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# Microalgae for livestock wastewater treatment: an overview

Rania Karoui

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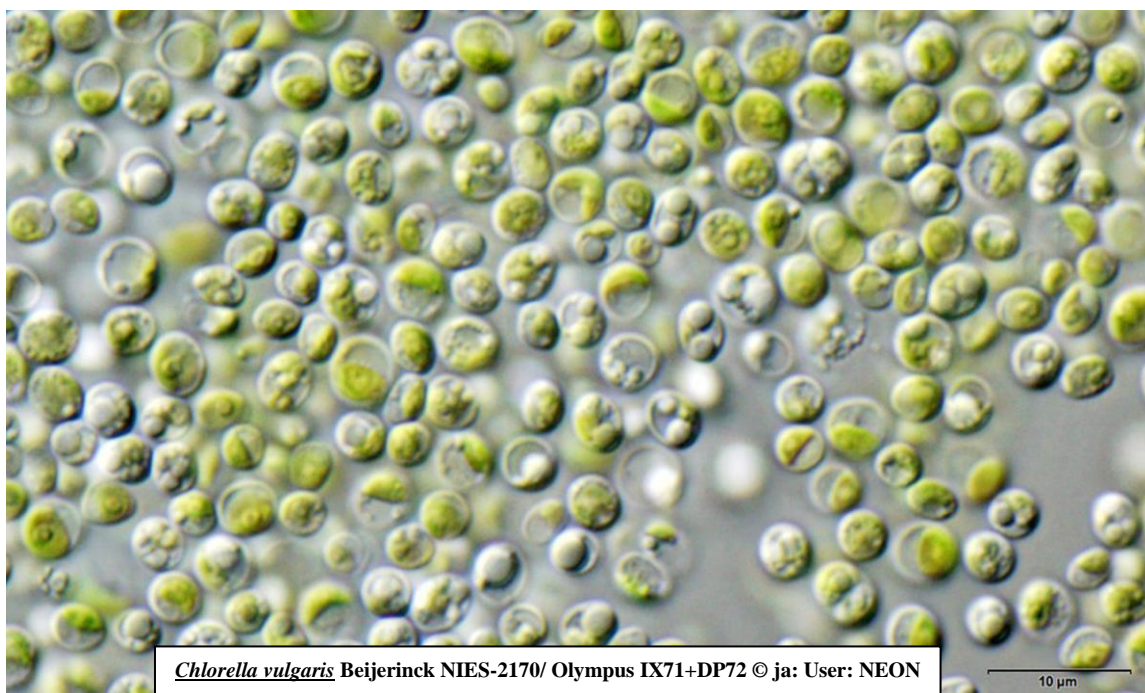
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## Microalgae for livestock wastewater treatment: an overview.

Par : Rania KAROUI



**Soutenu à Rennes le 16/03/2021**

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*Les analyses et les conclusions de ce travail d'étudiant n'engagent que la responsabilité de son auteur et non celle d'AGROCAMPUS OUEST*

# Microalgae for livestock wastewater treatment: an overview

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*My sincere thanks go also to my family, especially my parents Faïza and Khalifa and my brother Anas, as well as my friend Bega for their continuous caring and support.*

# Foreword

The initial objective of the internship was to determine whether air-washing water from hog buildings, which is highly (and exclusively) nitrogenous, can be used for the production of microalgae (*Chlorella vulgaris*) in photobioreactors. Experiments should first have been carried out in batch reactors in order to study, primarily, the influence of the ammonium ion concentration on the cultivation of *Chlorella vulgaris*. Water from a biological scrubber used in the laboratory (IMT-Atlantique) would have been used for the treatment of ammonia and would thus have constituted a real effluent, representative of the loaded water obtained in the livestock farming. Various parameters such as the duration and the intensity of illumination, as well as the temperature and the algal biomass produced, would have been investigated.

The bibliographical part of the subject dealing with photobioreactors for the treatment of, piggery effluents by algae and in particular *Chlorella vulgaris* have been already completed.

The detailed experimental protocol for the preliminary manipulations in Erlenmeyer's, as well as for the manipulations in batch was established.

Unfortunately, following the Covid-19 pandemic, access to the Lab has become forbidden because of the lockdown, making it impossible to continue on the same initial subject, which included a huge experimental part.

The new objective, which now consisted in carrying out a bibliographical synthesis on the treatment of liquid effluents generated by intensive pig, poultry and cattle farms with high nitrogen and phosphorus loads via the use of micro-algae, has led to the completion of the present work.

# Glossary:

**C:** carbon

**Ca<sup>2+</sup>:** calcium

**CI:** confidence interval

**Cl<sup>-</sup>:** chloride

**COD:** Chemical oxygen demand

**Cu<sup>2+</sup>:** copper

**DHA:** docosahexaenoic acid

**DM:** dry material

**EPS:** extracellular polymeric substance

**F<sup>2+</sup>:** iron

**HNO<sub>3</sub>:** Nitric acid

**H<sub>3</sub>PO<sub>4</sub>:** Phosphoric acid

**Hrt, HRT:** hydraulic retention time

**K<sup>+</sup>:** potassium

**LED:** Light-Emitting Diode

**MBPBR:** microalgae biofilm photobioreactor

**Mg:** magnesium

**MJ:** megajoule

**Mn<sup>2+</sup>:** manganese

**N:** nitrogen

**Na<sup>+</sup>:** sodium

**NH<sub>3</sub>, NH<sub>3</sub>-N:** ammonia

**NH<sub>4</sub><sup>+</sup>, NH<sub>4</sub>-N:** ammonium nitrogen

**NO<sup>2-</sup>:** nitrite

**NO<sup>3-</sup>:** nitrate

**P:** phosphorus

**PBR:** photobioreactor

**PO<sub>4</sub><sup>3-</sup>, PO<sub>4</sub>-P:** total orthophosphates

**RABPBR, RAB:** rotating microalga biofilm photobioreactor

**SO<sub>4</sub><sup>2-</sup>:** sulphate

**T<sup>°</sup>:** temperature

**TN:** total nitrogen

**TP:** total phosphorus

**Zeolite:** microporous, aluminosilicate minerals commonly used as commercial adsorbents and catalysts and as fertilizer, odor absorbent, and feed additive in agriculture (Król, 2020).

**Zn<sup>2+</sup>:** zinc

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## Introduction

The growing production of livestock manures represent an issue for sustainable agricultural development (Awasthi et al., 2019). According to (Williams, 2013), 7% of worldwide greenhouse gas emissions are generated by the decomposition and the degradation of animal manures, mainly bovine and cattle slurries, swine slurries (Zheng et al., 2019) and poultry excreta (Mohan Singh et al., 2020). They provide a median calorific value of  $5000 \pm 1000$  kcal/kg (Sahu et al., 2016) with methane ( $\text{CH}_4$ ) emissions 8 to 10 times higher than  $\text{CO}_2$  (Grant et al., 2015 ; Awasthi et al., 2019). A great amount of these manures are used as organic fertilizers. Thus, large quantities of their effluents are highly charged with ammonium ( $\text{NH}_4^+$ ), nitrite ( $\text{NO}_2^-$ ) and nitrate ( $\text{NO}_3^-$ ). Over the time, soil hold nutrients which reaches to natural water resources and creates environmental problems like eutrophication. Therefore, they must be treated before being discharged into nature. The removal of dissolved nitrogen from water is always an important subject of study. Removal can be achieved by physico-chemical (Dumont et al., 2019) or biological methods (Przytocka-Jusiak et al., 1984; Cai et al., 2013; Jia and Yuan, 2016; Amini, Amini Khoei and Erfanifar, 2019). However, physico-chemical methods (adsorption, chemical precipitation, reverse osmosis, ion exchange, air stripping, etc.) are not very effective when the water to be treated contains several forms of nitrogen simultaneously and at high concentrations (several hundred  $\text{mg N.L}^{-1}$ ) (Dumont et al., 2019). Moreover, certain biological methods, such as denitrification processes, are currently not recommended because of the production of large quantities of nitrous oxide ( $\text{N}_2\text{O}$ ), identified as one of the main greenhouse gases (Mezzari et al., 2013). Therefore, wastewater containing nutrients can be an alternative to high-cost nutrient media for the good growth of microalgae. This includes dairy wastewater (Kothari et al., 2013), municipal wastewater, poultry wastewaters (Ibekwe et al., 2017; Azam et al., 2019; Azam et al., 2020) and piggery wastewater (Zheng et al., 2019; Dumont et al., 2019; Azizi et al., 2020). Hence, the importance of taking into consideration the high concentrations of ammonium that can inhibit the growth of microalgae and consequently the need to optimize cultivation processes, in particular, using promising technologies such as Photobioreactors (Tam and Wong, 1996; Khan and Yoshida, 2008; Hady-Romdhane et al., 2012; Nwoba et al., 2016). Several species of microalgae, including *Chlorella vulgaris*, have been shown to be particularly tolerant to high ammonium concentrations (up to  $1000 \text{ mg N. L}^{-1}$ ) and could be an interesting opportunity for the treatment of these waters (Kwon et al., 2020) in addition to their important commercial value for bioenergy, cosmetics and livestock feeds production (Montalvo et al., 2020). Therefore, the present work has the goal of presenting (I) the main forms of microalgae valorisation the characteristics of livestock effluents, (II) the characteristics of livestock effluents, (III) the most studied technologies to different scales of recovery of carbon, nitrogen and phosphorus and other nutrients from livestock wastes, (IV) the main characteristics and parameters of the processes involved in these technologies, and finally, their (V) efficiency and feasibility.

## 1. Material and methods

In order to characterize the different livestock manure effluents, a data review from several studies and literatures has been collected in the Table 2 and Annexes I, II and III, with a focus on piggery, cattle and poultry livestock wastes.

The collected data has been processed using the SPSS 23.0 software, with Kruskal-Wallis nonparametric test, as it contains independent variables. Then, a One-way ANOVA test followed by a post-Hoc test of Tukey were applied.

## 2. Microalgae cultivation systems, biomass valorisation and by-products

Microalgae are ubiquitous and omnipresent microphytes, they are defined as aquatic unicellular microorganisms that can be both found in freshwater and marine systems, in many shapes and sizes, ranging from three to ten micrometres. They grow and reproduce naturally in lakes, rivers, lagoons, sea and oceans and are capable of photosynthesis. Due to their photosynthetic adsorption and absorption potential, microalgae and mainly Chlorophyceae (green microalgae), are worldwide cultivated, for several purposes and within different scales (Table 1). They were firstly grown in large-scale systems, for food chain supply as human super aliment, mainly *Spirulina*, in open ponds in Germany (Hamed, 2016) before the rise of the industrial cultivation techniques.

### 2.1 Microalgae cultivation systems:

There are several ways and scales of cultivating microalgae. However, there are generally two modes of culture; continuous or batch culture.

#### Continuous cultivation mode:

In this mode of cultivation, the growing medium is continuously renewed. The volume of the culture is kept constant by the application of an inlet and outlet flow rate or an overflow that ensures the removal of excess volume (Table 1, Hadj-Romdhane et al, 2012). This cultivation method is essentially used to maintain a constant algal production from the point of view of the produced biomass. That is, to always maintain production in exponential phase (Figure 1; Filali et al, 2011; Cerón-García et al, 2012; Wang et al, 2013; Ben Amor, 2015). According to Ben Amor (2015), the material balances on biomass between  $t$  and  $t+dt$  gives:

$$Q \cdot X \cdot dt + V dX = rV dt \quad (1)$$

With:  $Q$  = flow rate of liquid leaving the reactor ( $L h^{-1}$ ) assumed to be equal to the inlet flow rate  $V$  = crop volume (L),  $X$  = cell concentration ( $cell L^{-1}$  or  $g \text{ dry matter } L^{-1}$ ) and  $r$ : growth rate equal to  $\mu X$  with  $\mu$  specific growth rate ( $h^{-1}$ ). The growth rate  $r$  is defined as the change in the number of cells or weight of the biomass in a well-defined time and per unit of volume. Different models have been developed to estimate  $\mu$ . The best-known model is based on Monod's law in the case of growth limitation by light or carbon substrate. At steady state and in a continuous culture, the biomass rate is stable, so we have:

$$\mu = \frac{Q}{V} = D \quad (2)$$

With  $D$  the dilution factor (Ben Amor, 2015).

Table 1 : Summary of microalgal cultivation systems, purposes, and production scales

Microalgae strains	Biomass production	Country	Microalgae culture purposes	Treatment process : reactors	Harvesting technologie	References
<i>Spirulina</i>	3 (t dry weight /year)	China, India, USA, republic of Myanmar, Japan	Wastewater and liquid effluents treatments (digestates, agricultural and livestock effluents, industrial wastewater), livestock feed, Human nutrition, phycobiliproteins, cosmetics	Bioprocess : open ponds, PBR		Sialve and Steyer (2013)
<i>Chlorella</i>	2000 (t dry weight /year)	Taiwan, Germany, Japan	Metals and heavy metals treatment, aquaculture feed, cosmetics	Bioprocess (adsorption and assimilation by Microalgae): Open ponds, PBR	Precipitation, chelation, membrane filtration	
<i>Dunaliella</i>	1,2 (t dry weight /year)	Australia, USA, China	Gaseous effluents treatment (CO <sub>2</sub> fixation), Human nutrition, cosmetics-carotene	Physical process (CO <sub>2</sub> capture and storage)		
<i>Aphanizomenon</i>	500 (t dry weight /year)	USA	Bioenergy production; Biofuel, Ethanol, hydrogen, methane ...			
<i>Haematococcus</i>	300 (t dry weight /year)	USA, India	Aquaculture, Astaxanthine			
<i>Cryptocodinium</i>	240 t DHA	USA	Docosahexaenoic acid DHA(Omega-3)			
<i>Schizochytrium</i>	10 t DHA	USA	Docosahexaenoic acid DHA (Omega-3)			
<i>Scenedesmus obliquus</i>		USA	Biofuel production Carbon dioxide mitigation	Open ponds, PBR	Filtration, sedimentation, centrifugation, flocculation	Cai et al. (2013)
<i>Chlorella sp.</i>			Wastewater streams treatment (municipal, agricultural, and industrial)			
<i>Euglena gracilis</i>						
<i>Chlorella vulgaris</i>	Biomass production: 1200–1350 mgL <sup>-1</sup> Lipid productivity: up to 40mgL <sup>-1</sup> d <sup>-1</sup>		Bioenergy production	Flat-plate PBR (25L) with LED light Laboratory scale		Andersen, Jacobson, and Sexton (1991) Papapolymerou et al. (2018)
Microalgae strains	Biomass production	Country	Microalgae culture purposes	Treatment process : reactors	Harvesting technologies	References

<u><i>Scenedesmus obtusiusculus</i></u> BG-11	Biomass productivity : 0.90 gL <sup>-1</sup> d <sup>-1</sup>			Flat-plate PBRs (10 in series, 30L each) inclined 60 degrees Pilot scale.		Koller et al. (2018)
<u><i>Chlorella vulgaris</i></u>	Biomass productivity: 17.9mgL <sup>-1</sup> d <sup>-1</sup> Carbon content: 49.96%		Wastewater treatment	Air-lift Tubular PBR (7L) with 14-h light/10-h dark cycle and air bubbling Laboratory scale		Chaudhary, Dikshit, and Tong (2018)
<u><i>Chlorella sp. and Scenedesmus sp. consortium</i></u>	Biomass productivity: 60–61mgL <sup>-1</sup> d <sup>-1</sup> Lipid productivity: 5.4–21mgL <sup>-1</sup> d <sup>-1</sup>		Wastewater treatment: Anaerobically digested piggery effluent	Raceway pond (15 cm depth, 1500L). Cultivation in batch and semi continuous modes Pilot-scale		Raeisossadati et al. (2019)
<u><i>Chlorella vulgaris</i></u>	Biomass productivity: 0.57 g/L·d <sup>-1</sup>	Korea	Nutrients recovery from swine digestate wastes	Lab scale Flasks (500 mL)	Membrane filtration	Kwon et al. (2019)
<u><i>Dunaliella sp.</i></u>	Biomass productivity: 678 mg/L, β-carotene yield : 4.02 -7.26 mg/L	China	Digested poultry litter purification β-carotene production	Lab scale (500 mL) 15 days	Filtration, centrifugation	Han et al. (2019)

## Batch mode

In this culture mode, an inoculum of microalgae is added at time  $t$  to a culture medium at a given volume. The growth of the algal biomass will stop when there are no more nutrients to be consumed by the algae in the culture medium (Ben Amor, 2015).

According to Ben Amor (2015), this mode of culture contains five growth phases (Figure1):

- **Phase 1** "The latency phase" or phase of adaptation to the growing environment.
- **Phase 2** "The acceleration phase": the phase of the beginning of cell division.
- **Phase 3** "The exponential phase": The rate of cell multiplication is maximum. Environmental conditions are optimal for algal growth.
- **Phase 4** "The stationary phase": the concentration of microalgae remains constant. One of the essential elements is missing (light, nitrogen, phosphorus, carbon...); the growth rate decreases and just compensates for mortality.
- **Phase 5** "The decline phase": negative growth rate, the majority of the cells have exhausted their intracellular reserves.

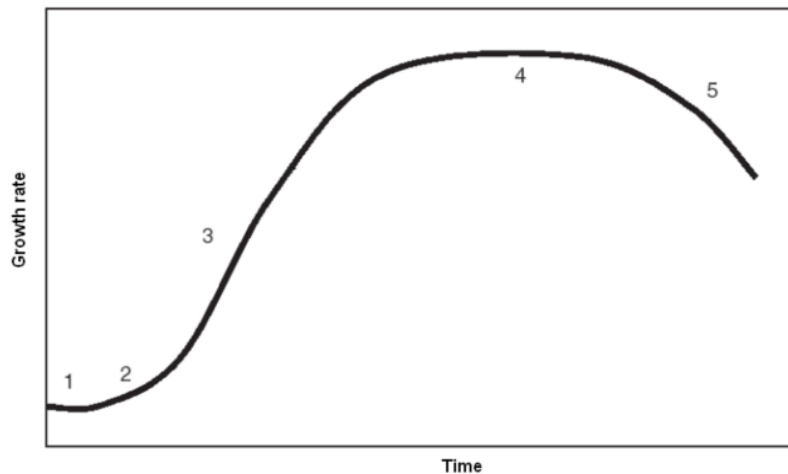


Figure 1 : Diagram of the kinetics of algae growth. 1: Latency phase; 2: Acceleration phase; 3: Exponential phase; 4: Stationary phase; 5: Decay phase (Richmond, 2007).

The material balance between times  $t$  and  $t+dt$ , in this mode of cultivation gives:

$$VdX = r V dt \quad (3)$$

We get:

$$\frac{dX}{dt} = \mu X \quad (4)$$

$\mu$  is only constant in the exponential phase, which is as follows:

$$\mu = \frac{\text{Ln} \left( \frac{X}{X_0} \right)}{t-t_0} \quad (5)$$

$X$  and  $X_0$ : cell concentrations at times  $t$  and  $t_0$  respectively. The generation or doubling time ( $td$ ) is defined as the time required for a number of cells to double their number or weight.

## Microalgae culture bioreactors

There are two systems of algae culture.

**Open-air systems or basins** (circular on the left or raceway at right (Figure 2)). This cultivation system is generally used for large-scale algae production, up to 2000 tonnes of dry weight/year (Table 1; El-Baz and Baky, 2018) under more or less controlled environmental conditions. (Ahsan et al, 2008; Ben Amor, 2015) and with almost no needed expensive flue gases, like  $\text{CO}_2$  supply.  $\text{O}_2$  is loaded via natural air mixing or via paddle wheels' outlets.

These cultivating systems are hence highly exposed to contamination such as grazers, bacteria, fungi, viruses, and other parasite microalgae (Bux and Chisti, 2016).



Figure 2 : Pictures of two types of algae tanks, circular tank on the left. raceway tanks on the right (Ben Amor, 2015).

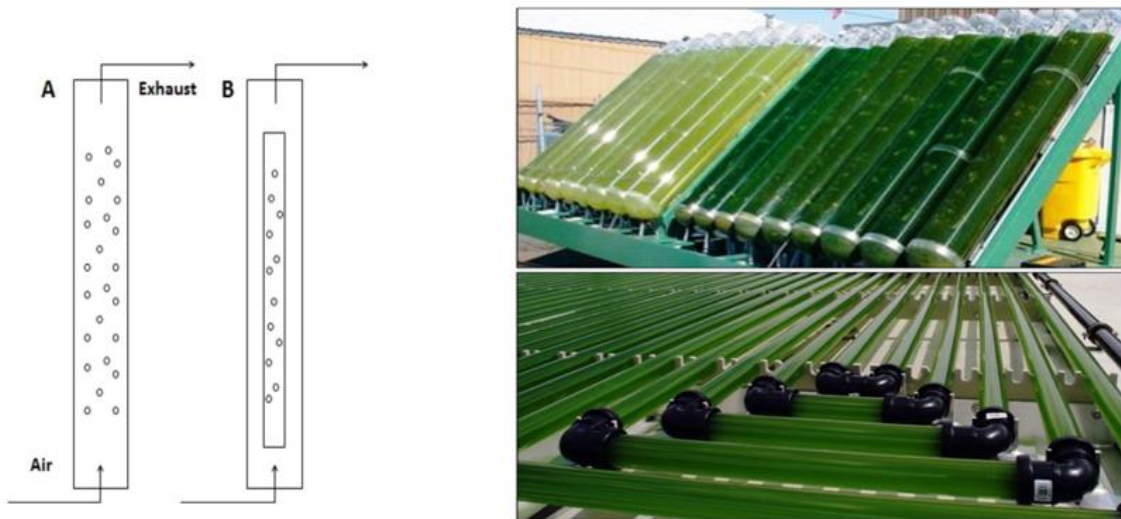


Figure 3 : The different types of photobioreactors (Ben Amor, 2015).

**Photobioreactors**, are closed, transparent containers to allow the penetration of light into the crop. Photobioreactors are designed to allow good light exposure and control of all culture parameters: pH, temperature, oxygen, agitation etc. (Hadj-Romdhane et al., 2012; Ben Amor, 2015), to reduce contamination risks and limit CO<sub>2</sub> losses (Moheimani et al., 2015) in addition to the increase in biomass productivity (Table 1) as they considered, generally, as continuous cultivation mode with stationary biomass growth; phase 5 in figure 1 doesn't exist. There are several configurations of photobioreactors; tubular reactors (Acien et al., 2017; El-Baz and Baky, 2018), coils, rigid panels, ring reactors (Richmond, 2007), Flat-plate PBRs (Acien et al., 2017; Płaczek, et al., 2017) plastic bags (Ben Amor, 2015) and bubble columns (Figure 3). Typically, the photobioreactors are closed loop, and the crop is constantly recycled. This optimizes the amount of water used and avoids evaporation losses compared to the pond culture system. The culture medium and carbon dioxide can be introduced by continuous or not, and at the end of the cycle, the algal biomass produced is recovered. (Richmond, 2007; Wang et al., 2012; Hadj-Romdhane et al., 2012; Ben Amor, 2015).



## 2.2 Microalgae biomass valorisation by-products and applications

Microalgae have a great commercial value. Indeed, they are seen as a considerable potential as a feedstock for different bioproducts (Figure 4; Borowitzka, 2013; Borowitzka and Moheimani, 2013; Klok *et al.*, 2014; Moheimani *et al.*, 2015; Raheem, *et al.*, 2018), from biofuels to precious biomolecules such as antioxidants, lipids (Feng *et al.*, 2011; Zhou *et al.*, 2011; Lam and Lee, 2012; Marjakangas *et al.*, 2015; Shen *et al.*, 2015; Zhou *et al.*, 2016, Jiang *et al.*, 2016; Lam *et al.*, 2017) and proteins. Moreover, they can be cultivated in different environmental conditions and systems (Table 2). Therefore, microalgae present as a promising solution to alleviate the ecological and environmental footprint of the increasing human population and its impacts on climate change, as they boost CO<sub>2</sub> sequestration, afford more food and livestock feed, can be a mean of renewable energy and wastewater treatment and recycling toward a sustainable bioeconomy. (Table 1, Table 3, Ferreira *et al.*, 2018 ; Rizwan *et al.*, 2018 ; Bussa *et al.*, 2019).

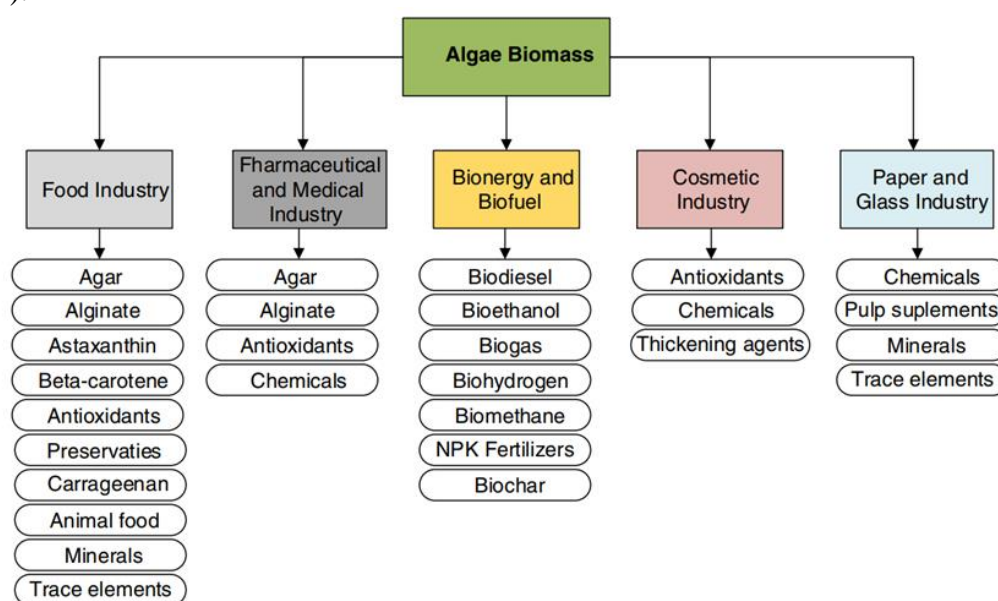


Figure 4 : Commercial application of algae biomass (Montalvo *et al.*, 2020)

### Bioenergy production

Among the various applications of algal biomass, its use in biodiesel, biomethane (Bishop and Rahman, 2017), bioethanol (Lee, *et al.*, 2015), biohydrogen (Khosravitar, 2019 ; Show *et al.*, 2019), bio-oil (Guo *et al.*, 2019), biogas (Toro, Perez, and Alzate, 2018), bioelectricity (Kumar *et al.*, 2019) and Biochar production (microalgae pyrolysis). The reorientation toward the use of biofuels as a renewable, sustainable, and clean bioenergy means (Harun and Daquan, 2011; Lakatos *et al.*, 2019), has gained increasing importance, since microalgae require much less land area, less time to mature (24–48 h), and yield about 30 times or higher volume in oil about 5% to 75% of dry biomass (Ohse *et al.*, 2015) than old fossil fuels and terrestrial oilseed crops (Kumar and Sharma, 2014).

### Chemical materials, cosmetics, and pharmaceutical industry

Among the many bioactive compounds known to be produced by microalgae are vitamins, lipids (Feng *et al.*, 2011; Zhou *et al.*, 2011; Lam and Lee, 2012; Marjakangas *et al.*, 2015; Shen *et al.*, 2015; Zhou *et al.*, 2016, Jiang *et al.*, 2016; Lam *et al.*, 2017), pigments like phycocyanine and  $\beta$ -carotene (Khanra *et al.*, 2018), and polysaccharides (Perin *et al.*, 2019). Some of those

chemicals, such as antioxidants, have demonstrated anticancer (Dewi et al., 2018), anti-infective, antioxidant (Agregán et al., 2018), immunostimulatory (Chen et al., 2019), anti-inflammatory (Lauritano et al., 2016), activities, and hypocholesterolaemia (Martínez-Bisbal et al., 2019) effect. These biological macromolecules are highly required in cosmetics, pharmaceutical and functional food, due to their virtue against oxidative and free radicals. (Tessmer Scaglioni et al., 2018). Their use in these industries is still growing (Joshi et al., 2018; Patel et al., 2019).

*Spirulina sp* and *Chlorella sp.* are considered among the super aliments with a high nutritional value. They are widely used as nutraceutical food supplement worldwide due to their contents in oligo minerals  $Mg^{2+}$ ,  $Fe^{2+}$  .... (Table 1 ; Ben Amor et al., 2015, Andrade et al., 2018 ; Khanra et al., 2018).

## **Agriculture**

### **Feed for livestock**

In Agriculture, microalgae are cultivated for their nutritional value, high content in proteins carbohydrates, lipids, vitamins, minerals, and bioactive compounds, such as carotenoids, as livestock feed and prebiotics for pigs (Yan et al., 2012 ; Kibria and Kim ,2019), chicken (Ribeiro et al., 2013 ; Evans et al., 2015 ; Kralik et al., 2020), rabbits (Kata et al., 2018 ; Abdel-Daim et al., 2019 ; Aladaileh et al., 2020) and ruminants (Lamminen et al., 2019) and in aquaculture (FAO, 2018) as fish (Eryalçin et al., 2015 ; El-Habashi et al., 2019) and shrimp (Adissin et al., 2020) diets. (Madeira et al., 2017; Table2). Among livestock animals, ruminants have the advantage of being capable to use the non-protein nitrogen and to digest the microalgae cell wall (Lum et al., 2013). The inclusion of 100 % microalgae meal in the dietary of ruminants (mainly cows) showed similar results of digestibility as a 100% soybean meal. Indeed, no change on milk production and animal growth has been detected (Lamminen, et al., 2019). Some changes in meat and milk composition has been evaluated, with an increase of some fatty acids' concentration such as docosahexaenoic acid DHA (Flaga et al., 2019), as well as on oxidative status and animal reproduction but with lesser extent (Mavrommatis et al., 2018; Tsiplakou et al., 2017; Tsiplakou et al., 2018) and animal reproduction (Senosy et al., 2017; Sinedino et al., 2017).

### **Soil fertilizers**

Cyanobacteria are reported as the well-studied as microalgal biofertilizer. Thanks to its specialized cells called heterocysts that contains the nitrogenase enzyme, Cyanobacteria act like a site for atmospheric nitrogen fixation (Karthikeyan et al., 2007; Babu et al., 2015). These diazotrophic cyanobacteria enhance the crops to the crops yield by improving the soil nitrogen and preventing its leaching (Nilsson et al., 2002; Renuka et al., 2016; Renuka et al., 2017; Renuka et al., 2018). Which results in the reduction of at least a 50% in the use of chemical fertilizers while maintaining the grain quality and yield (Pereira et al., 2009) for a variety of crops such as maize, wheat, cotton, and tomato (Osman et al., 2010; Prasanna et al., 2015; Dineshkumar et al., 2019). In addition to their use as bioenergy means, microalgae and biochar from microalgae contributes also to the maintenance of soil structure, physical properties (porosity, stable soil aggregates, water holding capacity, bulk density, water infiltration, and hydraulic conductivity) and quality and optimal levels of soil organic matter and nutrients by improving the microbial activity through facilitating the interaction and the growth of beneficial microbes (Leloup et al., 2013; Yan-Gui, et al., 2013; Li et al., 2018; Ronga

et al., 2019) Moreover, some microalgae species are considered as a sustainable biopesticide sources (Righini and Roberti, 2019).

### **Wastewater treatment and nutrients recovery**

Some algal strains are used for nutrients recovery mainly nitrogen phosphates, minerals from wastewater and agricultural effluents (Franchino et al., 2013; Franchino et al., 2016; Ding et al., 2016; Udaiyappan et al., 2017; Hazman et al., 2018; Khalid et al., 2019; Udaiyappan et al., 2020, Table 1). This microalgae application pathway will be further detailed and developed in the following parts of this article.

### **3.Characteristics of the livestock manure effluents; swine, cattle and poultry wastes:**

Nitrogen levels in pig manure are higher than in poultry and cattle manure (Table 2). Depending on the physiological stage and dilution level, they range from 63.7 to 6129 mg TN /L of for swine manure, from 63 to 5580 mg TN /L for cattle manure. The contents of poultry manure vary between 27.32 and 3050±212.13 mg TN /L (Table 2). Ammoniacal nitrogen ( $\text{NH}_4^+$ ) represents in average 70% of total nitrogen (TN) in pig manure and 40% in cattle manure, following rapid hydrolysis of urea. (Itavi, 2003 ; Benoît, 2014). According to Christensen et al. (2009) et Benoît et al. (2014),  $\text{NH}_4^+$  is 70% in the dissolved phase, the rest is associated with proteins or mineral precipitates. For poultry manure, the  $\text{NH}_4^+$  content changes over time due to the transformation of uric acid, an organic form which represents the main component of poultry manure with up to 65 % of the TN content (Koerkamp et al., 1998 in Benoît et al., 2014). P contents are variable but follow similar trends (Table 2; Figure 6). They are generally lower for bovine effluents than for porcine effluents. which are even lower than those from poultry manure (Awasthi et al., 2019) that could reach up to 4547 mg TP/L. Potassium K contents are about up to 2100 mg/L for chicken effluents (centrifuged digestate supernatant ) against up to 3418 mg/L for swine slurry and up to 3338 mg/L for Dairy cattle effluents (Table 2).  $\text{Mg}^{2+}$  and  $\text{Ca}^{2+}$  contents also have a similar tendency as the other nutrients; they vary identically regardless of the effluent type or the livestock effluent origin factors. Indeed, the Kruskal-Wallis test for non-parametric variables showed no significant differences between nutrients variability depending on the livestock effluent origin and the effluent form.  $\text{NH}_3$  and  $\text{N}_t$  and the pH, are the only parameters that showed a significant difference with p-value < 0.05 for 95 % of certainty interval (Figure 6). The One-way ANOVA and the post Hoc Tukey tests confirmed this tendency for the pH and the other parameters (p-value=0 < 0.05). Swine slurries present generally an alkaline pH with an average of  $8 \pm 2$ . Cattle wastes are generally neutral (pH= 7) with an alkaline tendency. Poultry excreta are mostly acid in average (pH=4), but they can be neutral depending on the effluent type and if it was pre-treated or not. For example, most of the poultry wastes reported in Table 1, were treated by adding acidifying substrates such as NaOCl, especially in the lab scale studies. Ammonium and total nitrogen are very variable depending on the effluent type and provenance, but they converge both to a maximum of 5g. L<sup>-1</sup>. The Kruskal-Wallis and the One –way ANOVA tests showed also a significant difference for the N: P ratio for  $\alpha = 0.05$  and 95 % of CI., for the three type of livestock farming types; porcine,

Tableau 2 : Characteristics of livestock manure: literature review

Livestock farming type /Effluent type	NH <sub>4</sub> <sup>+</sup> [mg /L]	NH <sub>3</sub> [mg /L]	N <sub>t</sub> [mg /L]	PO <sub>4</sub> <sup>3-</sup> [mg /L]	Pt [mg /L]	pH	SO <sub>4</sub> <sup>2-</sup> [mg/L]	C [g/Kg. DM]	TS [mg/L]	VS [mg/L]	TOC [mg/L]	IC [mg/L]	BOD [mg/L]	COD [mg/L]	Country	References
<b>Swine</b>																
Slurry	-	-	2161.5±1487.05 (a)	648.5±478.71	-	-	-	-	-	-	-	-	-	-	USA	Cai <i>et al.</i> (2012)
Slurry	1400	-	-	600	-	5.5	-	-	-	-	19500	-	-	89000	-	Khoshnevisan <i>et al.</i> (2021)
Slurry (b)	-	-	63.7	-	31.15±11.53	-	-	260	-	-	-	-	-	-	France	Sharpley <i>et al.</i> (2004) ; Fuchs <i>et al.</i> (2014)
Slurry	-	1434.3	-	18.69	-	8.95	-	-	-	-	-	-	-	-	Taiwan, China	Wang <i>et al.</i> (2015)
Slurry	-	-	1320	-	1210	-	-	-	-	-	-	-	-	-	Brazil	Bertagnoli <i>et al.</i> (2020)
Slurry	3418	-	6129(a)	-	2121	7.5	-	-	129643	100179	-	-	36536	99000	USA	ASAE, (2003) ; Ma <i>et al.</i> (2018)
Slurry	4900	-	4370	-	1470	6	-	-	65700	55000	-	-	25000	82000	Japan	Li and Kobayashi(2010) ; Ma <i>et al.</i> (2018)
Slurry	289	-	-	-	24	-	-	-	-	-	-	-	-	745	-	Mingzi <i>et al.</i> (2016) ; Lee <i>et al.</i> (2021)
Slurry	598.04 ± 13.96	-	798.36 ± 22.54	42.95 ± 0.31	42.95 ± 0.31	7.60 ± 0.15	-	-	-	-	-	-	-	2383.46 ± 65.10	China	Cao <i>et al.</i> (2018)
Slurry	1761±41	-	1.1± 1.0*	161± 2	-	-	-	-	4188±18	2112±88	-	-	-	3750±64	Spain	Molinuevo-Salces <i>et al.</i> (2016)
Slurry (c)	228.105±202.03	-	251.5±339.31/0.49*/0.12**	2.28±3.03/	5.99±8.27	-	-	-	-	-	-	-	-	-	-	Monfet and Unc (2016)
Raw slurry	-	850-1150	-	-	10.5-34	-	-	-	-	-	-	-	-	6870-8420	Korea	Lee <i>et al.</i> (2021)
Raw slurry	-	1388	980	158	-	-	-	-	7000	4500	-	-	-	11	China	Wang <i>et al.</i> (2012)
Raw slurry	-	-	547.78 ± 44.53	81.72 ± 14.25	81.72 ± 14.25	8.14	-	-	-	-	-	-	-	824.53 ± 31.58	China	Wen <i>et al.</i> (2017)
Raw slurry (d)	-	3.95 ± 0.03	5.9 ± 0.3/0.04 ± 0.01*	0.47 ± 0.01	-	7.63	-	-	59.6 ± 0.51	-	-	-	-	75.4 ± 0.1	Korea	Kwon <i>et al.</i> (2019)
Autoclaved slurry	-	-	313.25 ± 19.35	-	56.18 ± 12.44	8.36	-	-	-	-	-	-	-	796.21 ± 24.67	China	Wen <i>et al.</i> (2017)
Centrifuged and sterilized slurry	219.5 ± 11.3	-	277.6 ± 6.1	30.2 ± 0.0 (b)	30.2 ± 0.0	6.3 ± 0.0	-	-	47500 ± 0.8	5300 ± 0.1	152.4 ± 8.4	86.1 ± 5.0	-	485.2 ± 3.5	China	Zheng <i>et al.</i> (2019)
Liquide manure	-	-	1550±1060.66(a)	185±190.92 (b)	185±190.92	-	-	-	-	-	-	-	-	-	Africa	Nwoba <i>et al.</i> ,2020
Liquide manure	532	-	-	72	145	8±0.71	-	-	-	-	-	-	-	-	-	Suzuki <i>et al.</i> (2007) ; Kataki <i>et al.</i> (2016)
Liquide manure	234	-	-	42	-	9	-	-	-	-	-	-	-	-	-	Perera <i>et al.</i> (2007) ; Kataki <i>et al.</i> (2016)
Liquide manure	831.5±14.71	-	1039.5±113.84	43±18.38	163±82.73	9	-	-	-	-	-	-	-	-	-	Ryu et Lee, (2010) ; Kataki <i>et al.</i> (2016)
Liquide manure	985	-	1212	161	182	8.25±0.35	-	-	-	-	-	-	-	-	-	Huang <i>et al.</i> (2011a, b) ; Kataki <i>et al.</i> (2016)
Liquide manure	1219.5±292.04	-	1219.5±292.04	97±59.40	139±70.71	10	-	-	-	-	-	-	-	-	-	Zhang <i>et al.</i> (2012) ; Kataki <i>et al.</i> (2017)
Digestate	233	-	348(a)/5.5*	101.4	-	7.7	-	-	-	-	-	-	-	332	Taiwan, China	Marjakangas <i>et al.</i> (2015)
Digestate	-	-	3304 ± 195 (a)/ <2.5 *	-	193 ± 20	7.6	<40	-	7.1 ± 0.2	4.4 ± 0.04	-	-	-	12.152	Australia	Kumar <i>et al.</i> (2010)
Digestate	-	1093	1135	-	24.5	-	373.2	-	-	-	-	-	-	2000-4000	China	Cheng <i>et al.</i> (2015)
Digestate	255.4 ± 4.9	-	463.0 ± 6.7	-	113.3 ± 7.0	7.0 ± 0.03	-	-	-	-	256.23 ± 20.78 (g/L)	-	-	18.6 ± 0.2	USA	Deng <i>et al.</i> (2017)
Digestate	240-690	-	-	-	33-43	9	-	-	-	-	97-220	-	-	69-97	Australia	Ayre <i>et al.</i> (2017)

(a): Total Kjeldahl nitrogen (TKN); (b): in g/Kg; (c): mmol/L; (d): g/L; \*: NO<sub>3</sub>-N; \*\*: NO<sub>2</sub>-N; NH<sub>4</sub><sup>+</sup>: ammonium; TN: total nitrogen; TP: total phosphorus; TOC: total organic carbon; IC: inorganic carbon; BOD: biological oxygen demand; COD: chemical oxygen demand; TS: total solids; VS: volatile solids ..... (Table to be continued in the next page)

Digestate	-	268.93 ± 8.38	331.55 ± 3.18 / 32.17 ± 1.35 *	22.95 ± 0.26	27.25 ± 0.2	7.97 ± 0.02	-	-	-	-	-	-	-	346.15 ± 3.61	China	Zeng <i>et al.</i> (2020)
Digestate (c)	140 ± 133.85	-	122.96/7.93	8.94	11.045	-	-	-	-	-	-	-	-	-	-	Monfet and Unc (2016)
Digestate (d)	-	3.7 ± 0.06	4.0 ± 0.12/0.12 ± 0.01*	0.3 ± 0.01	-	8.49	-	-	53.4 ± 4.72	-	-	-	-	16.7 ± 0.15	Korea	Kwon <i>et al.</i> (2019)
<b>Livestock farming type /Effluent type</b>	<b>NH<sub>4</sub><sup>+</sup> [mg /L]</b>	<b>NH<sub>3</sub> [mg /L]</b>	<b>N<sub>i</sub> [mg /L]</b>	<b>PO<sub>4</sub><sup>3-</sup> [mg /L]</b>	<b>Pt [mg /L]</b>	<b>pH</b>	<b>SO<sub>4</sub><sup>2-</sup> [mg/L]</b>	<b>C [g/Kg MS]</b>	<b>TS [mg/L]</b>	<b>VS [mg/L]</b>	<b>TOC [mg/L]</b>	<b>IC [mg/L]</b>	<b>BOD [mg/L]</b>	<b>COD [mg/L]</b>	<b>Country</b>	<b>References</b>
<b>Cattle</b>																
Slurry	-	-	63(a)	-	14	-	-	-	-	-	-	-	-	-	Africa	Nwoba <i>et al.</i> (2020)
Slurry	1300	-	-	-	-	7	-	-	-	-	-	-	-	-	-	Khoshnevisan <i>et al.</i> (2021)
Centrifuged slurry (supernatant)	643.88 ± 2.53	-	89.63 ± 0.42*/1.14 ± 0.02**	-	79.37 ± 0.32	7.72 ± 0.01	-	-	-	-	-	-	-	9972 ± 36	China	Lu <i>et al.</i> (2018)
Beef slurry	-	-	2114±2900.5	-	604.5	-	-	-	-	-	-	-	-	-	USA	Cai <i>et al.</i> (2011)
Dairy cattle slurry	-	-	185(a)	-	30	-	-	-	-	-	-	-	-	-	Africa	Nwoba <i>et al.</i> ,2020
Dairy cattle slurry ***	1200	-	1950	395	-	7	-	-	47500	37500	-	-	6500	64000	Japan	Li and Kobayashi (2010); Ma <i>et al.</i> (2018)
Dairy cattle slurry (c)	65.36 ± 87.58	-	121.5±166.71	1.56	5.215	-	-	-	-	-	-	-	-	-	-	Monfet and Unc (2016)
Dairy cattle raw slurry	-	-	1200	-	500	-	300	-	-	-	-	-	-	-	USA	Dougherty <i>et al.</i> (2017)
Dairy cattle raw slurry	-	-	300	-	200	-	100	-	-	-	-	-	-	-	USA	Dougherty <i>et al.</i> (2017)
Dairy cattle raw slurry	-	-	600	-	100	-	100	-	-	-	-	-	-	-	USA	Dougherty <i>et al.</i> (2017)
Dairy cattle effluents	-	-	-	-	296± 29.70	8.5	-	-	-	-	-	-	-	-	-	Zhao <i>et al.</i> (2010) ; Kataki <i>et al.</i> (2016)
Dairy cattle effluents	387± 186.68	-	-	-	275± 247.49	8.85± 0.49	-	-	-	-	-	-	-	-	USA	Uludag-Demirer <i>et al.</i> (2005) ; Kataki <i>et al.</i> (2016)
Dairy cattle effluents	1405	-	2845	19	460	7.2	-	-	-	-	-	-	-	-	-	Shen <i>et al.</i> (2011) ; Kataki <i>et al.</i> (2016)
Dairy cattle effluents	909	-	5180 (a)	1082	1082	7	-	-	13814	115116	-	-	18419	126628	USA	ASAE (2003) ; Ma <i>et al.</i> (2018)
Dairy cattle effluents	2500	-	5530	1290	1291	7	-	-	136700	115000	-	-	24000	13000	Japan	Li and Kobayashi (2010); Ma <i>et al.</i> (2018)
Dairy cattle degistate (c)	82.715 ± 108.49	-	130.035±165.22	0.32	4.265	-	-	-	-	-	-	-	-	-	-	Monfet and Unc (2016)
Urine	7732	-	-	-	305	9	-	-	-	-	-	-	-	-	-	Prabhu and Mutnuri (2014) ; Kataki <i>et al.</i> (2016)

(a): Total Kjeldahl nitrogen (TKN); (b): in g/Kg; (c): mmol/L; (d): g/L; \*: NO<sub>3</sub>--N; \*\*: NO<sub>2</sub>--N; NH<sub>4</sub><sup>+</sup>: ammonium; TN: total nitrogen; TP: total phosphorus; TOC: total organic carbon; IC: inorganic carbon; BOD: biological oxygen demand; COD: chemical oxygen demand; TS: total solids; VS: volatile solids ..... (Table to be continued in the next page)

Livestock farming type /Effluent type	NH <sub>4</sub> <sup>+</sup> [mg /L]	NH <sub>3</sub> [mg /L]	N <sub>t</sub> [mg /L]	PO <sub>4</sub> <sup>3-</sup> [mg /L]	P <sub>t</sub> [mg /L]	pH	SO <sub>4</sub> <sup>2-</sup> [mg/L]	C [g/Kg MS]	TS [mg/L]	VS [mg/L]	TOC [mg/L]	IC [mg/L]	BOD [mg/L]	COD [mg/L]	Country	References
<b>Poultry</b>																
Manure (b)	-	-	38.5	14.35±1.63	-	-	-	304	-	-	-	-	-	-	-	Slaton <i>et al.</i> (2013) ; Fuchs <i>et al.</i> (2014)
Manure (b)	-	-	62.8	24	-	-	-	191	-	-	-	-	-	-	-	Guivarch and Morel (2001); Fuchs <i>et al.</i> (2014)
Manure (b)	-	-	50.2	27.8±11.74	-	-	-	364	-	-	-	-	-	-	-	Sharpley <i>et al.</i> ,2004 ; Fuchs <i>et al.</i> ,2014
Manure (b)	-	-	27.32	-	43.12	-	-	-	-	-	-	-	-	-	Brazil	Bertagnoli <i>et al.</i> (2020)
Liquide effluents	-	-	800(a)	-	50	-	-	-	-	-	-	-	-	-	Africa	Nwoba <i>et al.</i> (2020)
Slurry	-	-	1313.5±723.37	-	50-446	-	-	-	-	-	-	-	-	-	USA	Cai <i>et al.</i> (2012) ; Nwoba <i>et al.</i> (2020)
Slurry	1318	-	1580	-	371	9	-	-	-	-	-	-	-	-	-	Yetilmezsoy <i>et Sapci-Zengin</i> (2009) ; Kataki <i>et al.</i> (2016)
Slurry	3183	-	12731(a)	-	4547	6.9	-	-	242500	181.875	-	-	50016	166719	USA	ASAE (2003); Ma <i>et al.</i> (2018)
Treated slurry (acidified and filtered)	546.7 ± 4.6	-	46.1 ± 1.6 *	943.4 ± 24.9	-	1.0- 7.0	342.0 ± 8.2	-	-	-	-	-	-	2532.0 ± 10.1	USA	Shashvatt <i>et al.</i> (2018)
Treated slurry (acidified and filtered)	423.8 ± 6.7	-	-	1462.4 ± 55.3	-	1.0- 7.0	969.9 ± 49.6	-	-	-	-	-	-	3110.6 ± 14.0	USA	Shashvatt <i>et al.</i> (2018)
Treated slurry (acidified and filtered)	275.2 ± 1.9	-	0.2 ± 0.4*	1540.5 ± 62.3	-	1.0- 7.0	1000.3 ± 117.6	-	-	-	-	-	-	3309.3 ± 12.9	USA	Shashvatt <i>et al.</i> (2018)
Digestate	-	-	1480± 141.42	-	376± 8.49	-	-	-	-	-	-	-	-	-	USA	Cai <i>et al.</i> (2012)
Litter degistate	2500± 707.11	-	3050± 212.13	-	22.5± 3.54	9.5± 0.71	-	-	-	-	750± 353.55	2475± 176.78	-	650± 353.55	China	Han <i>et al.</i> (2019)
Digestate (b)	33.21- 308.2	-	144.39-254.64 / 0.40 *	2.68	3.10- 9.13	-	-	-	-	-	-	-	-	-	-	Monfet and Unc (2016)
Centrifuged degistate (supernatant)	1143-1787	-	1570-2473/ 0.55 10.7**	-	154-214	-	111-155	891- 876 (mg/L)	-	-	-	-	-	-	USA	Singh <i>et al.</i> (2011)

(a): Total Kjeldahl nitrogen (TKN); (b): in g/Kg; (c): mmol/L; (d): g/L; \*: NO<sub>3</sub><sup>-</sup>-N; \*\*: NO<sub>2</sub><sup>-</sup>-N; NH<sub>4</sub><sup>+</sup>: ammonium; TN: total nitrogen; TP: total phosphorus; TOC: total organic carbon; IC: inorganic carbon; BOD: biological oxygen demand; COD: chemical oxygen demand; TS: total solids; VS: volatile solids

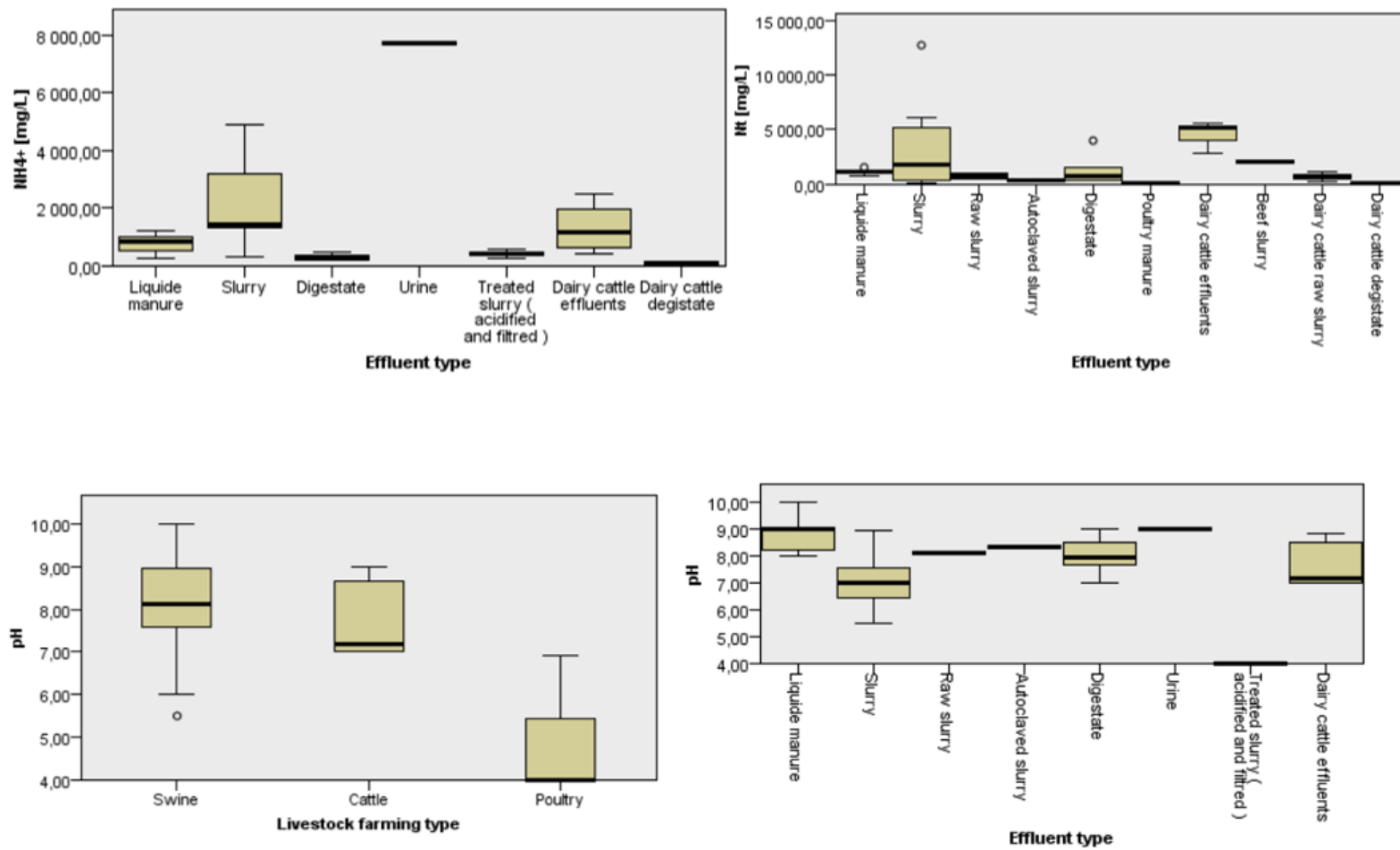


Figure 5 : Barplots of parameters with significant differences with a non-parametric Kruskal-WALLIS test analysis for alpha= 0.05 and 95 % of CI. All the other parameters have no significant differences for both the livestock farming type and effluent type factors.

bovine and poultry. This ratio varies between  $5.4 \pm 3.39$  (slurry) and  $14.5 \pm 3.54$  (liquid manure),  $3.25 \pm 1.77$  (Beef slurry) and 10 (slurry), and, between  $3.95 \pm 0.49$  (degistate) and 32 (liquid effluents), respectively for swine, cattle and poultry livestock wastes (Table 2).

This ratio is depending mainly on the livestock farming type, which is depending on the animals' species and feed. Indeed, as it was previously discussed, the poultry wastes are the richest in the nitrogenous and the phosphorous components. Moreover, the changing of the nitrogenous form due to the wastes form (solid manure or liquid slurries ...), generates differences in the N: P ratios.

A high N/P ratio (32) indicates a greater amount in nitrogen (800 mg/L of TN) than in phosphorus (50 mg/L of TP) within the composition of the slurry, or the livestock waste in general (Table 2). Such an elevated N: P ratios are generally observed in liquid wastes (N/P of the liquid poultry effluents is about 32 as it is shown in the Table 1), and this is due to the nature of the phosphorus components which are mainly under solid particulate form, while the nitrogenous components are mainly soluble in the liquid fraction of the effluent (mainly under the liquid and the gaseous forms).

Besides phosphate and nitrogen compounds, livestock wastes contain carbon, solid particulates as total solids (TS) and volatile solids (VS). These components can be present under the organic or the inorganic forms. The inorganic carbon (IC) is the bioavailable form for the algae strains, it's more present in the poultry litter digestates about  $2475 \pm 176.78$  mg/L, and about  $86.1 \pm 5.0$  mg/L for the centrifuged and sterilized swine slurry (Table 2). C indicates the organic carbon amount in the livestock wastes. It represents 260 g/Kg.DM in the swine slurry up to 891mg/L in the supernatant of the centrifuged poultry degistate.

COD (chemical oxygen demand) and BOD (biological oxygen demand) represent respectively the amount of dioxide required for the degradation of the organic matters in general (COD) and by the microorganisms (biodegradation; BOD). They indicate the amount of organic matters present in the livestock wastes. BOD is up to 36536 mg/L of swine slurry, up to 24000 of dairy cattle effluents and up to 50016 mg/L poultry slurry. Concerning the COD, it fluctuates between 12.152 and 99000 mg/L for the swine degistate and the swine slurry, respectively. They are between  $9972 \pm 36$  and 126628 mg/L in the supernatant of centrifuged cattle slurry and Dairy cattle effluents, respectively. Poultry litter degistate and poultry slurry contain respectively  $650 \pm 353.55$  mg COD /L and 166719 mg COD /L. The total organic carbon (TOC) are correlated to the COD amounts (Table 2). They both depend on the carried out treatment processes and the pH of the livestock wastes. Total solids and volatile solids represent respectively the amount of the total suspended solid particulates and the volatile (soluble) solid particulates. VS, TS, COD and BOD are very correlated to the suspended solid particulates and they highly influence the turbidity of the livestock effluents. A great value of these parameters indicates a great value of turbidity (NWOBA et al.,2020).

The inorganic soluble compounds that have been shown in Annexes I, II and III, are mainly sodium  $\text{Na}^+$ , calcium  $\text{Ca}^{2+}$ , potassium  $\text{K}^+$ , magnesium  $\text{Mg}^{2+}$ , chloride  $\text{Cl}^-$ , sulphate  $\text{SO}_4^{2-}$  (Table 1), iron  $\text{Fe}^{2+}$ , copper  $\text{Cu}^{2+}$ , zinc  $\text{Zn}^{2+}$  and manganese  $\text{Mn}^{2+}$ . These oligo nutrients are present in small amounts in livestock manures. They are considered as essential elements for plants growth, for animals feed and human alimentation. These chemical constituents (macro and micronutrients) of slurries could be extracted and valorised through microalgae cultivation (Ben Amor.,2015; Nwoba et al., 2020).

Wastewater from livestock facilities can also contain pathogenic microbes and toxins (Nwoba et al.,2020). Therefore, they must be well treated and managed in order to prevent environment contamination.



## 4. Wastewater and livestock manure effluents treatment technologies

### Manures and slurries: storage, treatment techniques and management policies

Agricultural farms, especially hog farms, are considered to be sources of odour emissions due to high ammonia levels in particular. These emissions are generated at the level of the barn at about 50%, against 25% of the emissions coming only from the spreading of manure, in liquid (slurry) or solid form (manure) (Guinand, 2003). However, the spreading and storage of dejecta are considered as a secondary source of nuisance in time, as they are managed by the regulatory obligations of the spreading schedule (ADEME,2012; ADEME,2018). As a result, pig farming is under increasing pressure from public opinion regarding its impact on the environment (ADEME,2012). Indeed, the number of farmer-neighbour conflicts has increased, and has led to a decrease or even a refusal of operating permits for installations or extensions of pig breeding buildings (Guinand,2008; Dumont *et al.*,2019). Due to the pollution impacts of the manure generated in livestock waste streams, in farms, they have to be treated before discharging into the environment (Awasthi *et al.*, 2019). Therefore, many special and strict regulations and policies have been established worldwide to manage livestock manures in breeding farms and regulate their valorisation as biomethane and biofertilization sources mainly through anaerobic digestion (Figure 6; Table 4; Awasthi *et al.*, 2019; Yao *et al.*, 2020). In Europe, several strict regulations, including “Nitrate Act (91/676/EC)”, “Drinking Water Quality Directive (80/778/EEC)”, “Water Framework Directive (2000)”, “Common Agriculture Policy (1992)” and “Good Agricultural Practices” are imposed to prevent manure field application (Yan *et al.*,2013; Awasthi *et al.*, 2019) with a fixed maximum of 170 Kg-N/ha from animal manure which is only applied in France (Yan *et al.*,2013; Awasthi *et al.*, 2019).

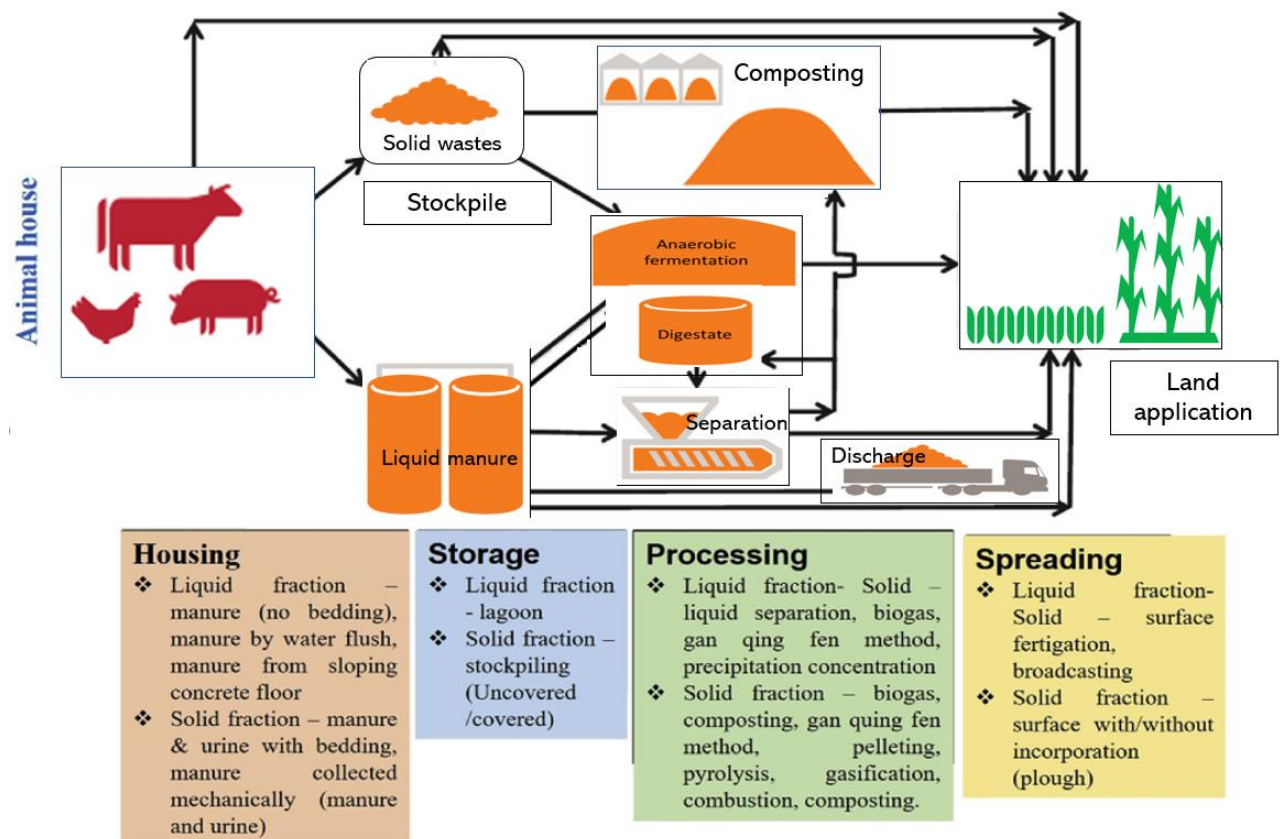


Figure 6 : Graphic outline of operated and critical stages for various livestock manure through combined crop-animal schemes in China. (adapted from Leenstra *et al.*,2019 and Awasthi *et al.*, 2019)

Denmark and Germany are promoting the of co-digestion concept via the installation of large centralized co-digestion manure-based plants for biogas production (Burg *et al.*, 2018; Fuchs *et al.*, 2018; Awasthi *et al.*, 2019). About below 10% of the manures are treated anaerobically in Denmark, this amount is supposed to reach up to 50% manures converted into biogas by 2050 (Hez *et al.*, 2016; Awasthi *et al.*, 2019). In China, the policies are promoting biogas power generation and composting technologies (Table 3; table 4; Yao *et al.*, 2020) by the “Recycling of Livestock Manure” in agriculture to substitute chemical fertilizers by 60–75% (Sun *et al.*, 2012; Adamović *et al.*, 2018; Awasthi *et al.*, 2019). In Sub-Saharan African countries, livestock housing shall be roofed, their floors must be waterproof and the manure storage systems under covered (Figure 7; Zahedi *et al.*, 2018; Awasthi *et al.*, 2019).

### **Physical and physico-chemical treatments, applied to digestates, and livestock manure.**

Based on their variable forms (slurry, excreta, solid manure) and consequently their variable composition (organic matter, minerals, soluble nutrients...) (Table 2), livestock wastes should be treated by several and different processes (Figure 7; Table 3, Table 4). These technologies involve various methods with variable parameters, mainly physical like solid-liquid separation (Table 4), that eliminates dust, solid organic and inorganic particulates ..etc. and hence, reduce the turbidity of the wastes through simple gravitational decantation (Table 4). The physicochemical and chemical approaches like air stripping (Table 3, Table 4), membrane filtration (Table 3; Elyssia Mayhead *et al.*, 2018; Zeng *et al.*, 2020), and struvite formation (Table 3; Szogi and Vanotti, 2009; Uludag-Demirer *et al.*, 2005; Kataki *et al.*, 2016), involve the addition or the passage of the effluent through a chemical medium or substratum to extract the demanded molecules. While air stripping technique allows the removal of up to 90% of total nitrogen and 93% of  $\text{NH}_4^+$ , membrane filtration enables the recovery of up to 94% of  $\text{NH}_3$ , less than 93 % of  $\text{NH}_4^+$  and less than 91 % of phosphorus (Table 3) by reverse osmosis (Lu *et al.*, 2014; Dumont *et al.*, 2019; Montalvo *et al.*, 2020). Via the struvite formation technique, up to 99 % of  $\text{PO}_4^{3-}$  and 80 % of  $\text{NH}_4^+$  could be reduced from the  $\text{NH}_4^+$  wastewater (Table 3). However the effectiveness of this approach is widely depending on the pH (alkaline) and the P: Mg: N ratio, that are affected by the manure composition itself (Kataki *et al.*, 2016; Shashvatt *et al.*, 2018; Bacelo *et al.*, 2020; Mazzini *et al.*, 2020; Montalvo *et al.*, 2020; Zeng *et al.*, 2020). In France, the recovery of nitrogen and phosphorus from liquid livestock effluents (e.g. slurry) and digestates is generally processed under pilot scale trials, in the form of ammonium sulphate, while phosphorus is recovered in the form of struvite (ammonium magnesium phosphate) or apatite (calcium phosphate) (Houot *et al.*, 2014).

Moreover, biochar floating biocovers are reported by Dougherty *et al.*, (2017) as a promising passive facility of nutrients recovery, odour and gas emissions reduction from livestock (mainly cattle) manure lagoons.

Experiments reported in the literature proved that technologies used for nutrient recovery from livestock waste, mainly swine, are mostly applied to the effluents previously treated by anaerobic digestion as a primary treatment. Greater efforts and assistance are required for the implementation of these technologies in large scales, because of the need of pH and chemicals concentrations adjustment and regular maintenance and servicing which are very costly. (Uludag-Demirer *et al.*, 2005; Montalvo *et al.*, 2020)005 ; Kataki *et al.*, 2016 ; Cao *et al.*, 2018 ; Ma *et al.*, 2018 ; Mazzini *et al.*, 2020 ; Montalvo *et al.*, 2020).

Tableau 3 : Nutrients recovery (carbon (C), nitrogen (N), phosphorus (P) technologies from livestock manure digestate; reproduced from Montalvo et al. (2020).

Technology	Nutrients recovery	Operating conditions	Nutrients removal efficiency	Reference
Algae biomass	C-conversion, P in algae biomass	-Algae Strain: <i>Chlorella sp.</i> -Digested Swine Manure -Semi-Continuous -Operation at Bench-Scale -Optimal Operational Conduction: Hydraulic -Retention Time (Hrt) = 2.26 Days -Dilution Rate = 8.0 Times -Two Layer PBR Volume = 17 L - Cultivation Time = 17 Days	751 mg COD L <sup>-1</sup> d <sup>-1</sup> 20 mg PO <sub>4</sub> -P L <sup>-1</sup> d <sup>-1</sup> 38 mg TN L <sup>-1</sup> 60 mg NH <sub>3</sub> -N L <sup>-1</sup>	Hu et al. (2013)
	C-conversion, P in algae biomass	-Algae strain: <i>Chlorella sp.</i> - Batch - mode = 1 month Continuous - mode = 1-month Pilot scale -Multilayer PBR -Volume = 7500 L -Cultivation time = 2 months	-Batch mode: 73% NH <sub>3</sub> -N removal 23% P removal -Nitrogen (TN) removal: 17% -PO <sub>4</sub> -P removal: 22% -Chemical oxygen demand (COD) removal -Continuous mode :72% NH <sub>3</sub> - N removal	Min et al. (2014)
Air stripping	N	-Laboratory scale -pH 9.4-11.9 -Temperature = 6-25 ° -Air flow rate = 1.15-90 L min <sup>-1</sup>	90% N removal	Darestani et al. (2017)
	N	-Bench scale -Continuous operation -Temperature = 303-343 K -Air flow = 333-2000 L min <sup>-1</sup> -Air/liquid -ratio = 1875	93% NH <sub>4</sub> <sup>+</sup> removal 88% TN removal	Lim et al. (2015)
Membrane	N	Liquid-liquid membrane stripping solutions: H <sub>3</sub> PO <sub>4</sub> , HNO <sub>3</sub> and H <sub>3</sub> PO <sub>4</sub> /HNO <sub>3</sub> ratios -Active membrane area: 1.4 m -Flow rates = 40 mL min <sup>-1</sup> Room temperature = 22 °C	Up to 94% NH <sub>3</sub> recovery	Vecino et al. (2019)
	N	Laboratory scale -Gas-permeable membrane -Flow rate: 5.8 L d <sup>-1</sup> -Temperature = 25 °C	55% NH <sub>3</sub> recovery (without pH adjustment) 81% NH <sub>3</sub> (with pH adjustment)	Garcia-González and Vanotti. (2015)
	N, P	-Gas-permeable membrane -Low aeration addition to magnesium chloride (MgCl <sub>2</sub> ) to P precipitation	> 93% NH <sub>4</sub> -N removal efficiency > 91% P removal efficiency	Szogi and Vanotti (2009)
Struvite formation	N, P	Bittern used for struvite precipitation	> 90% P removal, 23–29% NH <sub>4</sub> <sup>+</sup> reduction	Zhang et al. (2012)
	N, P	Effect of P: Mg: N ratio, Effect of stirring speed and pH Pilot scale Continuous operation Aeration	P: Mg: N = 1:1:1.294 – 99% PO <sub>4</sub> <sup>3-</sup> -removal >80% NH <sub>4</sub> -N removal	Limoli et al. (2016)

Times = 1–4 h ; HRT = 6–15 h; Temperature = 0–29.5 °C; Experimental period = 247 days; 85% P removal 60 N, P Zeolite addition, Zeolite modified at pH 8.0–9.5; 82% NH<sub>3</sub>-N removal; 98% PO<sub>4</sub><sup>3-</sup>removal; 82N, P Zeolite fly ash synthesized using alkaline hydrothermal technology ;74% N removal 96% P removal

Tableau 4 : Overview of manure treatment approaches; anaerobic digestion, reproduced from Yao et al., (2020)

Approaches	Descriptions and results
<b>Solid-liquid separation</b>	<ul style="list-style-type: none"> <li>• Separate manure slurry into solid and liquid fraction.</li> <li>• Types: sedimentation and mechanical separation.</li> <li>•Relative simplicity of the process and low costs of the equipment involved.</li> </ul>
<b>Thermal concentration</b>	<ul style="list-style-type: none"> <li>• Reduce the moisture content of manure by external heating.</li> <li>•Concentrate nutrients ion to reduce transportation costs.</li> </ul>
<b>Combustion/incineration</b>	<ul style="list-style-type: none"> <li>• Complete the thereto-chemical oxidation of organic incineration matter in manures in order to obtain recoverable heat, ashes and gasses.</li> <li>•For manure with high moisture contents, the pre-drying process is generally required to reduce moisture contents.</li> </ul>
<b>Acidification</b>	<ul style="list-style-type: none"> <li>• Application of acidic reagents such as sulfuric acid to manure slurry to lower its pH.</li> <li>•Reduce ammonia emissions and deactivate pathogens.</li> </ul>
<b>Anaerobic digestion</b>	<ul style="list-style-type: none"> <li>• Include a series of biological processes in which microorganisms break down organic substances of manure in absence of oxygen.</li> <li>•Produce a mixture of gases (mainly methane and carbon dioxide) that can be used as bioenergy.</li> </ul>
<b>Composting</b>	<ul style="list-style-type: none"> <li>• Aerobic biological decomposition and stabilization processes that convert the solid manure into compost.</li> <li>•Types: passive aeration, windrow/mechanical agitation, forced aeration/aerated static pile, in-vessel systems.</li> <li>•Inactivation of weed seeds and some pathogens.</li> <li>•Composted manure is less susceptible to leaching and further ammonia losses. Compost is sufficiently stable for storage and subsequent soil application.</li> </ul>
<b>Biological nitrogen removal</b>	<ul style="list-style-type: none"> <li>• Biological conversion of ammonium to inert nitrogen gas using classical nitrogen removal treatment.</li> <li>•Combine autotrophic nitrification and heterotrophic denitrification processes.</li> <li>•The feedstock is generally the liquid fractions separated from slurry.</li> </ul>
<b>Membrane filtration</b>	<ul style="list-style-type: none"> <li>• Removal of suspended solids (SS), colloids, bacteria and nutrients from liquid through membrane filtration.</li> <li>•Require manure pre-treatments to prevent fouling, maximize membrane life, and increase flux.</li> </ul>

### Volatile wastes' treatment techniques

Air washing is a technique that is widely applied in industries in various fields, such as in solvent production sites, waste incineration sites, bone degreasing units, and in urban wastewater treatment plants.... In agriculture, this technique is often used downstream of livestock (pigs, chicken and cattle) breeding farms mainly in the Netherlands. It has been introduced, later in France to regulate volatile gaseous emissions and particularly, ammonia. (Guingand, 2008). Indeed, the use of air scrubbers in pig breeding buildings has become an obligation to obtain the "Green Label". This regulatory approach has encouraged Dutch farmers to introduce and develop air washers in a consistent manner and to adapt them to the specific

constraints of pig farming. The implementation of this conditioned labelling process, with a view to protecting the environment, has significantly favoured the reduction of ammonia emissions related to pig farm activities (Guinand, 2008; Dumont *et al.*, 2019). This washing process consisting in the solubilising of volatile molecules present in the air of pig barns, under gaseous form (notably ammonia  $\text{NH}_3$ ) in the fresh water by converting them to ammonium ( $\text{NH}_4^+$ ); nitrite ions ( $\text{NO}_2^-$ ) and nitrate ions ( $\text{NO}_3^-$ ) forms by the denitrifying microbial communities present in the water, under controlled acid pH in order to increase ammonia solubility (Guinand, 2008; Van Der Heyden *et al.*, 2015; Loyon *et al.*, 2016; Dumont *et al.*, 2019). According to Van Der Heyden *et al.*, (2015), this process would allow the removal of up to 75 % of the odour nuisance (Guinand et Deforges, 2008), up to 95% of dust (Guinand et Garnier, 1996) and about up to 89% of ammonia (Lais *et al.*, 1997), 90% of ammonia removed can be reached by acid extraction in Netherlands, in France, acid scrubbing is legally banned as reported by Guinand, (2008). However, the progressive accumulation of nitrogen salts ( $\text{NH}_4^+ + \text{NO}_2^- + \text{NO}_3^-$ ) leads to saturation of the water in the buffer tank used in washing, which limits ammonia removal. This implies continuous monitoring of the quality of the water used in washing by adding a new volume of fresh water (Dumont *et al.*, 2019).

## **5. Bioprocess for livestock wastes and effluents treatment: Photobioreactors and microalgae strains.**

As it was highlighted in the section 4, the treatment of livestock effluents in agriculture can be carried out by several physico-chemical methods (Table 4; Dumont *et al.*, 2019; Montalvo *et al.*, 2020). However, these techniques are not very effective when the water to be treated contains several forms of nitrogen simultaneously and at high concentrations (several hundred  $\text{mg N.L}^{-1}$ ) (Dumont *et al.*, 2019). Hence, the tendency to use microalgae; photosynthetic organisms with various interesting biochemical characteristics such as accumulation of nitrogenous nutrients, phosphorus, inorganic carbon and heavy metals for a wide range of applications including wastewater treatment (Table 1; Table 3; Table 4). Then, these microorganisms can be used in various fields, namely: in human and animal food, in energy production (biofuels), and in the production of molecules with high added value for industrial purposes (Ben Amor, 2015; Table 1).

In this section, the main microalgae strains involved in the treatment of livestock wastes, and the bioprocesses deployed will be pointed and discussed.

### **Microalgae strains**

Many microalgae species are used in wastewater treatment dispositives including species of freshwater and seawater (Table 1; Table 5). Among the most used microalgae strains, the class Chlorophyceae are the most represented with the genera *Scenedesmus* (Gao *et al.*, 2016, Wang *et al.*, 2016; Prandini *et al.*, 2016; Labbe *et al.*, 2017; Daneshvar *et al.*, 2018), *Chlorella* (Wang *et al.*, 2015; Franchino *et al.*, 2016; Gao *et al.*, 2016; Wang *et al.*, 2016; Labbe *et al.*, 2017; Leite *et al.*, 2019), *Desmodesmus* (Mehrabadi *et al.*, 2017; Eze *et al.*, 2018; Hernández-García *et al.*, 2019) and *Chlamydomonas* (Kamyab *et al.*, 2015; Mohammadi *et al.*, 2018) as it has been reported in the Table 1 and Table 5.

Tableau 5 : Microalgae strains for wastewater treatment and their nutrient removal efficiency adapted from Nwoba et al. (2020).

Microalgae sp.	Wastewater source	%N removal	%P removal	Reference
<i>Scenedesmus obliquus</i>	Municipal wastewater	80-99	54-98	Martinez et al. (2000)
<i>Chlorella sp.</i>		93	81	Li et al. (2011)
<i>Chlorella sp.</i>	Digested manure	76-83	63-75	Wang et al. (2010)
<i>Chlorella pyrenoidosa</i>	Industrial wastewater	70	44	Hongyang et al. (2011)
<i>Chlorella pyrenoidosa</i>	Diluted piggery wastewater	60 77.7 <sup>a)</sup>	80	Wang et al. (2012)
<i>Chlorella vulgaris</i>	Municipal wastewater	24-99	<30-78	Aslan and Kapdan (2006)
<i>Chlorella soroldniana</i>		45-72	88	Hernandez. et al. (2006)
<i>Chlorella vulgaris</i>		55-88	12-100	Ruiz-Marin et al. (2010)
<i>Scenedesmus obliquus</i>		79-100 <sup>a)</sup>	30-100	Zhang et al. (2008)
<i>Spindina (Anthrospim) sp.</i>	Animal wastewater	84-90 <sup>a)</sup>	72-87 14 <sup>b)</sup>	Olguín et al. (2003)
<i>Anthrospira platensis</i>	Industrial wastewater	96-100 <sup>a)</sup>	87-99 <sup>b)</sup>	Phang et al. (2000)
<b>Polyculture</b>	Municipal wastewater	99	99	Woertz et al. (2009)
<i>Chlorella vulgaris</i> (Mixotrophic)	Swine degistates (8 folds dilution)	98.7-99.8 <sup>c)</sup>	41.0-62.5	Kwon et al. (2019)
<i>Chlorella vulgaris</i> (Mixotrophic)	Cattle	83.16-94.27 <sup>a)</sup>	90.98-94.41	Lv et al. (2018)
<i>Chlorella minutissima, Chlorella sorokiniana, Scenedesmus bijuga</i> (Mixotrophic consortium)	Poultry	Up to 60	Up to 80	Singh et al. (2011)
<i>Chlorella vulgaris</i>	Manure free piggery wastes	Up to 100 <sup>a)</sup>	Up to 99	Zheng et al. (2019)
<i>Dunaliella sp.</i>	Digested poultry litter	63.8	64.1	Han et al. (2019)

<sup>a)</sup> Ammonia nitrogen (NH<sub>4</sub><sup>+</sup>-N) ; <sup>b)</sup> Total orthophosphates (PO<sub>4</sub><sup>3-</sup>-P) ; <sup>c)</sup> (NH<sub>3</sub>-N)

These Chlorophyceae are widely used in livestock wastewater treatment bioprocesses not only for their potential for of dairy, piggery, poultry and aquaculture wastes remediation, but also for their high productivity, bioaccumulation of precious molecules and their tolerance to elevated concentration of nutrients and their valorisation into biomethane, biofuel, biohydrogene or bioelectricity (Carlsson et al., 2007; Rowley, 2010; Ben Amor, 2015).

The genus *Chlorella* has particularly a high capacity for photosynthesis (Richmond, 2004; Rowley, 2010; Clement-Larosiere, 2014; Ben Amor, 2015). It's considered among the most promising algae for biofuel production, mainly the species *Chlorella sp.*, thanks to its high lipid content between 14 and 40% of its dry weight (Powell et al, 2009; Taher et al, 2011; Feng et al.,2011; Zhou et al., 2011; Marjakangas et al., 2015; Shen et al.,2015; Lam et al.,2017).*Chlorella vulgaris*, is considered, therefore as a potential candidate algae for animal effluents purification (Table 5) thanks to its particular tolerance to high ammonium concentrations ,(Kwon et al., 2020) . It enables the recovery of respectively up to 94.27 % of ammonia ,94.41%of TP and 91.24 % of COD from the cattle waste medium. (Lv et al.,2018; Table 5). *Chlorella minutissima*, *Chlorella sorokiniana* and *Scenedesmus bijuga* consortium, cultivated in mixotrophy, allowed the recovery of respectively up to 60%,80% of TN and TP from the digested poultry litter (Singh et al.,2011; Table 5). The results for *Chlorella pyrenoidosa*, were up to 60%, 77.7%,80% and 57.6% of respectively TN, NH<sub>4</sub><sup>+</sup>, TP and COD removal from the initial substrate of diluted piggery wastewater (Wang et al.,2012)

Indeed, *Chlorella vulgaris*, is widely used in studies and laboratories, due to its easy cultivation and its biochemical characteristics; it is an ubiquitous alga, found in the aquatic environment, but also in the soil. It tolerates a great variation of salinity and a large pH range, from 3 to 10. (Richmond, 2004; Rowley, 2010; Clement-Larosiere, 2014; Ben Amor, 2015).

## Photobioreactors

The growth characteristics and metabolites accumulation strongly depend on the type of cultivation: phototrophic (using light and CO<sub>2</sub>), heterotrophic (without light and organic carbon), and mixotrophic (using light, CO<sub>2</sub>, and organic carbon). Phototrophic cultivation is the most common type used in large-scale microalgae production (Suparmaniam et al., 2019)

Generally, the different cultivation systems of microalgae in livestock wastewater have been carried out within lab scale into flask or batch systems (Table 5). Nevertheless, other photobioreactors (PBRs), microalgae biofilm photobioreactors (MBPBRs), a rotating microalga biofilm photobioreactor (RABPBR) or RAB (Abu Hasan et al.,2021) open ponds and raceways types and designs have been employed for the treatment of livestock wastewater with microalgae (Table 2; Table 6). Microalgae biofilms or autotrophic biofilms (Choudhary et al.,2017) are a consortium of microalgae (including cyanobacteria) with heterotrophic microorganisms (bacteria, fungi, and protozoa) and other microorganisms which are embedded in extracellular polymeric substance (EPS matrix) (Mantzorou and Ververidis, 2019).As it was explained in section 2, open ponds and raceways are open systems which are very dependent on the meteo parameters, PBRs (section 1) and MBPRs and RABs are closed systems, the cultivation parameters are controlled.

In the Table 6, some characteristics and operating conditions of different PBRs systems are detailed. These parameters influence the PBR nutrient recovery efficiency, in addition to the biotic conditions like the microalgal strain and its abiotic requirements (T°, C/N ratio, N/P ratio, luminosity...).



Tableau 6 : Operating conditions of some PBR treating livestock and municipal wastewater reproduced from Abu Hasan et al. (2021) and Rios Pinto, Ferreira and Tasic (2021).

<b>Raceway pond</b> (15 cm depth, 1500 L). Cultivation in batch and semi continuous modes Pilot-scale	<i>Chlorella sp.</i> and <i>Scenedesmus sp.</i> consortium Anaerobically digested piggery effluent	Biomass productivity: 60–61mgL <sup>-1</sup> d <sup>-1</sup> Lipid productivity: 5.4–21mgL <sup>-1</sup> d <sup>-1</sup>	Raeisossadati et al. (2019) ; (Rios Pinto, Ferreira and Tasic, 2021)
<b>MBPBR:</b> -Grid plastic, gauze +baffle, cotton cloth, glass fiber, and a standard cotton cloth -Light intensity: 20010 μmol.m <sup>-2</sup> s <sup>-1</sup> - HRT: 5days	<i>Chlorella vulgaris</i> UTEX 2714 Hog manure wastewater	COD: 95.67% TP: 64.40% TN: 69.55% NH <sub>4</sub> <sup>+</sup> -N: 91.24%	Chen et al. (2019) Abu Hasan et al. (2021)
<b>Thin layer and turf scrubber Thin-layer reactor</b> (0.5 cm depth, 350L). Cultivation in batch and semicontinuous modes Pilot-scale	<i>Chlorella sp.</i> and <i>Scenedesmus sp.</i> consortium Anaerobically digested piggery effluent	Biomass productivity: 31–45mgL <sup>-1</sup> d <sup>-1</sup> Lipid productivity: 8.1–29mgL <sup>-1</sup> d <sup>-1</sup>	Raeisossadati et al. (2019) (Rios Pinto, Ferreira and Tasic, 2021)
<b>MBPBR:</b> Glass plate and attached algal biofilm disks	<i>Chlorella pyrenoidosa</i> Swine wastewater	NH <sub>4</sub> <sup>+</sup> -N: 75.9% TP: 68.4% COD: 74.8% Zn <sup>2+</sup> : 65.7% Cu <sup>2+</sup> : 53.6% Fe <sup>2+</sup> : 58.9%	Cheng et al. (2017) Abu Hasan et al. (2021)
<b>RAB:</b> -Material of disk: Plastic drum (41 cm) - Rotating speed: 4.8rpm -Light intensity: 20020μmol.m <sup>-2</sup> s <sup>-1</sup> - HRT: 1 2-day 6-day	<i>Chlorella sorokiniana</i> Municipal wastewater	TN : 93.7 % TP : 80.6 %  TN : 81.3 % TP : 99.5%	Shayan et al. (2016)  Abu Hasan et al. (2021)

## 6. Efficiency and feasibility of these treatment: a critical overview

### 6.1. Efficiency:

Photobioreactors for treating livestock effluents, especially pig, bovine and poultry, have shown great performance for nutrients removal. These systems using microalgae as a mean for wastewater treatment, contribute to minimize the use of energy, chemical substances (flocculants, coagulants ...) and allow water recycling by minimizing evaporation losses, and by recycling treated water in watering, cleaning or even in the reuse of cultivation of other microalgae (Hadj-Romdhane et al.,2012). The microalgal biomass produced is also destined for valorization, which creates a kind of circular economy or green economy, contributing to the protection of exhaustible natural resources such as water and presenting a stable revenue due to the added value of the targeted products that creates an additional mean of incomes.



As it has been shown in the tables 5 and 6 and according to several reviews and studies (Solmaz and Işık, 2020; Galanakis, 2021) PBRs and especially MBPRs represent a promising way for wastewater treatment (mainly livestock wastes). The nutrients removal efficiency can reach up to 93.7%, 99.5% and 74.8% for TN, TP and COD removal. Biomass productivity is up to  $45\text{mgL}^{-1}\text{d}^{-1}$ , and lipid productivity is up to  $29\text{mgL}^{-1}\text{d}^{-1}$  (Table 6). However, the nutrient recovery efficiency is highly depending on the PBR system and design, the fixed cultivation conditions, mainly N/P ratio, HRT and luminosity or light intensity. According to several studies, the MBPR technique is considered to be the best approach for livestock wastes secondary treatment (Solmaz and Işık, 2020; Zhang *et al.*, 2020), as it enhances the total solids (TS) amount removal, which influences the cultivation medium turbidity and thus the light penetration and the photosynthesis that impacts in its turn the algal biomass productivity and nutrients removal (Solmaz and Işık, 2020; Zhang *et al.*, 2020). According to Ambat *et al.* (2019) Solmaz and Işık (2020), a high HRT period  $> 3$  days (Zhang *et al.*, 2020) leads to a low nutrient loading which impacts the N/P ratio due to P depletion in the cultivation medium. Nitrogen is not considered as a limiting factor in livestock wastewater for microalgae cultivation. Zhang *et al.* (2019) and (2020), reported that an optimal HRT of 2 days, an N:P ratio of 3.9: 1 had an optimal algal biomass production and nutrient removal for an MBPR system with *Chlorella vulgaris* and *Scenedesmus* consortium cultivated in mixotrophy. Indeed, Kwon *et al.* (2020), confirmed that in a heterotrophic, or mixotrophic culture medium, where algal biomass is associated with bacterial populations, N/P and C/N ratios play an important role in ensuring the equilibrium of the culture medium in terms of inorganic nutrients and carbon dioxide for the microalgae, and organic matter and  $\text{O}_2$  for the bacterial functioning (metabolism of nitrogen compounds and respiration). An N/P ratio of 5-10 (Li *et al.*, 2011; Wang *et al.*, 2012; Kwon *et al.*, 2020), C/N ratio of 5 -15, a light intensity of 1500 Lux were optimum for 99% of  $\text{NH}_3$  and 62% of TP removal from air-stripped piggery digested manure, with *Chlorella vulgaris* and bacteria consortium in mixotrophy (Kwon *et al.*, 2020) which matches the range of N/P ratio values of livestock wastes mentioned in Table 1, Annexes I, II and III. These N/P ratios fluctuate between  $5.4\pm 3.39$  (Cai *et al.*, 2012) and  $14.5\pm 3.54$  (Nwoba *et al.*, 2020) for hog manures (Annex I), between  $3.25\pm 1.77$  (Cai *et al.*, 2011) and 10 (Nwoba *et al.*, 2020) for cattle wastes (Annex II), and between  $3.95\pm 0.49$  (Cai *et al.*, 2012) and 32 (Nwoba *et al.*, 2020) for poultry excreta (Annex III).

Optimal N/P ratios vary according to the cultivation medium composition, the algal strains, and the bacterial consortia. According to Chen *et al.* (2012), consumed N/P ratio is in the range of 72.2-76.9 for freshwater algae cultivated in anaerobically-digested manure. Wang *et al.* (2013) reported that *Oedogonium sp.* grown in digested piggery wastewater has an optimal N/P ratio from 4.4 up to 16.0. Concerning *Chlorella sp.*, this ratio is about 7.5-9.5 for a growth medium supplemented with digested dairy wastewater (Wang *et al.*, 2010). Whereas *Chlorella vulgaris* has shown optimal growth and TN and TP removal for an N/P ratio of 11.7-14.4 in air-stripped aerobically digested piggery wastewater and air-stripped pre-treated raw swine wastewater (Kwon *et al.*, 2020). Indeed, Kwon *et al.*, 2020, confirmed that *C. vulgaris* used for the treatment of piggery wastewater hasn't shown any sign of inhibition for a 10-folds diluted air stripped pretreated swine wastes containing with initial concentrations of 2000 mg COD/L, 220 mg TN/L, 100 mg  $\text{NH}_3\text{-N/L}$ , 40 mg TP/L. Similar results were investigated by Zhu *et al.*, 2013, where under the controlled conditions of 1900 mg/L COD, 80 mg/L TN and 85 mg /L TP *Chlorella sp.*, has shown optimal growth in diluted piggery wastewater. Moreover, anaerobic digestate containing initial concentrations of 200 mg/L TN and 2.5 mg/L TP, provided optimal conditions for *C. zofingiensis* and *Chlorella sp.* cultivation according to Chen *et al.*, 2012. These concentrations are by far smaller than the initial concentrations in the raw livestock wastes (Table 2).

The efficiency of the system will also depend on the initial algal biomass amount inoculum introduced into the system., and the luminosity which can produce photoinhibition in case of excessive luminosity. On the contrary, it could be deficient (due to the accumulation of algal biomass on the walls of the photobioreactors), which results in a slowing of growth. Hence the importance of proper agitation in the culture system (Richmond, 2007; Hadj-Romdhane et al.,2012; Ben Amor, 2015; Abu Hasan et al.,2021). In the lab scale, the optimum light intensity is variable depending on the algal strain. In literature, it was reported to be in the range of 20–1000  $\mu\text{mol.m}^{-2}.\text{s}^{-1}$ , and the most commonly 150  $\mu\text{mol.m}^{-2}.\text{s}^{-1}$ . According to Ji et al., (2013) optimal luminosity for *Desmodesmus sp.*, is about 98  $\mu\text{mol.m}^{-2}.\text{s}^{-1}$ , on the other hand, it's about 700  $\mu\text{mol.m}^{-2}.\text{s}^{-1}$  for *Dunaliella viridis* (Gordillo et al.,1998; Galanakis, 2021). Light cycles (dark: light) as well, impact sharply the performance of the microalgae. Several studies reported that the minimum viable photoperiod is (8:16) and the maximum is (24:0). Whereas the most commonly used photoperiod is 12:12h (dark: light). Indeed, a 24h of light exposure increased biomass productivity (Rai et al., 2015; Galanakis, 2021).

Furthermore, the temperature and especially the pH (inhibition of the growth of *C. vulgaris* if alkaline pH > 10), will impact the growth velocity, cell dimension, metabolites composition and accumulation in the algal cells, and the concentrations of the different nitrogenous forms in the culture medium, as well as the CO<sub>2</sub> concentration. This leads to a change in the C/N, C/P and N/P ratios of the culture medium. The temperature depends on the algal strain but the optimum range for a lab scale is about 16-27°C (Ben Amor, 2015; Kwon et al.,2020; Galanakis, 2021).

## 6.2. Feasibility:

As it has been discussed in the previous section, PBRs have shown a promising treatment performance with high nutrients removal rates (Tables 5 and 6). However, these systems depend on several intrinsic and extrinsic, biotic and abiotic factors, and the majority of the reported studies concerned only lab scale's systems. Therefore, much more studies still be needed to overcome the challenges of the large-scale microalgae cultivation systems for wastewater treatment (Zhou et al., 2019; Liu et al., 2020).

The first factor that needs to be considered is the strain selection. The best strain is that one that has high productivity biomass, and high metabolic accumulation and uptake for the targeted chemical compounds to be extracted, then, a high tolerance for severe cultivation conditions. For livestock wastewater treatment, many literature reported that mixotrophic algal consortia isolated from these effluents are the best inoculum for an optimum treatment (Al-Jabri et al., 2020 ; Kwon et al.,2020; Nwoba et al., 2020) .However , the selection of the suitable strains still be needed for optimal results .

Water and nutrients availability is no longer a limiting factor for microalgae cultivation, since industries, agriculture and anthropic activities are loading a great amount of undervalued effluents containing abundant amounts of nutrients. Nevertheless, the abiotic parameters (T°, CO<sub>2</sub>, nutrients ..... ) that are compulsory for algal growth should be optimized and regulated depending on the algal strains.

These parameters that impact heavily biomass productivity and biochemical metabolism in microalgae cultivation are light intensity, photoperiod, temperature, nutrients, mixing, aeration, pH, and CO<sub>2</sub> absorption (Zhou et al., 2019; Liu et al., 2020; Al-Jabri et al., 2020; Galanakis, 2021). For lab scale, providing artificial illumination for indoor microalgae cultivation is necessary. Incandescent lamps and fluorescent tubes. For ideal photosynthesis performance in microalga, LEDs in the blue or red-light spectrum are especially used, as they are reported as the most active portions of light. They generally placed at 23 to 25 cm distance from the PBRs

for ensuring optimal luminosity (Rai et al., 2015; Koc, Anderson, and Kommareddy, 2013; Galanakis, 2021).

Moreover, biological (anaerobic digestion) and physico-chemical pre-treatment are reported as mandatory before processing to microalga cultivation. They must be applied to the raw livestock wastes as a primary treatment approach (Lv et al., 2018). In fact, all the reported studies in the previous sections confirm that PBRs are applied to diluted livestock digestates. This pre-treatment allows the removal of suspended matters and dark colour which reduce the PBRs productivity and efficiency, as they reduce luminosity (Montalvo et al., 2020). The PBRs design is also a very important factor of its efficiency (Abu Hasan et al., 2021). In fact, the higher the surface/volume ratio, the higher the cell concentration is, and consequently, the higher the photobioreactor volumetric productivity and rentability are.

Finally, the biggest challenge of the feasibility of these processes is mainly, energy consumption. As it has been previously discussed, many steps in the microalgae cultivation (in PBRs mainly) are involving high maintenance, high servicing and sophisticated technical assistance and processing. These are including upstream and downstream. In fact, closed indoor cultivation PBRs need power for mixing the culture, for water pumping, gas bubbling and distribution (CO<sub>2</sub>) pipelines, during the process. We should not forget as well, wastewater influents' pre-treatment, and PBRs design, manufacturing and conception, which are exclusively costly (Galanakis, 2021). Lower down in the processing chain of wastewater (respectively livestock wastes) treatment with microalga, harvesting (Table 7) and dewatering techniques are, among others the costliest steps. In fact, ultrasound assisted harvesting consume up to 200 MJ energy /Kg of microalgae for 83% of efficiency as reported by Bosma et al. (2003) (Table 7). Mastering these methods and choosing the appropriate ones is crucial, because they define the final yield product, the rentability of the whole treatment system and the quality of the treated water that could be reused and recycled for microalga cultivation or loaded in nature. Therefore, in this case, it should be meeting the international discharge standards. (Al-Jabri et al., 2020). Based on Table 7, several harvesting methods are used to separate the algal biomass from the bulk such as sedimentation, flocculation, filtration, coagulation .... etc. Whereas, the adoption of a harvesting technique would mainly depend on, the algal strain, the application of the produced biomass and the energy requirement per unit of biomass production. A comparison of different harvesting techniques, their energy consumption and their efficiency in accordance with the algal strain, the wastewater type and the cultivation system type has been detailed in the Table 7 as well.

Generally, to obtain a 20% solid matter biomass paste, a two-phase harvesting process is applied. During the first step, more or less inexpensive harvesting methods like sedimentation, flocculation and filtration are used in order to obtain a biomass slurry of about 1–4% solid matter. In a second step, more performant technique such as centrifugation is used to get biomass paste with in average above 20% solid matter concentration.

According to the cultivation system type and the final targeted product, the type of harvesting technique and the number of steps are fixed. Microalgae biomass destined for biogas production by anaerobic digestion, are harvested by inexpensive methods (filtration, sedimentation, flocculation, ...) since biogas production doesn't request a high quality or refined input product. Slurries are mostly used in anaerobic digestion (Al-Jabri et al., 2020).

Tableau 7 : Energy requirement and harvesting efficiencies in microalgae biomass harvesting reproduced from Al-Jabri et al. (2020)

Harvesting Method	Volume used (L)	Type of Cultivation System	Wastewater Type	Strain name	IBC (mg/L)	HE (%)	CF	FBC (g/L)	Energy Needed (MJ/Kg)	References
Self-cleaning Centrifugation	–	–	–	<i>Scenedesmus sp.</i> Mixed microalgae	1000 400– 700	– 90	120 ~100	120 40– 70	3.6 9.45*	Molina Grima et al. (2003) Sim et al. (1998)
Electrocoagulation	1	–	–	<i>Chlorella vulgaris</i>	300– 600	95	–	–	7.2	Vandamme et al. (2011)
Submerged Filtration	25	–	–	<i>Chlorella vulgaris</i>	410	98	14.7*	6.07	2.3	Bilad et al. (2012)
Vacuum Filtration (belt filter)	–	–	–	<i>Chlorella proboscideum</i>	1000	–	95	95	1.62	Molina Grima et al. (2003)
Tangential flow filtration	34.5	–	–	<i>Tetraselmis suecica</i>	600	–	78	46.8	11.82	Danquah et al. (2009)
Magnetic separation	200	–	–	<i>Botryococcus braunii</i>	1230	~92	–	–	13.17	Wang et al. (2014)
Dissolved air flotation	9750	–	–	Mixed microalga	400-700	–	~100	40- 70	5.87	Sim et al. (1998)
Ultrasound assisted harvesting	8.4	–	–	<i>Monodus subterraneus</i>	160-200	~83	~20	3.2- 4	~200	Bosma et al. (2003)
Pulse electrolysis	0.4	–	–	<i>Nannochloropsis oceanica</i>	~1000	96.4	–	–	1.8	Kim et al. (2014)
Coagulation flocculation	–	High rate algal ponds	Urban wastewater	Mixed microalga	–	90	–	–	–	Gutiérrez et al (2015)
Bioflocculation	–	Photobioreactor	Domestic wastewater	<i>Chlorella sp.</i>	–	98.9	–	–	–	Madkour et al. (2020)
Auto-flocculation	–	Photobioreactor	Pretreated swine wastewater	<i>Chlorella sp.</i>	–	39.5	–	–	–	Cheng et al. (2020)
Belt-filtration system	–	Photobioreactor	Domestic wastewater	<i>Scenedesmus sp.</i>	–	46– 84	–	–	–	Sutherland and Ralph (2019)

IBC = initial biomass concentration, CF = concentration factor, FBC = Final biomass concentration, HE = harvesting efficiency. \* was determined by the correlating centrifuge's electricity consumption, harvesting efficiency, and initial biomass concentration

Open systems like High rate algal ponds (HARAP) are also employed in livestock wastewater treatment, mainly outdoor in small agglomerations with cheaper installation. They are economical, since they do not require a huge amount of costly energy and assistance like PBRs. However, they don't afford fix biomass productivity since they are extremely dependent on meteorological conditions. The product quality is not well controlled. Thus, they are easy to construct and operate continuously. However, they encounter major limitations, such as poor light intensity, evaporative losses of water, diffusion of CO<sub>2</sub> to the atmosphere and requirement of large areas of land. Which is not faced in the case of PBRs (Rios et al., 2020; Abu Hasan et al., 2021, Galanakis, 2021).

## 7. Conclusion

The increasing amount of livestock manures makes their treatment and recycling mandatory. Their rich chemical composition makes them a good valuable medium for microalgal production through biological treatment such as MBPRs, PBRs and open ponds. The produced microalgal biomass is then valuable under several forms, such as bioenergy, biomethane source (after anaerobic digestion); bioelectricity, biofertilizer for crops and feed for animals and fish. They can also be valued as Human dietary supplementing pharmaceutical and food industries. However, MBPRs, PBRs as biological method of wastewater treatment, are not sufficient alone. A pre-treatment of raw livestock manure is compulsory as a primary step before MBPRs and PBRs treatment. These approaches have high treatment efficiency (up to 99% of NH<sub>3</sub> and 62% of TP removal) and a high yield of microalgal biomass of about 0.57 gL<sup>-1</sup>. d<sup>-1</sup> (Kwon et al., 2020), and is considered among the best alternative for low-cost wastewater treatment. Various species of microalgae, including freshwater and marine microalgae, are capable of forming biofilms and have been shown to act as bioremediation agents depending on environmental factors (light, pH, CO<sub>2</sub>, temperature), surface materials, and nutrients availability in the cultivation medium, and finally, on the PBR design.

Chlorophyceae microalgae, including *Chlorella vulgaris*, represent a great potential for nutrients recovery from high concentrated livestock wastes. Nevertheless, the culture parameters of these systems need to be well monitored to achieve optimal microalgal growth and nutrients removal, and to ensure the cost-effectiveness of these bioprocesses.

Indeed, efficiency and viability depend on several biotic and abiotic factors, such as the nutrients' initial concentrations, the microorganisms' composition in the effluents - that can affect the nutrient uptake by the microalgae-, the pH, the CO<sub>2</sub> concentration, the light intensity, the turbulence and the temperature. Therefore, obtaining optimal growth implies a good determination of these parameters.

A very important factor that could impact the PBRs effectiveness, is harvesting techniques of microalgal biomass or the biorefinery approach that, if not well mastered, could significantly increase the cost of the production, and thereby, reduce the economic viability of the whole process and production line. Therefore, more studies should be done to investigate these processes.

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# ANNEXES:

## Annex I: Characteristics of swine manure: literature review

Livestock farming type /Effluent type	K <sup>+</sup> [mg/L]	Fe <sup>2+</sup> [mg/L]	Ca <sup>2+</sup> [mg/L]	Cu <sup>2+</sup> [mg/L]	Mg <sup>2+</sup> [mg/L]	Zn <sup>2+</sup> [mg/L]	Mn <sup>2+</sup> [mg/L]	Na <sup>+</sup> [mg/L]	Cl <sup>-</sup> [mg/L]	N/P	Country	References
<b>Swine</b>												
Slurry	-	-	-	-	-	-	-	-	-	5.4±3.39	USA	Cai <i>et al.</i> (2012)
Slurry	-	-	-	-	-	-	-	-	-	-	-	Khoshnevisan <i>et al.</i> (2021)
Slurry (b)	-	-	-	-	-	-	-	-	-	-	France	Sharpley <i>et al.</i> (2004); Fuchs <i>et al.</i> (2014)
Slurry	-	-	-	-	-	-	-	-	-	-	Taiwan China	Wang <i>et al.</i> (2015)
Slurry	1010	-	620	4212	210	3105	715	-	-	-	Brazil	Bertagnoli <i>et al.</i> (2020)
Slurry	3418	-	-	-	-	-	-	-	-	-	USA	ASAE (2003); Ma <i>et al.</i> (2018)
Slurry	-	-	-	-	-	-	-	-	-	-	Japan	Li and Kobayashi (2010); Ma <i>et al.</i> (2018)
Slurry	-	-	-	-	-	-	-	-	-	-	-	Mingzi <i>et al.</i> (2016). Lee <i>et al.</i> (2021)
Slurry	-	-	-	0.39 ± 0.01	-	0.31 ± 0.00	-	-	-	-	China	Cao <i>et al.</i> (2018)
Slurry	-	-	-	-	-	-	-	-	-	-	Spain	Molinuevo-Salces <i>et al.</i> (2016)
Slurry (c)	-	-	-	-	-	-	-	-	-	-	-	Monfet et Unc (2016)
Raw slurry	-	-	-	-	-	-	-	-	-	-	Korea	Lee <i>et al.</i> (2021)
Raw slurry	-	-	-	-	-	-	-	-	-	-	China	Wang <i>et al.</i> (2012)
Raw slurry	385.12 ± 22.34	-	24.76 ± 12.83	<0.05	7.34 ± 0.27	<0.05	<0.01	-	-	-	China	Wen <i>et al.</i> (2017)
Raw slurry (d)	-	-	-	-	-	-	-	-	-	-	Korea	Kwon <i>et al.</i> (2019)
Autoclaved slurry	381.45 ± 22.58	-	23.77 ± 7.29	<0.05	7.06 ± 0.53	<0.05	<0.01	-	-	-	China	Wen <i>et al.</i> (2017)
Centrifuged and sterilized slurry	1.65 ± 0.08	1.29 ± 0.06	9.84 ± 0.07	5.18 ± 0.09	1.33 ± 0.09	3.95 ± 0.05	0.92 ± 0.02	-	-	-	China	Zheng <i>et al.</i> (2019)



Livestock farming type /Effluent type	K <sup>+</sup> [mg/L]	Fe <sup>2+</sup> [mg/L]	Ca <sup>2+</sup> [mg/L]	Cu <sup>2+</sup> [mg/L]	Mg <sup>2+</sup> [mg/L]	Zn <sup>2+</sup> [mg/L]	Mn <sup>2+</sup> [mg/L]	Na <sup>+</sup> [mg/L]	Cl <sup>-</sup> [mg/L]	N/P	Country	References
<b>Swine</b>												
Liquide manure	-	-	-	-	-	-	-	-	-	14.5± 3.54	Africa	Nwoba <i>et al.</i> (2020)
Liquide manure	-	-	255	-	-	-	-	-	-	-	-	Suzuki <i>et al.</i> (2007) ; Katakai <i>et al.</i> (2016)
Liquide manure	-	-	61	-	-	-	-	-	-	-	-	Perera <i>et al.</i> (2007) ; Katakai <i>et al.</i> (2016)
Liquide manure	-	-	-	-	-	-	-	-	-	-	-	Ryu <i>et Lee</i> (2010) ; Katakai <i>et al.</i> (2016)
Liquide manure	-	-	135	-	-	-	-	-	-	-	-	Huang <i>et al.</i> (2011a, b) ; Katakai <i>et al.</i> (2016)
Liquide manure	-	-	-	-	-	-	-	-	-	-	-	Zhang <i>et al.</i> (2012) ; Katakai <i>et al.</i> (2017)
Digestate	-	-	-	-	-	-	-	-	-	-	Taiwan, China	Marjakangas <i>et al.</i> (2015)
Digestate	-	-	-	-	-	-	-	-	-	-	Australia	Kumar <i>et al.</i> (2010)
Digestate	809.25	1.46	165.91	0.23	36.97	1.56	0.19	312.30	438.2	-	China	Cheng <i>et al.</i> (2015)
Digestate	-	-	-	-	-	-	-	-	-	-	USA	Deng <i>et al.</i> (2017)
Digestate	-	-	-	-	-	-	-	-	-	-	Australia	Ayre <i>et al.</i> (2017)
Digestate	-	-	-	-	-	-	-	-	-	-	China	Zeng <i>et al.</i> (2020)
Digestate (c)	-	-	-	-	-	-	-	-	-	-	-	Monfet <i>et Unc</i> (2016)
Digestate (d)	-	-	-	-	-	-	-	-	-	-	Korea	Kwon <i>et al.</i> (2019)

## Annex II: Characteristics of cattle manure: literature review

Livestock farming type /Effluent type	K <sup>+</sup> [mg/L]	Fe <sup>2+</sup> [mg/L]	Ca <sup>2+</sup> [mg/L]	Cu <sup>2+</sup> [mg/L]	Mg <sup>2+</sup> [mg/L]	Zn <sup>2+</sup> [mg/L]	Mn <sup>2+</sup> [mg/L]	Na <sup>+</sup> [mg/L]	Cl <sup>-</sup> [mg/L]	N/P	Country	References
<b>Cattle</b>												
Slurry	-	-	-	-	-	-	-	-	-	10	Africa	Nwoba et al. (2020)
Slurry	-	-	-	-	-	-	-	-	-	-	-	Khoshnevisan et al. (2021)
Centrifuged slurry (supernatant)	-	-	-	-	-	-	-	-	-	-	China	Lv et al. (2018)
Beef slurry	-	-	-	-	-	-	-	-	-	3.25±1.77	USA	Cai et al. (2011)
Dairy cattle slurry	-	-	-	-	-	-	-	-	-	4	Africa	Nwoba et al. (2020)
Dairy cattle slurry ***	-	-	-	-	-	-	-	-	-	-	Japan	Li and Kobayashi (2010) ; Ma et al.(2018)
Dairy cattle slurry (c)	-	-	-	-	-	-	-	-	-	-	-	Monfet et Unc (2016)
Dairy cattle raw slurry	1200	237	3700	14	600	18	16	336	-	-	USA	Dougherty et al. (2017)
Dairy cattle raw slurry	900	17	500	3	200	4	5	180	-	-	USA	Dougherty et al. (2017)
Dairy cattle raw slurry	1300	33	600	1	200	22	5	210	-	-	USA	Dougherty et al. (2017)
Dairy cattle effluents	-	-	80	-	-	-	-	-	-	-	-	Zhao et al. (2010) ; Kataki et al. (2016)
Dairy cattle effluents	-	-	-	-	-	-	-	-	-	-	USA	Uludag-Demirer et al. (2005) et al. (2005) ; Kataki et al. (2016)
Dairy cattle effluents	-	-	1735	-	-	-	-	-	-	-	-	Shen et al. (2011) ; Kataki et al. (2016)
Dairy cattle effluents	3338	-	-	-	-	-	-	-	-	-	USA	ASAE (2003) ; Ma et al. (2018)
Dairy cattle effluents	-	-	-	-	-	-	-	-	-	-	Japan	Li and Kobayashi (2010) ; Ma et al.(2018)
Dairy cattle degistate (c)	-	-	-	-	-	-	-	-	-	-	-	Monfet and Unc (2016)
Urine	-	-	-	-	-	-	-	-	-	-	-	Prabhu and Mutnuri (2014); Kataki et al. (2016)



### Annex III: Characteristics of poultry manure: literature review

Livestock farming type / Effluent type	K <sup>+</sup> [mg/L]	Fe <sup>2+</sup> [mg/L]	Ca <sup>2+</sup> [mg/L]	Cu <sup>2+</sup> [mg/L]	Mg <sup>2+</sup> [mg/L]	Zn <sup>2+</sup> [mg/L]	Mn <sup>2+</sup> [mg/L]	Na <sup>+</sup> [mg/L]	Cl <sup>-</sup> [mg/L]	N/P	Country	References
<b>Poultry</b>												
Manure (b)	-	-	-	-	-	-	-	-	-	-	-	Slaton <i>et al.</i> (2013) ; Fuchs <i>et al.</i> (2014)
Manure (b)	-	-	-	-	-	-	-	-	-	-	-	Guivarch et Morel (2001) ; Fuchs <i>et al.</i> (2014)
Manure (b)	-	-	-	-	-	-	-	-	-	-	-	Sharpley <i>et al.</i> (2004) ; Fuchs <i>et al.</i> (2014)
Manure (b)	22.45	-	51.03	93.41	10.12	1233.98	1012.3	-	-	-	Brazil	Bertagnoli <i>et al.</i> (2020)
Liquide effluents	-	-	-	-	-	-	-	-	-	32	Africa	Nwoba <i>et al.</i> (2020)
Slurry	-	-	-	-	-	-	-	-	-	10±8.49	USA	Cai <i>et al.</i> (2012) ; Nwoba <i>et al.</i> (2020)
Slurry	-	-	-	-	-	-	-	-	-	-	-	Yetilmezsoy and Sapci-Zengin (2009) ; Kataki <i>et al.</i> (2016)
Slurry	4547	-	-	-	-	-	-	-	-	-	USA	ASAE (2003). Ma <i>et al.</i> (2018)
Treated slurry (acidified and filtered)	1042.6 ± 19.0	-	631.6 ± 11.5	-	187.8 ± 3.0	-	-	177.9 ± 4.7	-	-	USA	Shashvatt <i>et al.</i> (2018)
Treated slurry (acidified and filtered)	1331.6 ± 23.4	-	952.4 ± 18.9	-	224.2 ± 5.1	-	-	433.6 ± 8.3	-	-	USA	Shashvatt <i>et al.</i> (2018)
Treated slurry (acidified and filtered)	1516.7 ± 17.8	-	754.1 ± 5.1	-	265.0 ± 2.9	-	-	334.8 ± 3.3	-	-	USA	Shashvatt <i>et al.</i> (2018)
Digestate	-	-	-	-	-	-	-	-	-	3.95±0.49	USA	Cai <i>et al.</i> (2012)
Litter degistate	1314.24	0.085	0.044	0.085	-	-	-	-	-	-	China	Han <i>et al.</i> (2019)
Digestate (b)	-	-	-	-	-	-	-	-	-	-	-	Monfet and Unc (2016)
Centrifuged degistate (supernatant)	1632-2100	22.4- 45.3	173-308	10.4 - 21.4	21.5 - 146	6.34- 13.4	4.57- 9.37	347-389	-	-	USA	Singh <i>et al.</i> (2011)

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Titre français : **Les microalgues pour le traitement des effluents d'élevage : bilan**

Titre anglais: **Microalgae for livestock wastewater treatment: an overview**

**Résumé :**

Les émissions élevées d'ammoniac ( $\text{NH}_3$ ) des installations destinées aux animaux, en particulier les porcs, le bétail et la volaille, ont un impact sur la pollution de l'air et de l'eau, ce qui peut entraîner des problèmes environnementaux (tels que l'eutrophisation) et des conflits entre les agriculteurs et leurs voisins. Plusieurs technologies sont utilisées pour gérer l'augmentation des productions d'effluents d'élevage dans le monde. Les déchets humides peuvent être éliminés directement par méthanisation (digestion anaérobie), ou par compostage (décomposition aérobie). Les émissions gazeuses d'ammoniac peuvent être traitées par biofiltration à l'aide d'épurateurs d'air. Cependant, ces techniques ne sont pas très efficaces lorsque l'eau à traiter contient plusieurs formes d'azote simultanément et à des concentrations élevées (jusqu'à  $1000 \text{ mg N. L}^{-1}$ ). C'est pourquoi on a tendance à utiliser des microalgues, organismes photosynthétiques présentant diverses caractéristiques biochimiques intéressantes (accumulation d'azote, de phosphore, d'oligo-éléments, de métaux traces et lourds) pour un large éventail d'applications, y compris le traitement des eaux usées. En outre, ces micro-organismes peuvent être utilisés dans divers domaines, à savoir : dans l'alimentation humaine et animale, dans la production d'énergie (biocarburants), et dans la production de molécules à haute valeur ajoutée à des fins industrielles (pharmaceutiques et cosmétiques). Les bioprocédés, tels que les photobioréacteurs utilisant des microalgues, principalement la *Chlorella sp.*, comme organisme extracteur de nutriments, sont considérés par les scientifiques comme des techniques prometteuses pour traiter les effluents d'agriculture.

**Abstract:**

The elevated ammonia ( $\text{NH}_3$ ) emissions from animal facilities, especially pigs, cattle and poultry, impact the air and water pollution which can lead to environmental problems (such as eutrophication) and conflicts between farmers and their neighbours. Several technologies are employed to manage the increasing of animal manure productions worldwide. Wet wastes can be removed directly by methanization (anaerobic digestion), or composting (aerobic decomposition). The gaseous ammonia emissions can be treated by biofiltration using air scrubbers. However, these techniques are not very effective when the water to be treated contains several forms of nitrogen simultaneously and at high concentrations (up to  $1000 \text{ mg N. L}^{-1}$ ) Hence, the tendency is to use microalgae; photosynthetic organisms with various interesting biochemical characteristics (accumulation of nitrogenous, phosphorus, oligo nutrients, trace and heavy metals) for a wide range of applications including wastewater treatment. Then, these microorganisms can be used in various fields, namely: in human and animal food, in energy production (biofuels), and in the production of molecules with high added value for industrial purposes (pharmaceutical and cosmetic fields). Bioprocess, such as photobioreactors using microalgae, mainly *Chlorella sp.*, for nutrients recovery, are considered by scientists as promising techniques for treating wastewater within a circular green economy process.

**Mots-clés :** Microalgues, *Chlorella sp.*, photobioréacteurs, effluents d'élevage, fumier, lisier, digestat, digestion anaérobie, bioénergie, traitement des effluents, volaille, porc, bovin.

**Key Words :** Microalgae, *Chlorella sp.*, photobioreactors, Livestock effluents, manure, slurry, digestate, anaerobic digestion, bioenergy, wastewater treatment, poultry, swine, cattle.