Two hypotheses were put forward to explain why the nitrification of ammoniacal nitrogen did not take place.

Firstly, low nitrification may be due to a low or non-existent population of nitrifying micro-organisms. In the experiments carried out, the raw material was oven-dried before use. This drying could have killed the micro-organisms present in the fresh material. Mineralisation would therefore proceed more slowly, while a new population of nitrifying micro-organisms develops (Hsieh et al., 2019). This hypothesis is supported by the work of Thomas Tomson and Félicien Mununga katebe who, by working with fresh raw materials, have obtained higher ratios between nitrite and nitrate concentrations in relation to the amount of TAN released in solution (Tomson, 2019). To verify this hypothesis, analyses of the microorganisms present in the droppings of fresh and dried hens and goats could be carried out.

The second hypothesis that may explain the low nitrite and nitrate concentrations is nitrogen volatilization. Several reactions may be possible. Firstly, nitrogen can volatilize as NH3. This loss of nitrogen is due to the non-stabilisation of NH3 into NH4+ and can be caused by an increase in pH. This reaction would explain the ammonia odours present in the room during the first days of the tests. However, the evolution of the pH of the test and of the two experiments does not allow the identification of such a reaction since a strong increase in pH was not observed for all modalities. The second possible reaction is anammox. Anammox uses the NH4+ and NO2- present to form nitrogen gas in the N2 form. However, this reaction only takes place under anaerobic conditions and is therefore not applicable for buckets under aerobic conditions. Another phenomenon is therefore the source of the weak nitrification of ammoniacal nitrogen. The hypothesis of volatilisation of nitrogen in NH3- or N2 form should therefore only be considered if the first hypothesis is refuted.

6.2.4 Phosphorus

Phosphorus concentrations are influenced by digestion. Anaerobic digestion seems to help the release of phosphorus in solution. In fact, as shown in the analysis of TAN concentrations, anaerobic digestion allows a higher decomposition of OM than anaerobic digestion (Tomson, 2019). Moreover, the phosphorus concentrations obtained during the test and experiments are extremely low compared to the other two nutrients (nitrogen and potassium). Moreover, their behaviour varies between the test and the two experiments. This behaviour is difficult to explain during the tests carried out. One of the hypotheses that can be formulated to explain the variations in phosphorus is the presence of phosphate ions. Indeed, calcium, iron or aluminium can react with phosphate to form a salt that can precipitate. The phosphorus is then no longer in solution (Youcef et al., 2005). The solubility of the salts formed depends on the temperature and pH of the solution. This phosphate precipitation reaction is the same as that used in wastewater treatment (Tétreault, 2015). However, more research is needed to assess the likelihood of this hypothesis.

6.2.5 Potassium

In the end, the potassium concentrations obtained in the tests are much higher than for the other two nutrients. No explanation was found for this difference. As with EC and TAN, potassium concentrations for modalities containing chicken droppings are higher due to the nature of the droppings.

6.3 NPK CONCENTRATION AND FERTILIZING CHARACTERISTICS OF THE ORGANIC FERTILIZER CREATED

The nutritional characteristics of the solution are evaluated based on the results obtained from Experiment 2.

A first estimate of the concentrations of salts in solution is given by the EC. The optimal value of the latter is between 1 and 4 mS/cm depending on the crop studied for hydroponics (Hardeep et al., 2016). In the case of experiment 2, the EC varies between 2.31 and 13.24 mS/cm. Indeed, only Goat/water (A) and Chicken/water (D) solutions have ECs below 4 mS/cm.

The NPK concentrations of the solutions created (Table 16) are compared to an average NPK content obtained from several nutrient solutions found in the literature (Table 17). Among the solutions created, the goat/water solution seems to have the most favourable fertilisation characteristics for obtaining a healthy crop. However, the NH4+ concentrations remain high, varying between 41.7 and 502.4 mg/l, compared to an average of 26 mg/l in the literature. Potassium concentrations are also extremely high. In fact, the literature mentions an average of 246 mg/l, while the potassium concentrations of the solutions created vary between 1360 and 3600 mg/l. On the other hand, NO3- and phosphorus concentrations are low. In fact, the solutions created do not contain NO3-, whereas the solutions found in the literature have an average of 156 mg/l. Concerning phosphorus concentrations, solutions with a free pH have 10 to more than 15 times less phosphorus than the average solutions in the literature.

These results therefore pose problems because the solutions are not balanced. On the one hand, the use of these solutions would create problems of deficiencies in phosphorus, for free pH modalities, and in NO_3^-, the form of nitrogen most assimilable by plants. Nitrogen deficiencies are identified by reduced growth and progressive yellowing beginning on old leaves, while phosphorus deficiencies are characterized by stunting of the plant and dark green coloration on mature leaves (Resh, 1978).

Besides, in organic hydroponics, a fertilizer containing only nitrogen in the form of NH_4^+ will significantly suppress plant growth (Fang et al., 2018). In addition, phosphorus toxicity, for pH-controlled modalities, and potassium toxicity may also occur. Excess potassium could lead to deficiencies in magnesium, zinc, or iron. On the other hand, there are no symptoms of phosphorus toxicity. However, deficiencies in copper and zinc can sometimes occur (Resh, 1978).

Table 16: NPK concentration of the solutions created in experiment 2 in mg/l

Modalities	N as NH4+	N as NO3-	P as PO43-	K
A: Goat/water	41.7	13	13	1600
B: Goat/buffer	105.8	0	/	1360
C: Chicken/buffer	502.4	0	/	3600
D: Chicken/water	104.7	0	3	3600

Table 17: NPK concentration of nutrient solutions found in the literature in mg/l (Resh, 2013)

References		N as NH4+	N as NO3-	P as PO43-	K
Jones & Shive (1921)		39	204	65	102
Hoagland & Arnon (1938)		14	196	31	234
Purdue (1948)	A	28	70	63	390
	B	28	140	63	390
	C	14	224	63	390
Schwartz (Californie)		15	196	31	234
Schwartz (New Jersey)		20	126	71	90
CDA	A	33	93	36.7	209
Saanichton	B	33	135	36.7	209
B.C.Canada	C	33	177	36.7	209
MEANS		**26**	**156**	**50**	**246**

To conclude, an imbalance exists between the NPK concentrations obtained in Experiment 2. This imbalance could lead to nitrogen and phosphorus deficiencies (for free pH modalities) and potassium and phosphorus toxicities (for controlled pH modalities). These results, although of little interest for fertilization in hydroponics, could be used in aquaponics where potassium is the limiting element (Goudeau, 2020)

6.4 PERSPECTIVES

As presented in the introduction, this work is part of Félicien Munungakatebe's thesis. The latter aims to exploit polluted land for food in the Democratic Republic of Congo. An experiment like those presented in this work has therefore already been carried out by Mr Munungakatebe under uncontrolled conditions and with locally available materials to create the digestion system.

Further research will be carried out following this study. Indeed, the nutritive solutions realized during Experiment 2 will be tested on hydroponic lettuce culture and a comparison with a commercial nutritive solution will be made. In addition, analyses of the microbiological and NPK composition of dried goat and chicken droppings are also planned.

A research project "Innovative irrigation systems in the Saharawi refugee camps in Tindouf (South-West Algeria)" has also been funded to continue this study with a direct application of bioponics in developing countries.

In addition to this research, many other subjects of study could be explored in the field of bioponics. Indeed, the possibilities of creating nutrient solutions from OM are extremely broad. It would be interesting, for example, to carry out the mineralisation of other animals' excreta. Studying the impact of the type of breeding and feeding on the NPK concentrations obtained after mineralisation would also be relevant. Furthermore, a more in-depth analysis of the parameters influencing mineralisation could increase knowledge of bioponics and allow the optimisation of the digestion system.

Ultimately, this work is carried out with the long-term objective of the establishment and daily use of bioponics systems in developing countries where the soil is not usable for natural (flooding, erosion, ...) or anthropogenic (soil pollution) reasons.

7 CONCLUSION

Since its revival in urban agriculture, hydroponics has shown its many advantages in terms of space, productivity, and water savings. Unfortunately, it has a major disadvantage: its dependence on mineral fertilisers. Bioponics is an innovative technique that aims to replace mineral fertilisers in hydroponic systems with organic fertilisers. These can be produced in different ways with many possible raw materials. This flexibility makes it possible to use bioponics worldwide. Indeed, in addition to its use in cities, it can also have a direct application in developing countries allowing the cultivation of vegetables on soils not suitable for agriculture. This asset of bioponics is still little studied. The aim of this work has therefore been to evaluate the possibilities and effectiveness of a system of mineralisation of organic matter allowing the creation of a nutritive solution from the excrement of goats and chickens, commonly found in the Democratic Republic of Congo.

To achieve this objective, several parameters were studied such as OM concentration, type of digestion (aerobic, anaerobic), pH management and type of buffer used. The impact of these parameters on the system was analysed by studying pH, temperature, EC and NPK concentrations released in solution.

The results of this master thesis allowed the creation of a functional mineralisation system of low technicality that allows the replication of the digestion process. However, three weaknesses were also identified. Indeed, heat losses were observed during the experiment. These could be linked to the poor insulation of the system set up. In addition, losses of raw materials could also be observed. These are mainly due to the presence of bubblers which lead to the creation of foam and the overflow of the reactors. Sealing these reactors while allowing the air flow to pass through, or the performance of the experiment on smaller volumes would allow this problem to be resolved. Finally, the sampling technique implemented has allowed significant time saving but is not optimal. The study of the three repetitions independently of each other would be more interesting and would allow more precise results over the duration of the experiment.

Moreover, the mineralisation process carried out by the system is not optimal. Indeed, nitrogen nitrification did not take place during the experiments set up. Furthermore, the balance between the concentrations of nitrogen, phosphorus and potassium is not ideal for a complete nutrient solution for optimal plant growth. Indeed, NH_4 concentrations are 2 to 25 times higher than the values found in the literature, while potassium concentrations are 5 to 15 times higher. On the other hand, the nitrate concentrations of the solutions created are non-existent and the phosphorus concentrations are 5 to 15 times lower than the values found in the literature.

Despite these shortcomings, several interesting conclusions have been drawn. Firstly, the use of chicken droppings resulted in a higher mineral concentration in the nutrient solution than goat droppings. Secondly, anaerobic digestion seems to have advantages in terms of phosphorus concentrations compared to aerobic digestion. Finally, the presence of a phosphate buffer, in addition to increasing the EC, seems to have a positive impact on the nitrogen concentrations found in solution.

To conclude, the system currently in place therefore does not allow the creation of an ideal organic solution for hydroponics. The study of the impact of the nutrient solutions on lettuce growth will allow the precise identification of deficiencies and toxicities encountered by the plant. However, due to its potassium enrichment, the solution created could be used as a supplement in aquaponic systems that are regularly deficient in potassium. It is therefore important to continue to improve the system and to study in greater depth the different parameters that influence the system to allow a precise characterisation of the mineralisation reactions.

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9 APPENDICES

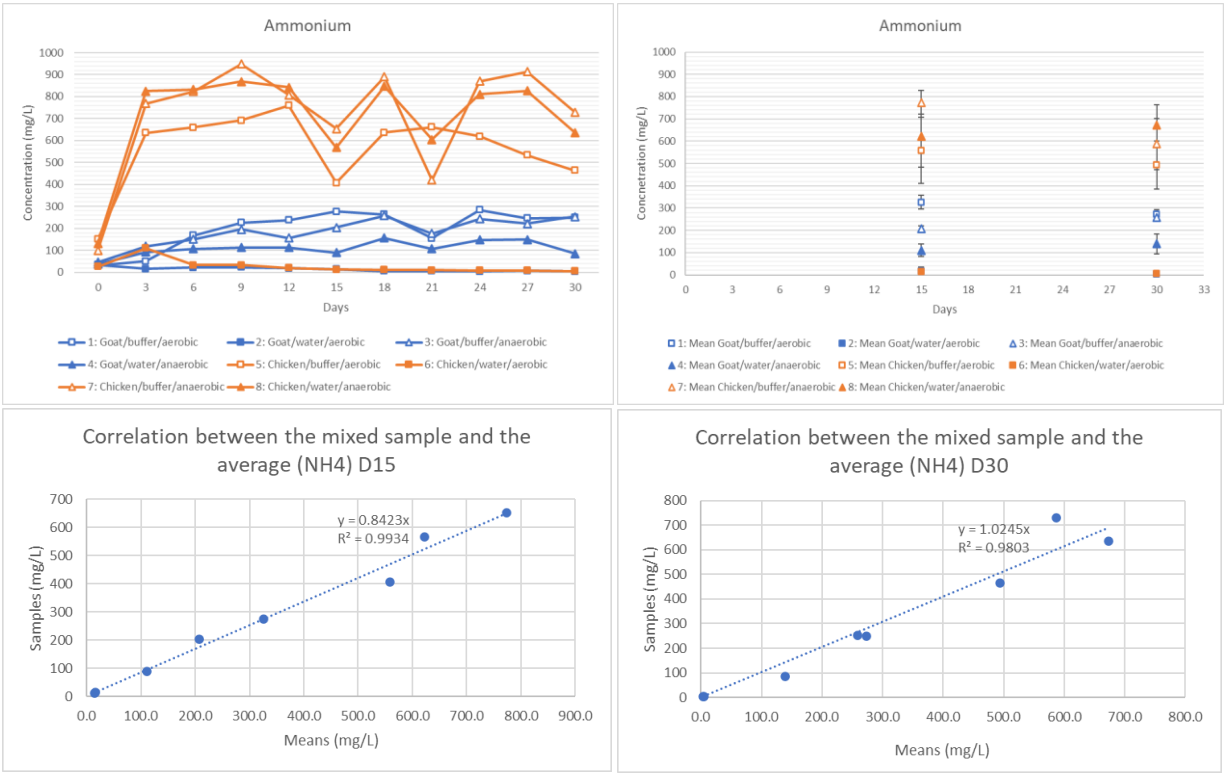
APPENDIX 1

Appendix 1: Calculation of the percentage of dry matter

CHICKEN	Fresh weight	Tare	Gross weight	Dry weight
1	3.33	0.60	3.52	2.92
2	3.77	0.67	3.98	3.31
3	3.29	0.65	3.51	2.86
Total				9.09
% Mean				87.49%
GOAT				
1	3.41	0.86	3.98	3.12
2	2.96	0.76	3.49	2.73
3	3.41	0.64	3.76	3.12
Total				8.97
% Mean				91.72%

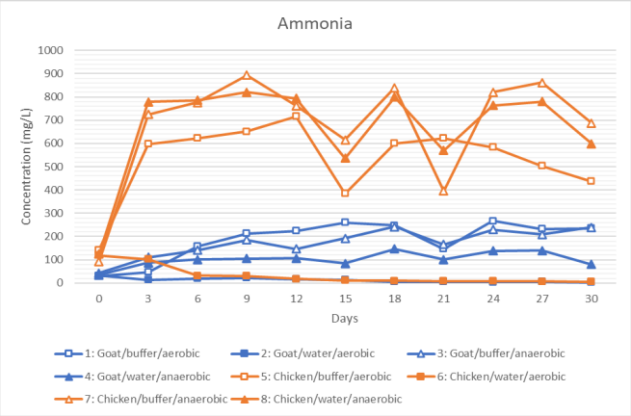
APPENDIX 2

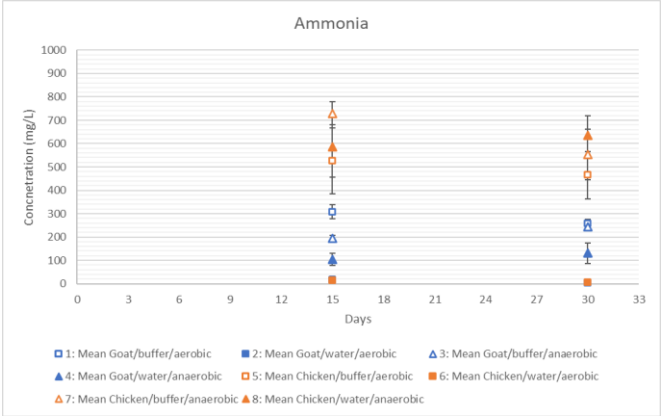
Appendix 2: Results of experiment 1 on ammonium

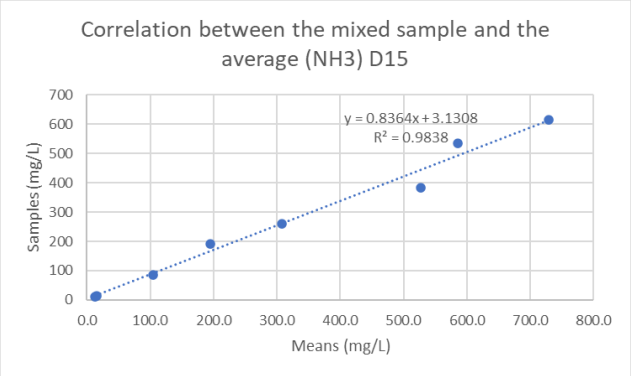


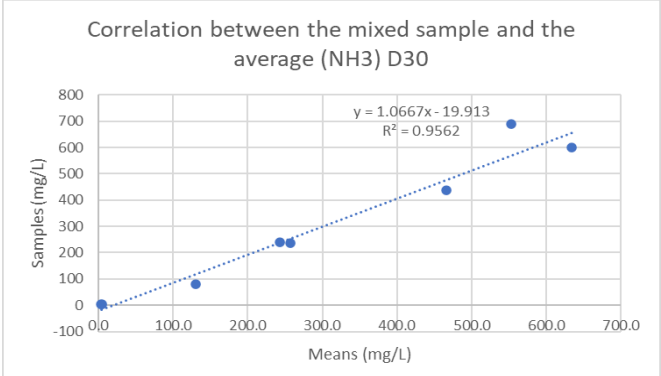
APPENDIX 3

Appendix 3: Results of experiment 1 on ammonia









APPENDIX 4

Appendix 4: Results of preliminary test 2 at the end of de experiment

Ech	Modalities				NPK				
	OM	% dry matter	Digestion	pH	TAN	NO3-	NO2-	P	K
1	Goat	2.5	aerobic	5.5	304.2	0	0		1120
2	Goat	2.5	aerobic	6	325.6	0	0		1040
3	Goat	5	aerobic	6	444.4	0	0		2640
4	Goat	7.5	aerobic	6	441.2	6	0		3760
5	Goat	10	aerobic	6	653	0	0		8600
6	Goat	2.5	aerobic	6.5	275.4	0	0		1520
7	Goat	2.5	aerobic	7	253.2	0	0		1640
8	Goat	2.5	aerobic	No buffer	15.8	0	0	160	1280
9	Goat	5	aerobic	No buffer	50.2	0	0	80	1440
10	Goat	7.5	aerobic	No buffer	84.8	0	0	120	2880
11	Goat	10	aerobic	No buffer	131.4	0	0	180	5400
12	Goat	2.5	anaerobic	6	190.2	82	0.1		1120
13	Goat	2.5	anaerobic	No buffer	248.4	0	0	116	1440
14	Chicken	2.5	aerobic	5.5	676.8	0	0		960
15	Chicken	2.5	aerobic	6	866.4	0	0		2640
16	Chicken	5	aerobic	6	1073	38	0		6400
17	Chicken	7.5	aerobic	6	354	0	0		8400
18	Chicken	10	aerobic	6	655	0	0		12800
19	Chicken	2.5	aerobic	6.5	959.6	0	0		3200
20	Chicken	2.5	aerobic	7	317.2	0	0		3400
21	Chicken	2.5	aerobic	No buffer	110.4	0	0	12	2600
22	Chicken	5	aerobic	No buffer	271.2	0	0	4	5000
23	Chicken	7.5	aerobic	No buffer	230	0	0	16	9800
24	Chicken	10	aerobic	No buffer	1278	0	0	2	12400
25	Chicken	2.5	anaerobic	6	849.2	34	0		3200
26	Chicken	2.5	anaerobic	No buffer	690	0	0.2	0	2600

APPENDIX 5

Appendix 5: Results of preliminary test 2 for pH, EC and NPK concentrations

APPENDIX 6

Appendix 6: Results of experiment 2 for pH, temperature, EC and NPK concentrations