UNIVERSITÀ DEGLI STUDI DI MILANO

Livestock manure treatment for nutrients removal: consolidated techniques, emerging problems and new approaches

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PhD Thesis

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PhD in Environmental Sciences
XXXI° Cycle
Academic year: 2017-2018

Per te nonna Rina,
ti penso sempre.

Abstract

Intensive farming practices and their continuous spread generate a large amount of livestock manure that cannot be disposed properly. This can cause water, air and soil contamination. Therefore, the identification of strategies for correct management of livestock manure is necessary for limiting environmental pollution.

Nitrification/denitrification process (NDN) represents one of the most applied solutions. However, at real scale, many of those facilities that adopt this process suffer from managing issues and malfunctioning.

Recently, alternative techniques have been applied to the recovery of nutrients along with their removal. An example is solid/liquid separation using chemicals to remove phosphorus and concentrate it into solid fraction, easier to handle.

Other concerns are ammonia and greenhouse gases emissions derived from the management and the treatment of livestock manure. Their assessment is the first step to identify and apply proper mitigation strategies focused on limiting emissions.

In this thesis, the application of biological processes for the removal of nitrogen from livestock manure was investigated to find possible improvements.

First, a review of the state of the art of nitrification/denitrification process applied in the sequencing batch reactor system (SBR) to livestock manure was carried out to produce an overview on the SBR technology, monitoring parameters and process optimization. This can help the identification of SBR strengths and weaknesses when these systems are adopted for the livestock manure treatment and can support the definition of the best operation settings to adopt for reaching desired removal rates.

Several treatment plants were monitored to attest operational and removal efficiencies achieved. The aim of these monitoring campaigns was the identification of possible improvements for NDN (nitrification/denitrification) plants.

Eventually, the application of the NDN process in laboratory SBR systems was analysed to study how the nitrogen removal rate changes with the

variation of the slurry inlet characteristics and of the applied operational settings.

Concerning emissions, the use of different types of dynamic hoods and static chamber for estimating GHGs and NH_3 emissions derived from livestock manure was investigated.

Regarding recovery of phosphorus (P), the dissertation dealt with the evaluation of the effect of two additives (calcium hydroxide $Ca(OH)_2$ and aluminium sulfate $Al_2(SO_4)_3$) on the solid/liquid separation process of P, Copper and Zinc from raw slurry and co-digested slurry before and after a physico-chemical ammonia stripping treatment.

The results outlined that the performance of monitored SBR treatment plants at real scale are less than expected. Rarely TAN removal exceeds 50% even if at lab-scale observed removal rates were higher (up to 95%).

Pre-treatments are necessary to improve performance of treatment plants. Moreover, the upgrading of proper process control system and its correct use are required.

These should be considered also if a new SBR is designed.

Ammonia and GHGs emissions occur during the entire livestock manure management chain and their evaluation is possible applying the direct method. In particular, dynamic hoods can be useful tool for measuring site-specific NH_3 and GHGs emissions except for CH_4.

The application of solid/liquid separation process for phosphorus removal using aluminium sulphate allowed to reach removal rates higher than 70%. This additive enhanced also the removal of suspended solids (58%), copper (up to 94%) and zinc (up to 93%).

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SECTION 1 - INTRODUCTION AND BACKGROUND

1.The management of animal slurry for environmental sustainability

Growing populations, rising affluence and urbanization are translating into increased demand for livestock products, particularly in developing countries.
An increase of 70% of these products is foreseen to feed a population estimated to reach 9,6 billion by 2050 (FAO, 2018).

The emerging larger, more regionally intensive and more specialized animal production system can result in the possibility of manure mismanagement. This, consequently, can lead to a risk of pollution (Sommer et al., 2013). In addition, the intensive livestock production generates a large amount of manure that cannot be disposed properly.

Livestock manure represent any organic material that supplies organic matter to soils together with plant nutrients, usually in lower concentrations compared to inorganic fertilizer, produced by housed livestock. Typically, is a mixture of faces and urine with or without bedding material, depending on the type of animal housing system (Pain et al., 2011).
Often, the produced volume exceeds the capacity of environment around the livestock facilities to absorb nutrients contained in manure. One effect is that crops receive an amount of nutrients that exceeds the dosage required for their growth. Overall, there is an increased risk of salinization in semi-arid regions, and of water pollution (both surface and underground) and soil.
Uncontrolled and excessive manure applications and incorrect and inefficient manure management practices may severely damage environment. Negative effects affect multiple scales, ranging from the aforementioned local and regional contamination of water and soil to the global emission of greenhouse gases (GHGs)(FAO,2018) (Table 1).

The following sections illustrate an overview of different environmental impacts derived from the erroneous manure management.

Table 1 Animal slurry management contributes to environmental pollution at local, regional and global scale, depending on the substances involved (Sommer et al. 2013).

	Scale of Environmental impact		
Substance	Local	Regional	Global
Odour, H_2S	+		
Nitrate	+	+	
Phosphorus	+	+	
Ammonia	+	+	
Greenhouse gases			+
Heavy metals	+	+	
Pathogens	+	+	

1.1. The central role of nitrogen in agriculture

Nitrogen (N) is needed for growth and survival of all organisms.

It has indeed a major role as a nutrient in the agricultural production system. Nevertheless, the extensive use of nitrogen raises environmental concern (Burton et al., 2003). Besides, in some parts of the world, it led to an excess of food and unhealthy diets (Galloway et al., 2008).

The natural biological cycle of nitrogen (Figure 1) is a complex sequence of chemical transformations involving nitrogen in organic and inorganic compounds. This cycle takes place between the atmosphere and the living world and also includes an inner cycle between plants, animals and bacteria.

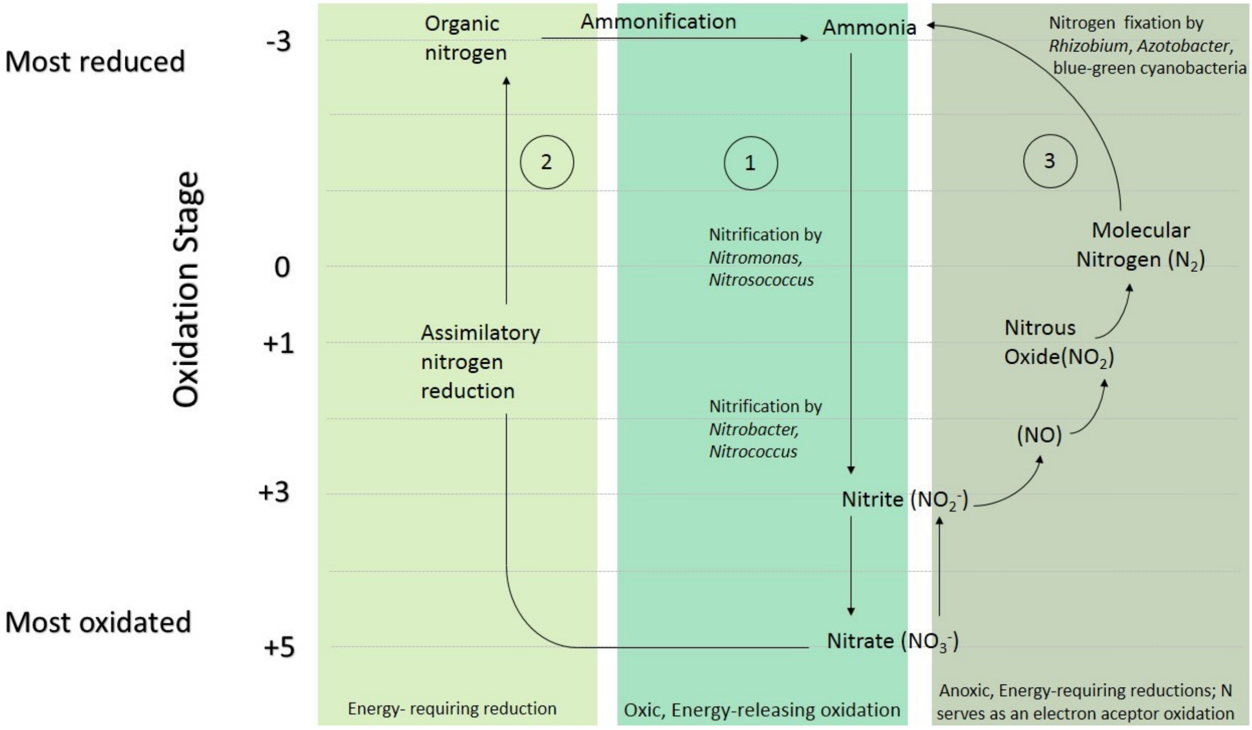

Figure 1 N-cycle (Adapted from "Ecologia course", Gatto M.; Casagrande R., Politecnico di Milano).

Over time, nitrogen is transferred from the atmosphere (N_2) to the biosphere. Thanks to natural fixation processes, atmospheric elemental nitrogen is turned into ammonium or nitrate ions, i.e., the only forms that are useful to plants. The ammonium taken by plants (a small fraction of that obtained from atmospheric nitrogen, though) is incorporated into many organic molecules by an assimilative reduction. These molecules are subsequently ingested by consumers of various orders. Nitrogen is then released to soil by means of the decomposition of plant debris and corpses of animals. Here, through the ammonification process, nitrogen returns to the form of ammonium ion. It is then transformed into nitrate by the nitrifying bacteria of the soil, and thus made available again for plants. If the soil is poor of oxygen, denitrifying bacteria can further transform nitrate into nitrite and, subsequently, nitrite into elemental nitrogen, which goes back in the atmosphere.

Nitrogen production is increasing every year. The demand comes dominantly from agricultural activities, followed by fossil fuel energy, and, more recently, bio-fuels (Galloway et al., 2008).

Impact of human activity on the N-cycle is unmistakable.

In areas with negligible human influence, nitrogen deposition to ecosystems generally amounts to about 0,5 kgN/ha/yr or less. Instead, there are now large regions of the world where average N deposition rates exceed 10 kg N/ha/yr, greater than an order of magnitude increase compared with natural rates. Being the main consumer of nitrogen, agriculture (whose productive system can be divided into three subcomponents, indicated in Figure 2, in the left-hand side dashed box) can alter the natural cycle of nitrogen.

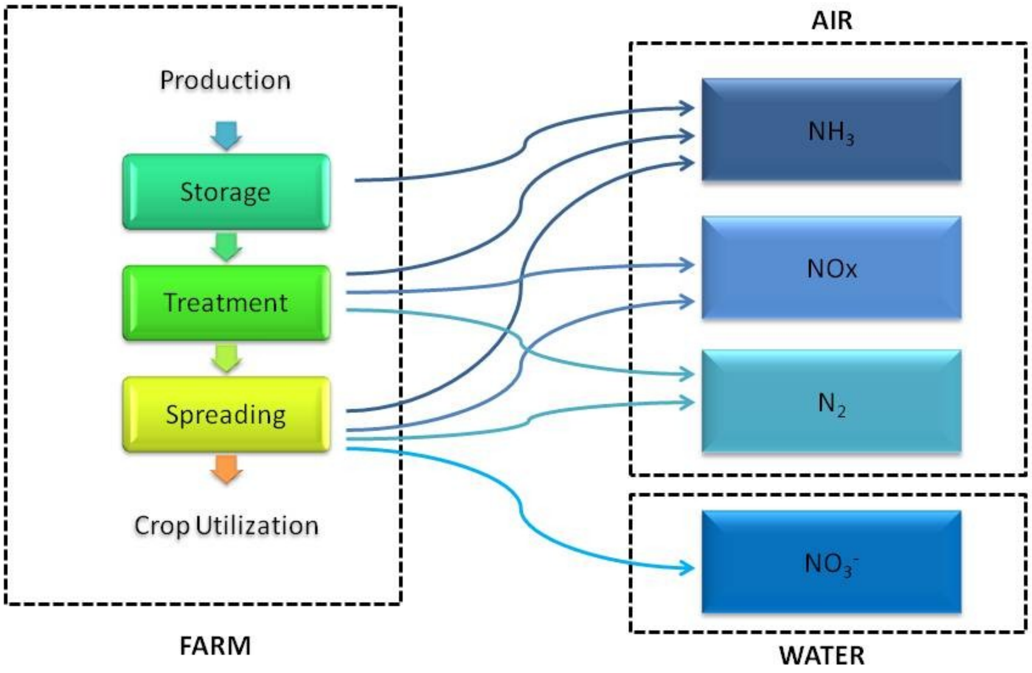

Figure 2 Major nitrogen losses during the management of livestock manure. In the left-hand side dashed box are reported different subcomponents of agriculture productive system. In the right-hand side are reported different pollutants for each environment compartment. Adapted from Burton et al., 2003.

The input of nitrogen to agriculture comes from the diet of animals, in particular from proteins contained in feed or grazed grassland. Only a part of the ingested nitrogen is metabolized by the animals, which consequently expel the excess N through urine and dung. In these residues, nitrogen is found both as urea and as other organic compounds (proteins and peptides).

The excretion of nitrogen is the input of the manure storage subsystem (Figure 2). The manure is excreted and subsequently transported to the manure storage areas where it comes into contact with the air. The naturally occurring enzyme urease degrades the urea into ammonia (NH_3), that, in the can volatilize. Volatilization is related to the amount of inorganic nitrogen in the slurry. All the housing conditions also have influence on the loss of ammonia, as well as the type of accumulation expected, the storage time, the size of the structures used for it etc.

The amount of nitrogen remained in manure when it is distributed to field is called N-manure. Applied spreading methods obviously influence the degree of volatilization that can occurs during and after manure application. Nitrogen not used by plants, called N-surplus, can be divided into N-leached, N-volatilized and N-denitrified. There is also a part that is run-off. The loss of nitrogen in the atmosphere that can occur in the first steps of the management of slurry represents a decrease in the potential benefit of proper use and recycling of nitrogen through crop production (Burton et al., 2003). Therefore, the correct management of livestock manure is necessary to limit the incorrect and uncontrolled nitrogen introduction in the environment (in its various impacting forms).

1.2. Environmental issues related to livestock manure

As reported by FAO organization, the livestock sector has an important role in climate change; in fact, it is estimated to emit 7,1 gigatones of carbon dioxide equivalent (CO_2-eq/year, representing 14,5 percent of all human-induced emissions). In particular, the milk and beef production have a biggest emission contribution (19% and 41%) rather than pig meat and poultry meat and eggs production that contribute respectively 9% and 8% to the sector's emissions (FAO, 2018).

In Europe agriculture sector contributes for 10,1% of greenhouse gases emissions (the first source is Energy with 78% of contribute) (European Environmental Agency, 2016).

In the year 2014, CH_4, N_2O and CO_2 emissions from agriculture sector were 23,3%, 34,7%, and 0,13% of total EU28+ISL CH_4, N_2O, and CO_2 emissions, respectively. Total emissions from agriculture were 436 Mt CO_2-eq with contributions from CH_4, N_2O, and CO_2 of 237 Mt CO_2-eq, 188 Mt CO_2-eq and 10,2 Mt CO_2-eq, respectively. Thus, CH_4, N_2O, and CO_2 contributed with 23%, 35% and 0,13% total EU28+ISL GHG emissions.

Agriculture can be considered as the most important source of anthropogenic CH_4 source.

CH_4 methane is estimated to have a global warming potential (GWP) of 28–36 over 100 years. The normal metabolic activities of ruminants produce significant amounts of CH_4. The production of CH_4 involves degradation and hydrolysis of organic material to organic compounds, which are then degraded to long-chain acids, proteins or alcohols (Perazzolo, 2015). The amount of CH_4 generated by a specific manure management system is affected by the extent of anaerobic conditions present, the temperature of the system, and the retention time of organic material in the system (Santonja, G. G., 2017).

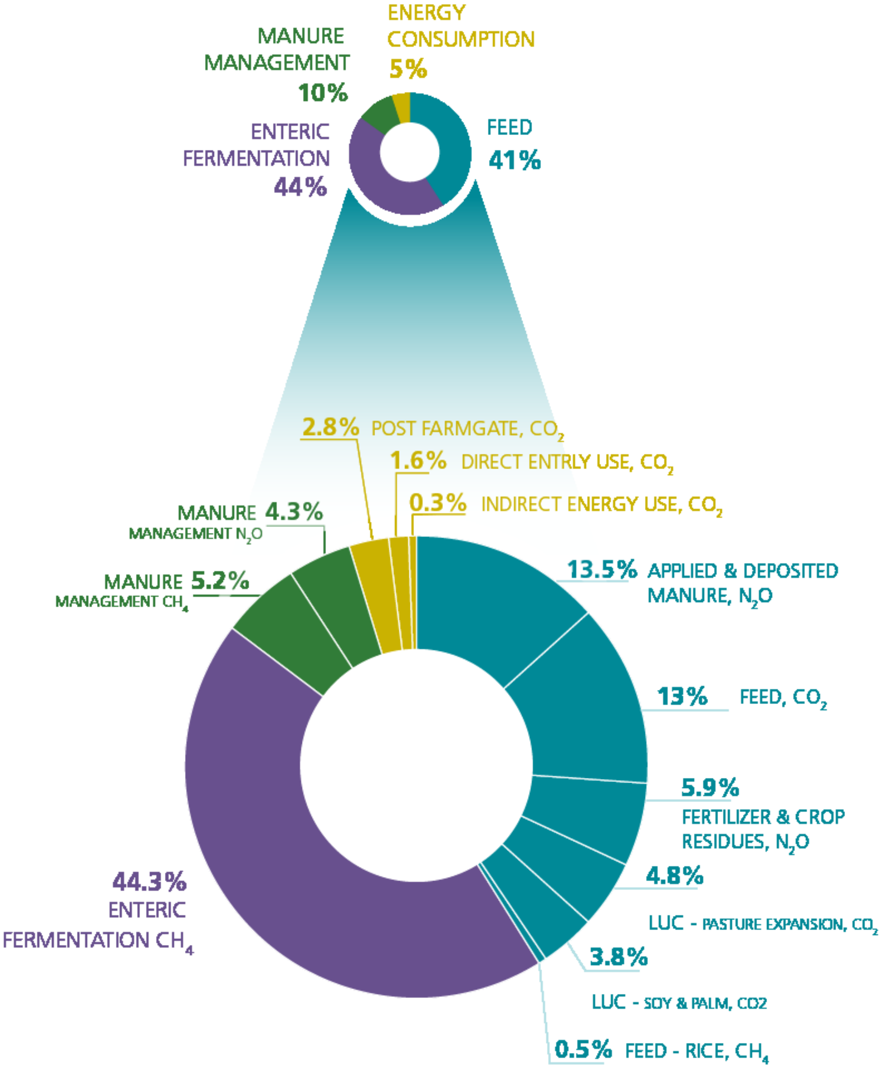

Figure 3 Global emissions by source. Relative contribution of main sources of emissions from global livestock supply chains. GLEAM 2.0 - Assessment of greenhouse gas emissions and mitigation potential. FAO, 2017

In addition, some manure treatment approaches can lead to rise of CH_4 emissions (Chadwick et al., 2011). Storage systems are also sources of CH_4 because the environment in these stores, if not actively aerated, favours methanogenesis or if it mostly stored over long period. Pig slurry generally has the potential to emit more CH_4 than cattle slurry, because it has a higher content of degradable organic matter than cattle slurry (Moller et al., 2004).

Nitrous Oxide (N_2O) has a GWP 265–298 times that of CO_2 for a 100-year timescale.
In nature, emissions of N_2O results also from a variety of agricultural practices and activities as the use of synthetic and organic fertilizers, the cultivation of organic soils, the production of nitrogen-fixing crops and from the application of livestock manure to cropland and pasture (Perazzolo, 2015).
Main sources of N_2O emissions are manure management and the application and deposition of manure (Montes et al., 2013).

Although not a GHG, NH_3 (and its ionized form, NH_4^+) is an important component of the environment because it is the dominant gaseous base specie in the atmosphere. Its properties (e.g., water solubility) make NH_3 important in atmospheric chemistry and physics. Once released into the atmosphere, NH_3 has a lifetime of less than 1 day to 5 days (Aneja et al., 2001). It will, therefore, most likely deposit to the Earth's surface closed to its source. However, part of NH_3 can react with acidic species such as sulphuric acid (H_2SO_4), nitric acid (HNO_3), and hydrochloric acid (HCl) to form ammonium sulfate, ammonium nitrate, or ammonium chloride, respectively. In Europe and the US approximately 75% of NH_3 emissions derive from livestock production (Webb et al., 2005). The N collected in manure is in reduced form such as ammonium (NH_4^+), proteins and urea. For these reasons, the livestock manure management is a strategic sector for the reduction of ammonia emissions

Concerning other environmental sector, is evident that the biggest problems of twenty-first century are related to water quantity and/or water quality issues (Schwarzenbach et al., 2010). Obviously, these problems are strictly connected with climate change that aggravates the already precarious situation of the quality, availability and accessibility of the water resource.

The incorrect livestock manure management is one of the main reasons for water pollution (diffuse source) and eutrophication. This is also due to an inappropriate and extensive use of some chemical products as fertilizers, pesticides or directly the erroneously use of manure, sludge and compost, both agronomic practices that promote the release of these in the environment (Mateo-Sagasta, J., et al., 2017,2018).

Leaching and runoff of manure nutrients applied to fields can reach surface water and groundwater with a possible consequent contamination (Figure 4).

In addition, animal diets can contain much higher trace metal concentrations than plants used for animal feeding, for responding to necessity of animal to take different essential nutrients elements. For example, Cu is added to pigs' diet as a cost-effective method of enhancing performance and as an anti-bacterial agent. In addition, Zinc is added to animal feed for weaned pigs for control of post-weaning scours (Sommer et al., 2013).

The risk to health caused by poor manure management includes different pathogens transfer pathways. These can be diffused in the environment through air, water, crops and vegetables. Pollution of shallow drinking water wells is a considerable risk to human and animal health. Therefore, the dumping of animal manure into rivers can pose a much greater risk of infection than careful recycling of manure in agricultural land with adequate precautions during spreading (Sommer et al., 2013).

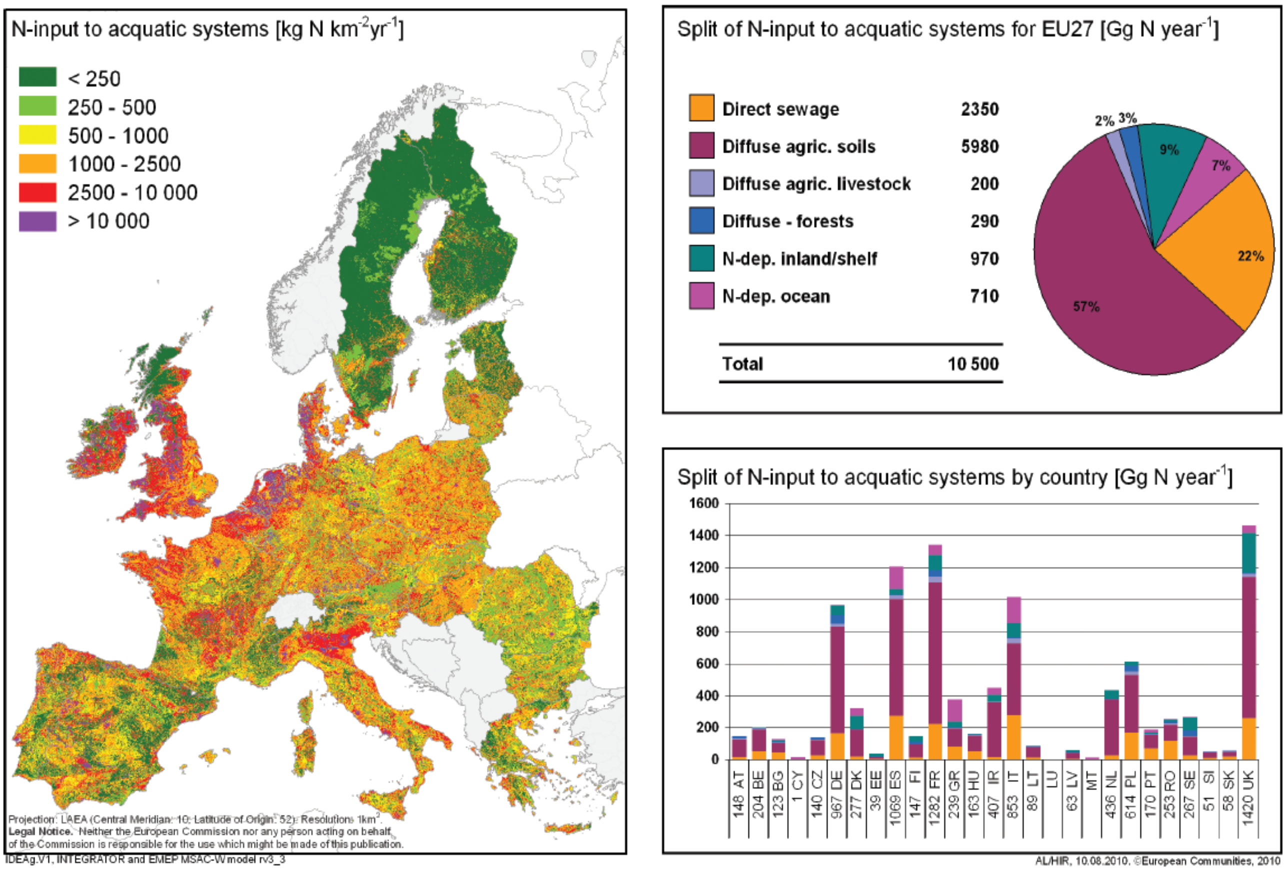

Figure 4 Total reactive N input to the hydrosphere (rivers and groundwater) in EU-27 for the year 2002. The map shows total N r point sources from sewerage systems and diffuse sources from agriculture and forest soils and atmospheric N r deposition to inland water surfaces for a grid at of 1 km × 1 km. The values are in kg N per total pixel area [kg N/km^2/yr. total area]. The pie diagram at the right side gives the split of N input to the hydrosphere [Gg N/year, rounded to 10 Gg N/year] for EU27: point sources (sewage systems) and diffuse sources (agriculture leaching, run-off, and forest soils). The histogram shows the split of N r input to the hydrosphere [Gg N/ year] by country. Basis: Sewage systems and agriculture: Indicator Database for European Agriculture V1, 2009; forest soils: INTEGRATOR, 2009. Leip et al., 2009.

Nowadays, also due to emerging problems of scarcity of resources, the recovery of nitrogen (N) and other elements (phosphorus, heavy metals etc.) is strategic for economic and environmental reasons considering that manure represents one of the most significant contributors to nitrogen (N) sources.

In Table 2 are reported categories of major water pollutants in agriculture and the relative contributions of the three main agricultural production systems.

Table 2 Categories of major water pollutants in agriculture and the relative contributions of the three main agricultural production systems (Mateo-Sagasta et al., 2017). *: low contribute; ** medium contribute; ***: high contribute.

Pollutant category	Indicators/examples	Relative contribution by: Crops	Livestock	Aquaculture
Nutrients	Primarily nitrogen and phosphorus in chemical and organic fertilizers as well as animal excreta and normally found in water as nitrate, ammonia or phosphate	***	***	*
Pesticides	Herbicides, insecticides, fungicides and bactericides, including organophosphates, carbamates, pyrethroids, organochlorine pesticides and others (many, such as DDT, are banned in most countries but are still	***	-	-
Salt	E.g. ions of sodium, chloride, potassium, magnesium, sulfate, calcium and bicarbonate. Measured in water, either directly as total dissolved solids or indirectly as electric conductivity	***	*	*
Sediment	Measured in water as total suspended solids or nephelometric turbidity units – especially from pond drainage during harvesting	***	***	*
Organic matter	Chemical or biochemical oxygen demanding substances (e.g. organic materials such as plant matter and livestock excreta), which use up dissolved oxygen in water when they degrade	*	***	**
Pathogens	Bacteria and pathogen indicators. E.g. Escherichia coli, total coliforms, faecal coliforms and enterococci	*	***	*
Metals	E.g. selenium, lead, copper, mercury, arsenic and manganese	*	*	*
Emerging pollutants	E.g. drug residues, hormones and feed additives	-	***	**

For these reasons, currently new-technological approaches are studied for valorisation and reuse of substances that in the "traditional" technologies are classified as potentially polluting and therefore removed. It is consequently evident the need of a correct management of livestock slurry, both for the protection of the environment, but, above all, for the valorisation of a product that turns out to be a precious resource.

1.3. Main legislation concerning the prevention of environmental pollution caused by the management of livestock manure

Environmental protection is nowadays a primary global interest issue.

Environmental problems related to the management of livestock manures are commonly the result of ignorance, negligence or not calculated risk. Therefore, as long as no preventive measures or measures of mitigation are taken, environmental problems will continue to exist (Sommer et al., 2013).

In response to the problems of society, where culture, markets and the pressure of civil society collectively fail to find a resolution, public or government policy can intervene to solve certain problems. Government policy aims to change the individual behaviour to achieve society goals. Among the available policy tools there are: regulatory or command and control instruments, economic or market-based instruments and communicative or persuasive tools. Multilateral environmental agreements (conventions and protocols) as well as intergovernmental organizations have largely contributed to consider seriously many of the known environmental problems.

The main European directives concerning the management of livestock manures are related to the protection of water and to the prevention and control of pollution, such as pollutants air emission.

1.3.1. European Directives

A good example of joint of regional and national regulation can be found in European Union. Environmental European policy is mostly established through Directives that imposed to member states environmental objectives to be achieved (Sommer et al., 2013). In general, the communitarian directives concerning management of livestock manure are related to protection, prevention and control of pollution, including air pollution resulting from greenhouse gas emissions and water pollution caused by over-nutrient content and by the presence of micro-pollutants. Here below some of most important directives are reported and briefly explained.

1.3.1.1. 2010/75/EC Directive on Industrial Emissions concerning Integrated Pollution Prevention and Control (IED).

This Directive has substituted the 2008/1/CE, which in turn had replaced Directive 96/61/EC. The upgrade integrates the Directive of 2008 with other 6 directives concerning the industrial emissions. It establishes a permit procedure and lays down requirements, in particular about emissions

The IED directive concerns industrial and agricultural activities with high polluting potential.

Introduces the innovative concept of the integrated approach to the prevention and reduction of emissions in the air, water and soil, waste management and energy efficiency. It provides for the gradual adoption of technical solutions available on the market (plant engineering, management and control), with the aim of preventing, reducing and, as much as possible, eliminating pollution due to industrial activities.

The objective of directive is to avoid or minimize polluting emissions in the atmosphere, water and soil, as well as waste from industrial and agricultural installations, with the aim of achieving a high level of environmental and health protection.

The industrial activities indicated in the IED Directive, to ensure the prevention and reduction of pollution, must operate only if they are in possession of an authorization.

To gain a permit operator must show that no significant pollution is caused and that the BATs (best available technologies) are applied, considering the technical characteristics, geographical locations and local environmental circumstances of the installations concerned.

"Best" means the most effective to achieve a high overall level of overall environmental protection.

"Available" means those techniques that have been developed on a scale that allows implementation in economically and technically viable conditions but does not necessarily mean that the technique must be an industry standard or actually widely available, since the IED's intention it is about changing practices to new, less polluting alternatives.

“Techniques” includes both the technology used how the installation is designed, built, maintained, operated and decommissioned. The EU member states' authorities and industries cooperate on the development of so-called “BAT” reference documents (BREFs). A BREF assists the regulatory authorities and those applying for licenses by describing reference techniques and reference levels for a specific economical sector. One of the BREFs is dedicated to installations for intensive livestock production (entitled 'Intensive Rearing of Poultry or Pigs’). This document covers intensive livestock farms with more than 40 000 places for poultry, or 2000 places for production pigs over 30 kg, or 750 places for sows. The BREF discusses good agricultural practice, nutritional management, housing systems, water and energy use, manure storage, manure processing, and land spreading of manure.

1.3.1.2. 2016/2284/EU National Emissions Ceiling (NEC) Directive.

The latest NEC ("National Emission Ceiling") directive of December 14, 2016 n. 2016/2284 / EU, introduced by the European Union concerning the reduction of national emissions of certain atmospheric pollutants, amends Directive 2003/35 / EC and repeals Directive 2001/81 / EC (1).

The new NEC Directive sets 2020 and 2030 emission reduction commitments for five main air pollutants: sulphur dioxide (SO_2), nitrogen oxides (NO_x), non-methane volatile organic compounds (NMVOC), ammonia (NH_3) and fine particulate matter (PM2,5).

Furthermore, it requires the development, adoption and implementation of national air pollution control programs and that the emissions of these pollutants and other pollutants listed in Annex I, as well as their impacts, are monitored and reported.

The main objective of this directive is to combat acidification, achieving long-term goals by taking the years 2010 and 2020 as a reference.

This Directive establishes the maximum limits for total emissions responsible for the acidification of these 5 pollutants for each Member State for emissions responsible for acidification, eutrophication and ground-level ozone pollution, but leaves Member States the freedom to adopt reduction measures. The directive is basically the EU translation of the Göteborg protocol.

Furthermore, Member States have to prepare and update national inventories of emissions and sulphur dioxide emissions, nitric oxide, volatile organic compounds and ammonia. These inventories and projections must be reported to the Commission and the European Environment Agency each year by 31 December.

1.3.1.3. 2000/60/EC Water Framework Directive

This Directive aims at maintaining and improving the aquatic environment in the Community.

The aim of directive is to protect superficial, deep, transaction, coastal waters. Defence and reduction of pollution, promotion of a sustainable water use, environment protection, improvement of aquatic ecosystems and mitigation of flood and drought effects.

1.3.1.4. 91/676/CEE Nitrates Directive

The EU Nitrates Directives now an integral part of the Water Framework Directive and is one of the key instruments for **protecting water** against agricultural pressures. Furthermore, it has also affected the control of greenhouse gas emissions.

The aim of this directive is “to reduce water pollution caused or induced by nitrates from agricultural sources and prevent further such pollution”.

The Nitrates Directive (91/676/EEC) requires Member States (MS) to:

- Monitor water quality and Identify polluted waters or water at risk of pollution (trophic status);
- Designate the vulnerable zones (NVZ);
- Establish codes of good agricultural practices (CGAP) (voluntary implementation outside NVZ/obligatory within NVZ) and action programmer;

- Every 4 years review the designation of vulnerable zones and action program. The measures concern the correct use of nitrogenous fertilizers in general and livestock manure.

  The definition of polluted or potentially threatened waters is attributed if nitrate concentrations in groundwater and surface waters could be higher than 50 mg NO_3^-/L if no action is taken, or if surface waters. Including fresh water bodies, estuaries, costal and marine waters, are found to eutrophic or in the near future may become eutrophic if no action is taken.

In order to reduce pollution, the action programs must comprehend obligatory measures connecting to:

- a period during which the spreading of animal manure and fermenters is prohibited;
- Capacity and construction of livestock effluent storage, including measures to prevent water pollution caused by flow and infiltration into ground and surface waters of liquids containing livestock manure and effluents from plant material such as silage fodder;
- Limits to the amounts of animal manure and fertilisers applied to land, which balanced fertilisation.

The Nitrate Directive has had an enormous impact on the management of effluents in the EU-27 countries, therefore in this area there is a more effective and advanced management of animal livestock compared to the one applied in the United States, Japan and China.

1.3.2. Italian regulations

1.3.2.1. D.Lgs. 152/2006 "*Testo Unico Ambientale*"

Numerous European directives have been implemented in this law, which repeals old decrees.
This act regulates the following subjects:

- in second part, the procedures for the strategic environmental assessment (SEA), for the environmental impact assessment (EIA) and for the integrated environmental authorization (IPPC);
- in the third part, the protection of the soil and the fight against desertification, the protection of water from pollution and the management of water resources;
- in the fourth part, the waste management and remediation of contaminated sites;
- in the fifth part, the protection of the air and the reduction of emissions into the atmosphere;
- In the sixth part, the compensation protection against damage to the environment.

Its primary objective the promotion of the levels of quality of human life, to be achieved through the preservation and improvement of environmental conditions and the prudent and rational use of natural resources. For this purpose, it affords reorganization, coordination and integration of the environment laws. In that decree the vulnerable zone introduce in Nitrate Directive are identified and the required criteria for agronomic use are given, delegating to the regions the laws actuation. Once the zones have been defined, the implementation of the action plan, which must follow the directions of the directive, is the responsibility of the region.

1.4. Existing treatments for nutrients reduction and removal from livestock manure

Many options and treatments can be used for the correct handling of livestock waste. Manure processing applied for improving manageability and utilization of livestock manure can be modulated according to the needs of the individual farms in order to respond to specific needs. It is not possible to identify univocal solution valid in all situations.

It is necessary to consider every single reality and the choice of treatment must be carefully evaluated.

The decision on which treatment has to be applied must include three aspects: environmental protection, economic feasibility and agronomic needs.
The first point regards the objectives of atmosphere emissions reduction (which includes NH_3, odours, GHG, etc.), the water protection (e.g. compliance with Nitrate Directive), the removal of pathogens, or the removal of xenobiotic compounds (emerging pollutants). *Economic applicability* regards the need to match environmental needs and expendable costs or the possibility of spending reduction. Finally, the need to apply treatment that balancing the quantity of nutrients remaining in the treated livestock manure with the crop requirements, for returning the organic matter and nutrients to land in a more controlled way and improving the stability and plant availability of nitrogen and phosphorus. A processing strategy can consist of a single process or a combination of various unitary processes.
In the Figure 5 are shown various alternative treatment lines, which foresee the use in sequence of different chemical, physical and biological treatments that are currently in use or that can be potentially applied. Not all the treatments have been reported. It has been chosen to represent the treatments that widespread applied, and some treatments that today represent a novelty in the field of waste manure treatment.
In some contexts, treatment is not strictly required, and the produced livestock waste is stored in a tank and used at the appropriate time. In this case, as evidenced by the literature, problems related with GHG emissions occur (in the Figure 5 “emission problems” are identified with an “E”). For example, the **storage** of solid manure has also been shown to be a source of CH_4, with losses from cattle heaps representing between 0,4 -9,7 % of the total C content of small heaps, end loss can be higher in larger field scale heaps (Chadwick et al., 2005).

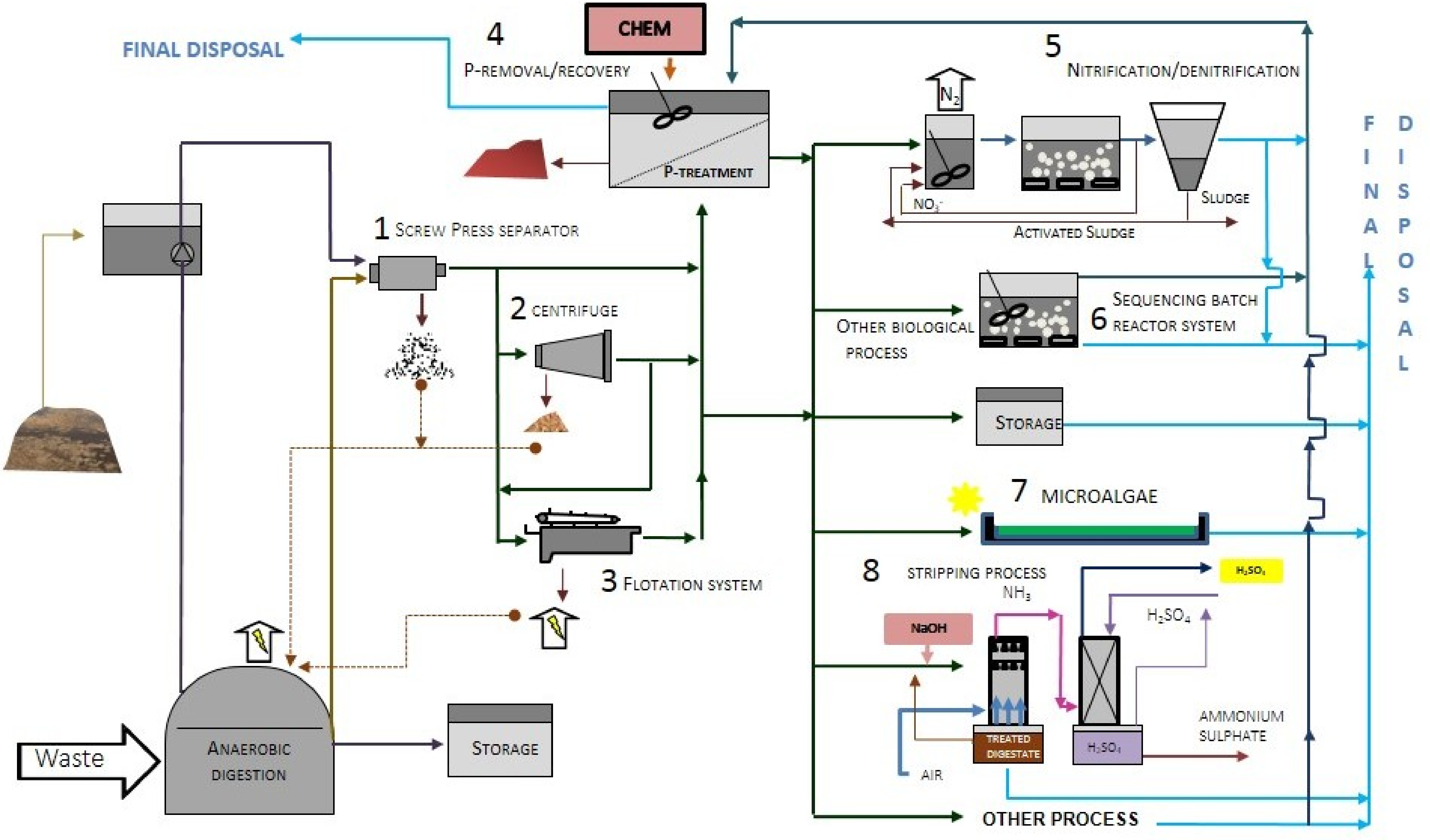

Figure 5 Different possible treatment lines for nutrients managing for livestock manure. 1: Separation through separator (e.g. vertical helical separator); 2: Centrifugation; 3: Flotation system; 4: Chemical process for P-precipitation; 5: Nitrification/denitrification process in a conventional scheme; 6: Sequencing batch reactor system; 7: treatment using microalgae; 8: stripping process. With "E" are identify the potential problems connected with emissions of GHG and ammonia.

In animal house with slurry collection, the manure remains in a predominantly anaerobic state with little opportunity for the NH_4^+ to be nitrified. Here N_2O may theoretically be produced at air liquid interface of stored slutty or on slats and solid floors where urine and faeces are deposited. In general, emissions of N_2O are a function of production and consumption of N_2O and the air exchange rate in the heap. Thus emissions of N_2O typically range from less than 1% to 4,3% of the total N in stored cattle and pig manure heaps. In addition, ammonia emissions can occur in a storage system.

Emissions from slurry stores may be reduced by decreasing or eliminating the airflow across the surface by installing a floating cover (different types), by allowing the formation of a surface crust, or by reducing the surface area per unit volume of the slurry store (Perazzolo, 2015).

For these reasons and obviously for those farms that do not have enough land on which they can spread the nitrogen load produced (Nitrate Directive), a further treatment is expected. There are different technologies based on chemical-physical or biological process finalized to the correctly livestock management or to the elimination of nitrogen compounds (and in general of nutrients).

1.4.1. Solid-liquid separation treatments

Among the chemical-physical processes, the **solid-liquid separation** is a frequently used treatment for managing livestock manure. This technique consists in the partial removal of suspended particles (both coarse and small) from the liquid manure.

The final goal of this treatment is to obtain two fractions: liquid one, characterized by a lower content of DM (dry matter), lower concentration of nutrient (P and N), heavy metals, and a solid fraction with a high concentration of DM (from about 15 to 30% depending on the technique used and the incoming material) and nutrients.

Due to their completely different composition and consistence, the two fractions can be used in different ways. The liquid fraction can be used directly on the crops, for fertigation, but it can be further treated through membrane filtration (Masse et al., 2007), evaporation (Veeken et al., 2004), crystallization and struvite precipitation (Hjorth et al., 2010).

With that kind of treatment, it is possible to reach the more suitable N/P ratio for the crops requirements and reduce P losses. Appling solid/liquid separation, the TAN /TN ratio (TAN: total ammonia nitrogen; TN: total nitrogen) in liquid fraction also increases, so more N is readily available for plants. Obviously, this treatment makes the liquid manure easier to handle, dropping also the consequent risks of blockages in pipelines (Møller et al., 2002) and reduces the cost and energy consumption.

The same benefit can be obtained by handling the solid fraction that, thanks to its texture, SF (solid fraction) can be transported with low costs and energy consumption. In addition, although the low quantity, after the S/L treatment solid fraction results rich in DM, phosphorus and organic matter (Møller et al., 2002). It may also be used for green energy production through anaerobic digestion, or can be used for composting treatment, as improver of soils, or as input material to produce mineral fertilizer.

In order to carry out solid liquid separation, there are many different options:

- filtration;
- drainage;
- centrifugation;
- sedimentation;
- flotation.

The combination of physical and mechanical separations can be applied for reaching highest separation efficiency. As example, in a Table 3 are reported results of separation efficiency, reachable with different types of separator.

Table 3 Separation efficiency reachable with different type of separator (Provolo et al., 2016).

		ST % Total Solid	SV % Volatile Solids	TN %	TAN %	P_2O_5 %	K_2O %	Volume (%)
Coarse Solids	Sieve	20÷25	25÷30	4÷7	5	8÷12	5÷10	5÷20
	Cylindrical	28÷48	32÷55	8÷15	5÷10	30÷42	5÷10	5÷20
	Helicoidal	30÷48	35÷45	10÷20	5÷10	28÷42	5÷10	5÷15
Small particles	Sedimentation	55÷60	70÷80	25÷35	20÷30	41÷57	-	15÷25
	Flotation	70÷90	80÷95	30÷40	15÷20	70÷90	-	20÷50
	Centrifuge	45÷70	60÷75	20÷40	16÷26	62÷80	5÷20	15÷30
	Belt press	42÷47	55÷62	24÷33	20÷30	60÷80	20÷30	20÷30

Specifically, the percentage represents the ratio between the amount of element in the solid fraction and that contained in the sewage entering the treatment obtainable with different types of separator. In literature, there are many works and research aimed at optimization of solid/liquid separation that, as explained before, allows livestock waste's volume reduction and optimized management of the fractions obtained. Separation is also performed to reduce the risk of phosphorus accumulation on agricultural land, after livestock manure disposal. The type of separation technique depends to the goal of treatment. When it is necessary to achieve the maximum level of pollutant's removal (ex. discharge into surface waters) many stages of separation are planned and different S/L separation processes in cascade are foreseen.

In the Figure 5, as example is reported a possible line of sequence of separation treatments that remove ever finer material at each step (1-2-3-4).

Nevertheless, even in these cases there is high risk of incurring high emissions of ammonia, (Loyon et al., 2007).

The solid fraction produced by the treatment has characteristics like untreated solid manure and has been shown to produce higher N_2O emissions during storage, compared to this latter due to the mixture of aerobic/anaerobic conditions on solid heap. The storage of the liquid fraction can lead to a lower N_2O emission than the untreated slurry. Regarding CH_4 emissions, it is difficult to say whether the separation of sewage increases or decreases it, since it depends mainly on the conditions of conservation and the characteristics of the obtained slurry fractions (Petersen et al., 2013). Likewise, the combined emissions of CH_4 and N_2O from storage of both fractions usually, but not always, were lower than those of the untreated fertilizer (Dinuccio et al., 2008). Additionally, slurry separation not always resulted in decreasing the NH_3 emissions. For example, when an emissions increment is observed, it is caused by the presence of a liquid fraction characterized by a higher TAN/TKN ratio and a lower TS% content, which slows the crust formation. This separation treatment is not enough for the objectives set (Nitrate directive), unless the separate solid obtained is exported. Therefore, often, the fractions obtained are sent to further dealings, which may include other S/L separation processes, or, more often, may involve other types of processes.

1.4.2. Other physico-chemical treatments

In order to handle livestock manure, other type of treatment can be applied, for example:

- membrane filtration,
- reverse osmosis,
- air stripping (in the Figure 5, n°8).

Membrane filtration and reverse osmosis are processes that can be defined as an “upgrade” of the simplest liquid solid separation process. The main difference is that, in this case, the separation occurs thanks to application of a driving force that allows the passage of the liquid through permeable membranes that retains the solid part. Based on membrane permeability, there are: microfiltration, ultra-filtration, nano-filtration and reverse osmosis. These treatments are often applied in cascade: in this way, micro-filtration is a pre-treatment for ultra-filtration, which is considered a pre-treatment for nano-filtration.

This type of process can be applied either when the usual solid / liquid separation treatments do not allow reaching the request solid removal rate necessary for subsequent treatments, or as separate process, maybe applied at the end of treatment line for removing the finest particles.

In addition, these processes allow producing a concentrated flow of nutrients that can be further use and which can be commercialized.

In the Table 4 the characteristics of different process are summarized, differentiated according to the applied pressure and a membrane cut off. This type of processes, however, have a series of operational problems related to the difficult management of filtering membranes that are very sensitive to wear and clogging phenomena (fouling).

Table 4 Characteristics of different type of filtrations (adapted from "Supply water treatment course". Nurizzo C., Politecnico di Milano)

	MF micro-filtration	UF ultra-filtration	NF Nano-filtration	RO reverse osmosis
Pressure	1 bar	2÷5 bar	5÷15 bar	>10 bar (variable)
Cut-off	≈ 50000 Da ≥ 500000 Da (for fine membrane)	2000÷80000 Da	200÷5000 Da	< 100 Da
Solid dimension retained	≥ 0,1 µ	≥ 0,003 µ	$\geq 10^{-3}$ µ	$\geq 10^{-4}$ µ
What holds?	Bacteria, organic waste, flocks' algae, protozoa; emulsion	low molecular weight organic molecules; proteins; bacteria; virus	Turbidity; colour; microbiological load; water hardness; emulsion	All organic molecules; ions; microbiological and viral load;

In fact, the presence of colloidal suspended solids, organic macromolecules, microorganisms as well as salt precipitation, the high-applied pressure and the high concentration of the incoming effluent can cause a change in the operating conditions due to the obstruction of the membrane pores. Even if the application of that kind of filtration allows reaching high level of removal rate, in the livestock treatment filed they are not widespread.

Changing type of process, the stripping process is a valid alterative for nutrient managing in livestock manure treatment. Simplifying process complexity, ammonia stripping is a physical process of removing a compound from liquid phase, using a gaseous carrier medium. It takes place based on the specific chemical-physical characteristics of the compound and on the elevation of the total contact surface of the liquid phase with the surrounding medium. This process, widely widespread in a chemical sector, it was adapted and applied to livestock treatment sector; it usually exploits use of high temperature and use of chemical products for rise pH in order to improve ammonia volatilization. Since that treatment is focused to removal N as ammonia, can be well applied to that kind of slurry that contains high concentration of TAN, as digestate, namely the product obtained downstream of the anaerobic degradation processes, which will be mentioned. With an optimized treatment, it is possible to obtain removal efficiency higher than 60% of TAN (considering only the stripping phase). Recently, possible simplification modifications of the "classic" industrial stripping process have been studied to best adapt it context in order to treat correctly livestock manure, allowing economic sustainability, as well as management and implementation simplicity (Provolo et al., 2017).

1.4.3. Biological treatments

As said before, in order to reduce nitrogen and nutrients in livestock manure biological treatments can be applied. These last include conventional nitrification and denitrification processes (NDN), simultaneous **nitrification-denitrification** process, **short-cut nitrogen removal** process, **anammox** process, enhanced biological phosphorus removal (**EBPR**), simultaneous removal of nitrogen and phosphorus. Nowadays there are also new approaches, for example based on the use of microalgae.

In conventional treatment plants (Figure 5, n° 5), biological removal of nitrogen occurs in two-step process, involving nitrification followed by denitrification process. Each reaction takes place through the biological activity of different bacteria. The final aim is that the nitrogen as ammonia in the slurry is converted in N_2 and re-enter the N-cycle. In this kind of treatment plants, there is neither the recovery of energy nor of matter. In fact, the last metabolic step envisages the release of nitrogen as N_2 which is naturally a gas. Leaving the slurry and entering again in the atmosphere the nitrogen released will be again available for the N-cycle. In this way nitrogen is somehow lost and wasted. The same consideration can be done for carbonaceous compound that are used in metabolic reactions and returns in the environment as CO_2.

Another inconvenient for these types of treatments are the high-energy consumptions and costs that often discourage the adoption of these plants by farmers. In addition, in these systems it is necessary to install suitable equipment and instrumentation, as well as to control continuously the process to ensure its correct operation: the mishandling of these plants can lead to the failure of prefixed objectives set and, indeed, to a waste of energy, money and resources.

In the recent years, mainly for economic aspects connected with the energy request for operation of NDN process, different kind of process have been developed, as a system for nitrogen removal based on partial nitritation with Anammox. This process does not require external carbon addition (necessary for post-denitrification treatment), has a negligible sludge production and need less energy and oxygen than the conventional process (Ahn 2006). A scheme reported in Figure 6 explains different nitrogen cycle re-classified according to potential biochemical pathway (Ahn 2006). During the biological processes, emissions can occur, mostly due to aeration of slurry during nitrification phase. In fact, the air blown into the slurry can primarily promote the stripping of ammonia, with the additional risk of producing N_2O emissions.

Another biological process widely applied to livestock waste is anaerobic digestion (AD) (Figure 5). This is not aimed at removing nutrients, rather at the energy recover from effluents, control of odours and stabilization of effluents before their agronomic reuse.

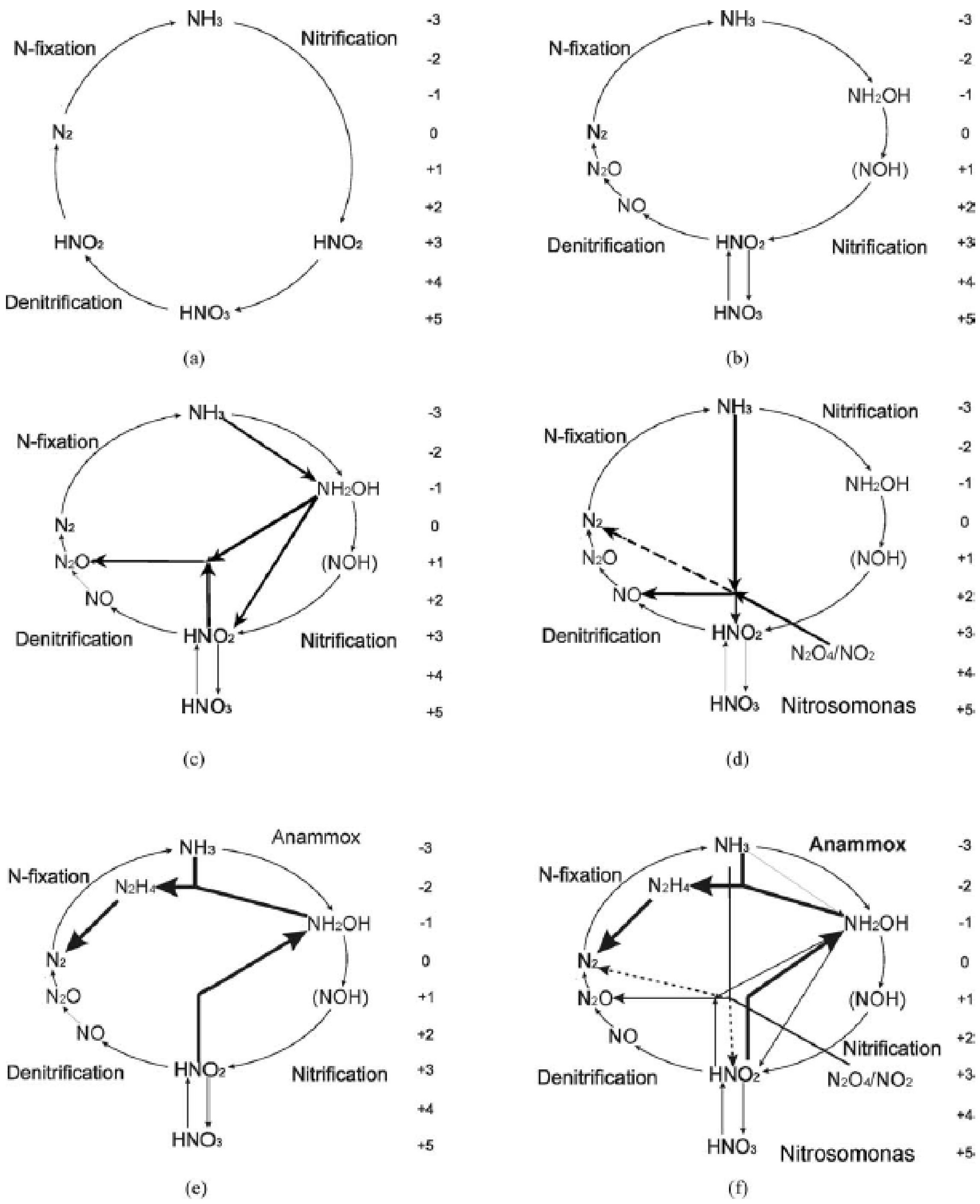

Figure 6. Nitrogen cycle (a) Classical N-cycle, (b) Sharon process, (c) Nitrosomonas aerobic denitrification or aerobic de-ammonification, (d) Nitrosomonas denitrification, (e) anaerobic ammonium oxidation and (f) overall nitrogen web. Ahn 2006

In particular, the overall objective of anaerobic digestion process is to convert organic waste into two categories of valuable products: biogas, a renewable fuel used further to produce green electricity, heat or vehicle fuel, and the digested substrate, commonly called digestate that can be used as fertilizer in agriculture. Digestate can undergo further treatments for the recovery of other products.

The process of anaerobic digestion can potentially increase the emission of NH_3 due to particular conditions that occur during the process as a higher concentration of TAN (Total Ammonium Nitrogen) and a higher pH. Besides the increase of the concentration of TAN, AD (anaerobic digestion) reduces the concentration of volatile fatty acids (VFA) and increases the concentration of total inorganic carbon (Sommer, 1997).

Another possible technology that can be applied for nitrogen removal and, in general, for nutrients reduction is the use of microalgae (Figure 5, n°8). In fact, piggery wastewater, as other livestock manure and digestate, containing high ammonia nitrogen and phosphorus as well as having a high COD content (considering untreated sludge). These pollutants, however, can serve as nutrients for the growth of some microalgae, which use N, phosphorus (P) etc. contained in the slurry for their growth, removing them from the effluent. Furthermore, microalgae produced can provide an additional by-product, which can be re-used as feed in aquaculture, plant fertilizer or intended to produce bio-fuels (Ayre et al., 2017)

Composting is another potential treatment that can be applied to livestock manure. It is a biological aerobic process that uses naturally occurring microorganisms to convert biodegradable organic matter in a product similar to humus. The process it destroys pathogens, converts nitrogen from the unstable ammonia to stable organic forms and reduces the volume of waste. Some limits of this process are connected with the increased risk of incurring emissions of ammonia, greenhouse gases including, especially methane and nitrous oxide.

Another applied treatment are wetlands and them can be natural or constructed wetlands (CWs). This method takes advantage of using of plants (macrophytes) and is managed as water quality improvement systems. The reduction of pollutants in wetlands is obtained by a combination of physical, chemical and biological processes: sedimentation, filtration, precipitation, absorption, absorption of plants, microbial decomposition and microbes nitrogen transformations; the application of constructed wetlands can treat a large quantity of livestock manure (Poach et al.2003; Meers et al., 2004).

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2. Motivation and goals of this dissertation

Currently, many information may be found concerning livestock manure treatment (see paragraph 1.3) although there is still the necessity to investigate deeper some aspects to improve the available best practices and eventually investigate new ones.

Despite the wide attention given to this theme, there are issues that, if solved, would allow achieving the desired results and therefore process optimization.

The major aim of this thesis was to investigate some issues regarding (1) nitrification and denitrification treatment, which still arise interest, as they are involved in the proper management of livestock manures.
In fact, in Italy, many nitrification/denitrification treatment plants were installed at farm facilities but very few showed a correct functioning. These malfunctions prevent farms to achieve the nitrogen removal goals, with furthermore potential risks in terms of emissions of ammonia and greenhouse gases.
In addition, due to rising interest in the new approaches finalized to nutrients recovery and not only to their removal, this work is also finalized (2) to study a possible application of solid/liquid treatment aimed to recovery of phosphorus, that is considered a scares resource.
As also say before, other theme (3) that rising worries and interest is the problem of ammonia emissions and greenhouse gases emissions. Emissions along the whole handling and treating process represent an undesirable problem that must be solved and contained, either with a correct mitigation solution or with an appropriate treatment. In this context for a correct emission evaluation, it is necessary to use a valid method to estimate the ammonia and greenhouse gases emissions. Therefore, the PhD research is also aimed to compare different instrument for measure emissions.

In particular, the dissertation is divided into different sections, each of which delves into a theme of interest in the aforementioned area (Figure 7).

Especially, first contextualization part (*Chapter 3*), is focused on the description and analysis's results of the state of the art of biological process treatment (*nitrification/denitrification process* mainly) applied in sequencing batch reactor systems. In particular, studying scientific literature on this theme, operational and management settings that guarantee excellent nutrient removal yields from livestock manure were identified, both on a laboratory scale and on a real scale. This research was done also for recognizing the possible control strategies useful for optimizing the achievable results. With the results obtained, the operative problems that occur at real scale and how they can be solved were inspected.

The followed part, instead, contains the experimental activity were carried out:

a) (*Chapter 4*) different monitoring campaigns of real scale treatment plants (both SBR and nitrification/denitrification “classic” plants) have been conducted and different experimental trials were been carried out. I*n particu*lar, in order to understand how the SBR plants in Lombardy work and what are the applied strategies of management of these systems, 5-treatment plants located in different provinces in Lombardy were monitored. All the analysis concerned this part were conducted at Università di Milano.
b) (*Chapter 5*) Always at Università di Milano, 6 lab-scaled SBR prototypes were designed and built, in order to study in a controlled environment, the biological process applied to livestock manure for nitrogen removal. In particular, 4 different experiment sets were performed in order to analyze how the removal rates changed with the variation of some operational parameters.

c) (*Chapter 6*) Different experiments concerning gas emission measurements were carried out during a PhD abroad period, that was conducted at IRTA (Institut de Recerca i Tecnologia Agroalimentàries), and in particular in collaboration with the GIRO group, (Organic Waste Integral Management Programme). Trails were performed in order to analyse the complex problem of NH_3 and GHGs emissions that can occur during different steps of livestock manure treatment chain. For these reasons, within the thesis’s research, the application of dynamic method, in particular the use of different type of dynamic hoods and static chamber for estimating GHGs and NH_3 emissions was investigated.

d) Then (*Chapter 7*), phosphorus recovery treatment was analysed. In particular, phosphorus precipitation/flocculation trials were carried out using different chemical compounds before and after a simplified ammonia stripping process. The aim is to analyse how phosphorus precipitation process can advantage following treatment and how phosphorus can be recovered. These experiments were conducted within the ReNeWal project. All the analysis concerned this part were conducted at Università di Milano.

e) Finally (Chapter 8), considering main results of different research that were carried out, a general pointing about the correct development of biological livestock treatment process finalizing to reduce ammonia emission and optimize reachable nitrogen removal rate was given. In particular a treatment line for pig manure was proposed. It foresees 2 solid liquid/separation processes, an SBR system and an alternative separation treatment focused on phosphorus recovery.

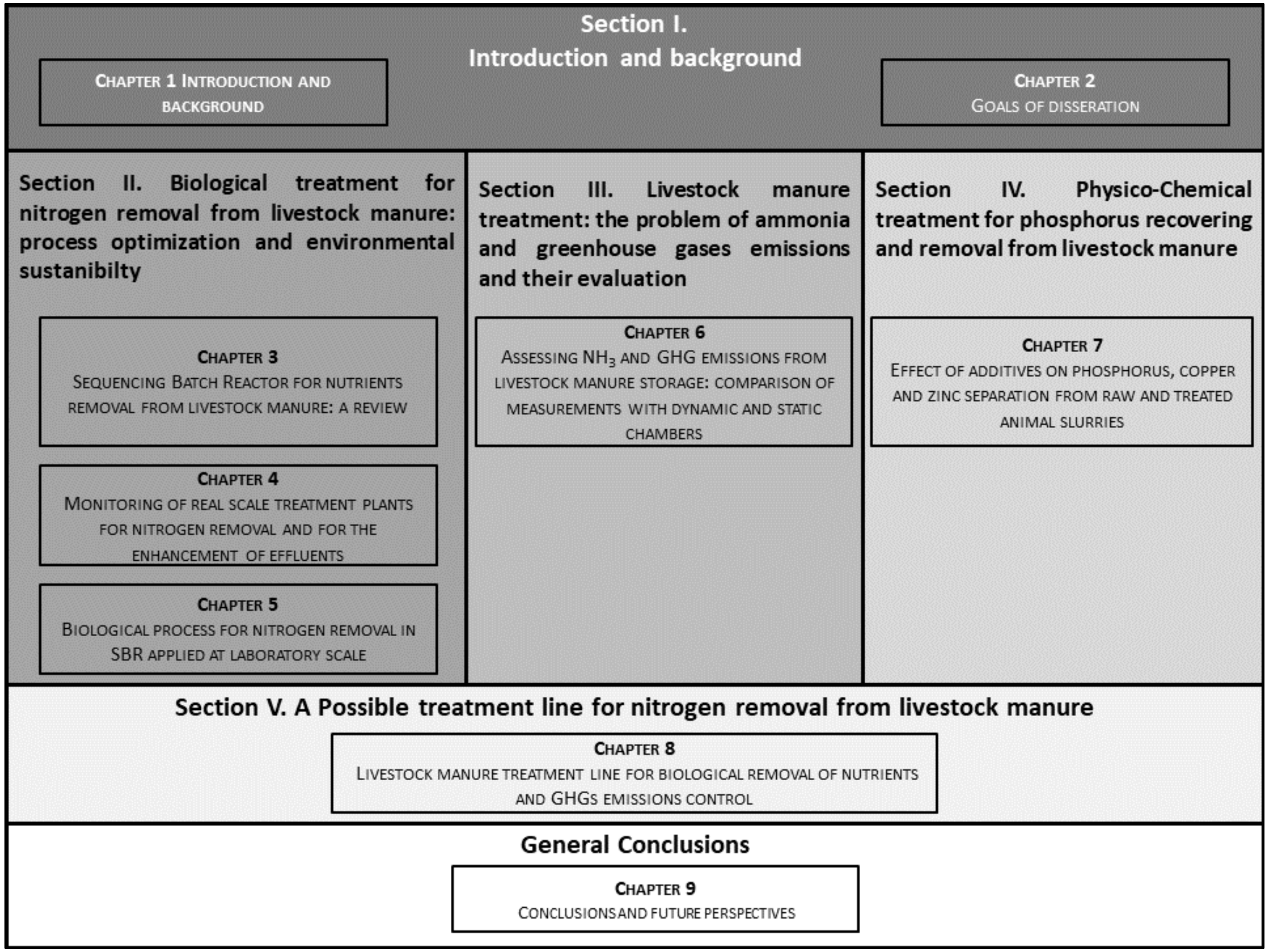

Figure 7 Organization and structure of the dissertation.

SECTION 2 - BIOLOGICAL TREATMENT FOR NITROGEN REMOVAL FROM LIVESTOCK MANURE: PROCESS OPTIMIZATION AND ENVIRONMENTAL SUSTAINABILITY

3. Sequencing Batch Reactor for nutrients removal from livestock manure: a review

The content of this chapter will be used for preparing an article Review that will be submitted.

3.1. Introduction

Intensive farming practices and their continuous spread generate a large amount of biological waste, which need proper management in order to avoid a negative impact on the environment (Bernal et al., 2009).

In order to maintain a sustainable local and global environmental balance and for securing a renewable source of nitrogen and phosphorus, is necessary to remove and recover nutrients from agricultural runoffs and animal manure, which are characterized by high load of nitrogen and phosphorus.

When livestock manure are applied to the soil, the excess of nutrients contents, besides other kind of micro-pollutants or heavy metals, through runoff and leaching, could contaminate surface watercourses and potentially led to arouse serious groundwater contamination or eutrophication of surface water (Han, Wu, Zhu, & Chen, 2008).

This nutrients imbalance between the crop requirements and livestock nutrients contents forces to find a viable alternatives for the direct use of manure as fertilizer (Deng et al., 2008; Magrí & Flotats, 2008).

The problem concerning nitrogen and nutrient pollution connected with the livestock manure utilization changes a lot considering different country, farms, restrictions concerning the quality of discharged effluents, way of breeding, etc.. In EU regions, for example, the limit imposed by the European directive “Nitrates Directive” for nitrogen utilization on land is 170 kg N/ha/y in Nitrate Vulnerable Zones –NVZ, and in several regions, there is a significant nitrogen surplus (European Commission, 1991). Accordingly, that the correct management of livestock manure production appears as a necessity, with attention to nitrogen and phosphorus contents.

Among the different possibility to reduce the nitrogen load, such as the transport of nutrient surplus to other areas, the introduction of a treatment for removing nutrients excess might be a suitable solution.

There are different technologies finalized to the elimination of nitrogen compounds based on chemical-physical processes (reverse osmosis, air stripping, membrane filtration, ion exchange, and breakpoint chlorination) or biological processes. These last include nitrification and denitrification processes (NDN), simultaneous nitrification-denitrification process, short-cut nitrogen removal process, anammox process, enhanced biological phosphorus removal (EBPR), simultaneous removal of nitrogen and phosphorus. Nevertheless, NDN process generally provides the most economical and efficient means for controlling nitrogen in wastewater effluents, transferring nitrogen to the atmosphere as N_2 (Azhdarpoor et al., 2015; Otawa et al., 2006). For example, up to 60% of total nitrogen initially present in pig slurry can be remove with NDN (Magrí & Flotats, 2008).

The nitrification/denitrification process applied in the sequencing batch reactor (SBR), currently, is widely used process for wastewater treatment of domestic and industrial wastewater from a variety of industrial categories.

Initially, SBR systems were applied to manure for removing only organic compounds aerobically, but that requirement it was soon transformed into the will and the need to remove also nutrients (Bortone C.H., 1992). Subsequently, the sequencing batch reactor is soon extended to remove nitrogen and phosphorus by applying intermittent aeration. With this goal, different process (suspended versus retained biomass) and configurations (single reactors versus series of reactors) were tested (Magrí & Flotats, 2008). The studies on SBR systems were then moved on the process layout: different kinds of reactors and configuration have been tested.

The latest scientific researchers are still focused on these issues, but mostly of the recent ones are concentrated on the process optimization applied for ensuring appropriate removal rates and guarantee energy and money saving. In fact, the great success and development of SBR system in many sectors is due not only to its design flexibility, but also to the development of modern technology, which allows better process control.

The global attention on SBR technology is also attested by the real allocation of SBR WWTPs (wastewater treatment plants), which processing industrial and civil wastewater, operating successfully in Japan, in the United States, Australia and in China, where new constructed wastewater plants have adopted the SBR process (Yang et al., 2010). This technique has been widely applied also to pig slurries and co-digested slurries (Choi et al., 2004; Deng et al., 2007; Deng et al., 2008; Lo & Liao, 2007; Magrí & Flotats, 2008; Poo et al., 2005).

Despite other type of biological processes (for example anammox process) currently succeed to the NDN applied to SBR technology, there is still much to be investigated on the process optimization and on process optimization tools easy to use, especially for the treatment of livestock manure. In fact, the application to of biological nitrogen removal to livestock slurry was considered as BAT (best available technology) and also for this reason, a lot of real scale farms, have applied this kind of treatment. Consequently, although the new plants will probably be built by applying other technologies, the existing ones need to function at their best.

In addition, although the NDN process operated in SBR has been widely studied, modelled and optimized especially in the field of urban wastewater, it is necessary to do further investigation on the application and optimization of this kind of system applied to livestock manure treatment and to the possibility of adapting this technology to more and more stringent limits and emerging issues in the field of new pollutants.

In this review, numerous studies on SBR systems used for biological nutrient removal from livestock manure are analysed. The aim of this work, besides to produce an overview on the SBR technology and SBR monitoring parameters and process optimization, is to individuate its strengths and weaknesses of when is applied and used for treating livestock manure. This study can be useful also to understand how improve ad optimize nitrification/denitrification process carried out in real scale SBR systems present on the territory that are been built for livestock slurry treatment and which do not always reach the intended target

3.2. Sequencing Batch Reactor: general aspects

The Sequencing Batch Reactor is a system where biological degradation of organic matter, nitrogen and phosphorus occurs in batch instead that in stationary continuous flow condition. In these systems, as well as for the continuous activated sludge process, the reduction of nutrients from wastewater and in general from effluents, can be obtained through microorganism activity. That solution represents one of the possible process tooled in an SBR system (Flores-Tlacuahuac & Pedraza-Segura, 2016).

Differently from a common activated sludge process, in the SBR the reactions occur in a single tank (or multiple, if is foreseen more lines) and the different biological processes are engineered based on temporal distribution rather than spatial distribution (Dutta & Sarkar, 2015). In this way, the duration of each phase can be adjusted in order to meet the different treatment needs, such as low COD (Chemical oxygen demand) in the effluent.

SBR systems can be defined as unsteady-state activated sludge process (Flores-Tlacuahuac & Pedraza-Segura, 2016).

SBR operates with biological nitrogen removal is an activated sludge system operating with a fill and draw sequence (cycle) that is repeated over time. Each cycle is composed of various consecutive phases (fill, reaction phase-anoxic/oxic reaction-, settle, draw and idle) that may overlap depending on the treatment goal (Han et al., 2008; Magrí & Flotats, 2008; Yang et al., 2010) (Figure 8).

The cyclical trend of SBR operation, based on nutrient removal, confers to this system flexibility for treating effluent of various kind, including the liquid fraction of pig slurry (Magrí & Flotats, 2008). The time of each cycle that spans the duration between beginning of fill and end of idle phase for a single tank SBR system, is essential for the proper optimization of SBR system; to this parameter is connected the number of cycle and duration of different phases (filling phase, reaction phase, settling phase, draw phase and idle phase).

During the ***Fill phase***, raw wastewater enters the tank and is mixed with the active biomass, which remains inside the tank at the end of previous cycle.

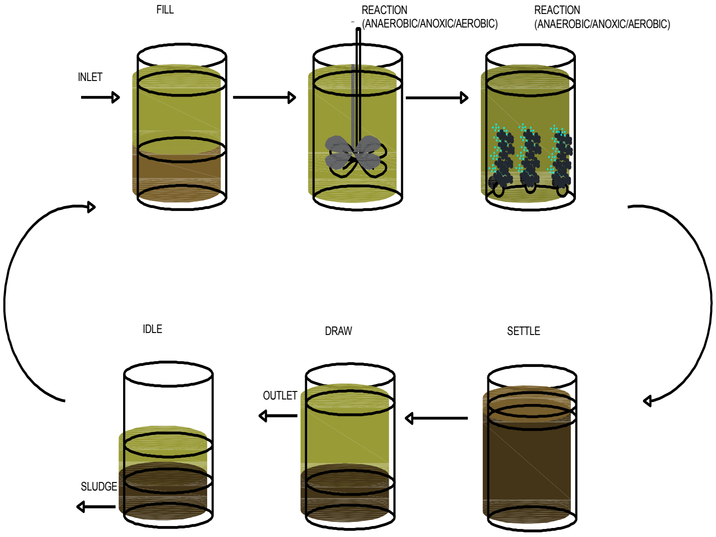

Figure 8 Schematic configuration of SBR operation.

This phase can be accomplished in different way, in a single time or in a multiple phase (for example as in the fed step strategy). In particular, depending on the wastewater characteristics and the target nutrient removal it is possible to define the filling phase as:

- Static fill: influent wastewater is added without mixing; resembling almost a plug flow situation creating a high food to microorganisms (F/M) ratio, similar to a selector compartment used in an activated sludge process, and in that way promote the growth of floc-forming bacteria by suppressing the filamentous ones (Dutta et el., 2015). In addition, due to high F/M ratio it is possible to select that type of fill when a biological phosphorus removal process is foreseen. This kind of filling can be useful if is denitrification phase occurs immediately after the feeding.

- Mixed fill: influent wastewater and organics is mixing with the biomass, which initiates biological reactions (EPA, 1999). During mixed fill, bacteria biologically degrade the organics and use residual oxygen or alternative electron acceptors. In this environment, denitrification may occur.
- Aerated filling: this type of filling is characterized by the simultaneous aeration of the sewage and the entry of new sewage. In this way, the aerobic reactions that will be completed in the Reaction phase are initialized. This type of filling can reduce the air requirement of the reaction phase.

During ***React phase***, the core of SBR operation, the completion of the biological reactions responsible primarily for the degradation of organic compound and nutrients occur. The treatment is controlled by air that is on and off for producing anaerobic, anoxic, or aerobic conditions. Obviously, the react phase operation depend on the requirement and on the type of biological process applied.

During the ***Settle phase***, the reactor tank acts as a batch clarifier, without any inflow or outflow. This phase is aimed to separate the flock of activated sludge to the clarified effluents, that will be discharge. This phase usually lasts 0,75-1 h.

In ***Draw phase***, the treated supernatant is withdrawn. Is necessary to leave in the thank over the active settled sludge a portion of supernatant.

Idle phase is the pause between the draw and the following fill phase. The introduction of this phase can be useful, for example, when there are several parallel working reactors and it serves as buffer in time. During this phase, mixing of the biomass to condition the reactive contents, and wasting of excess sludge, may be taken up, depending on the operating strategy (EPA, 1999).

In order to optimize the duration of the single phase for each case, a proper design of the time cycle is necessary. The permanence of the biomass in the system depends on SRT (Sludge Retention Time), consequently the efficiency of removal of nutrients. Generally, too high SRT lead to an inert environment, with accumulation of non-activated sludge and lower SRT mean that the biomass that has to slow growth times (such as nitrifying bacteria) is washed away. Various advantages are offered by SBR systems where nitrification and denitrification process is carried out, as the elimination of settling tank, the less space requirements and the possibility of modifications of process setting during the trial phases thanks to the great compatibility of SBR to automation (Artan et al., 2003).

3.2.1. SBR main sizing project parameters

For the correct design of any treatment plant, the first thing to be defined is the characterization of the inlet flow that have to be treated and the second one is defined the effluent’s characteristics that are being achieved.

Sequencing batch reactor systems allow infinite combinations of sequences aimed at achieving optimal operation based on the characteristics of the slurry that will be treated.

During operation, some phases are always present as settling and withdrawal; other phases, such as filling and reaction phases can be present or one phase can exclude the other.

Here below are reported, the main parameters generally necessary for sizing correctly of an SBR system, and which characterize its design and project (Sirini et al., 2006).

- Time of Cycle Tc, spans the duration between beginning of Fill and end of Idle phase for a single tank SBR system. Represent the duration of each cycle:

$$\boldsymbol{T_C = \frac{24}{n}} \ ; [\boldsymbol{h}]$$ Equation 1

where *n* is the number of cycles [cycle/day]; Tc can be expressed also as *1/n* as "fraction for day".

- Time of cycle can be also defined as follow:

$$\boldsymbol{T_C = T_F + T_R + T_S + T_D + T_I} \ ; \ [\boldsymbol{d}]$$ Equation 2

where T_F is Filling Phase, T_R is Reaction Phase, T_S is Settling Phase, T_D is Draw Phase and T_I is the Idle Phase.

The duration of ***Fill*** and ***React phases*** can be adjusted to impart the SBR system a CSTR-like or ideal PFR-like treatment characteristics (Dutta and Sarkar, 2015).

- V_F, volume feed in each cycle:

$$\boldsymbol{V_F = \frac{Q}{n}} \ ; [\boldsymbol{\frac{m^3}{d * cycle}}]$$ Equation 3

where Q is daily flow, feed in SBR reactor.

The overall volume of the reactor consists in two volumetric fractions: V_0 and V_F, where V_0 represents the volume where remains the settled sludge, and volumetric fraction V_F, the volume feeds and discharges in each cycle of operation.

- Θ, HRT, hydraulic retention time:

$$\boldsymbol{\theta = \frac{V_T}{Q}} ; [\boldsymbol{d}]$$ Equation 4

where $V_T = V_F + V_0$, and $Q = n * V_F$; the equation 4 becomes:

$$\boldsymbol{\theta = \frac{V_F + V_0}{n * V_F} = T_C * (1 + \frac{V_0}{V_F})}$$ Equation 5

If V_0/V_F is defined as *recycle ratio R*, Θ can be defined as follow:

$$\boldsymbol{\theta = T_C * (1 + R)}$$ Equation 6

V_F/V_T can be also defined as volumetric exchange ratio; consequently, the hydraulic retention time, can be expresses as follow:

Equation 7

$$\boldsymbol{\theta = \frac{V_T}{(n * V_F)} = T_C * VER}$$

VER allows to identify easily the volume percentage which will be treated.

- ***FTR*** -fill time ratio-:

Equation 8

$$\boldsymbol{FTR} = \left(\frac{\boldsymbol{T_F}}{\boldsymbol{T_C}}\right)[-]$$

- ***Sludge Retention Time*** SRT, is defined as follow:

Equation 9

$$\boldsymbol{SRT = \frac{V_T * x}{q_s * x_r + (q - q_s) * x_e}} \, [\boldsymbol{d}]$$

where *x* is concentration of biological mass in the reactor; x_r is biological mass in the sludge and x_e the biological mass in the effluent; q_s is the purged flow of sludge.

- ***Sludge Retention Time*** for autotrophic biomass SRT_A, is defined as follow:

Equation 10

$$\boldsymbol{SRT_A = SRT * \frac{TA_e}{T_C}} \, [\boldsymbol{d}]$$

where TA_e represents the duration of aerobic phase, which one involved in the growth of autotrophic biomass. Usually, a SBR cycle can last 6-8 h (4-3 cycles/day) (Obaja et al., 2003, 2005) but its duration can reach 24 hours. A low cycle's time needs higher volume; too many cycles for day lead to an incomplete reaction phase: in fact, if sedimentation time, extraction (withdrawn), have fixed time, the reaction time have to be reduce for reducing cycle length (Masotti 2011). Obaja et al., 2005, support that the time cycle and the duration of each single phase within the time cycle, must be designed properly in order to optimize the removal of nutrients, arguing that the choice of hydraulic residence time (HRT) and solid retention time (SRT) will depend of this optimization.

3.3. Nitrogen biological removal applied in SBR

In the livestock manure, as for civil wastewater, nitrogen is mainly present in organic form (proteins) and urea contained in the urine; in both cases, these substances undergo to a rapid process of ammonification that lead to ammonia nitrogen formation. Among different treatments, nitrogen biological removal can be achieved through nitrification/denitrification process that transforms ammonia present in the slurry to nitrogen gas.

Specifically, ***nitrification*** is a *two-step process* that is catalysed by aerobic chemoautotrophic AOB (Ammonia-Oxidizing Bacteria) and nitrite-oxidizing bacteria (NOB). In the first step, knows named ***nitritation***, according to reactions 11a and 11b AOB, often *Nitromonas spp*, through aerobic autotrophic process, oxide ammonium NH_4^+ to nitrite NO_2^-. During this step approximately 75% of the oxygen needed to complete the oxidation of ammonium is used and the maximum rate of nitrification has been observed to occur at a DO (dissolved oxygen) level greater than 2,0 mg/L (Dutta & Sarkar, 2015).

In the second phase, ***nitratation*** process, NOB bacteria, often *Nitrobacterspp*, oxide nitrite to nitrate NO_3^-, according to reactions 11c).

The complete reaction is reported in equation 11d.

$$NH_4{}^+ + 0.5O_2 \rightarrow NH_2OH + H^+ \qquad a)$$

$$\mathrm{NH_2OH} + O_2 \rightarrow N0_2{}^- + H_2O + H^+ \qquad b) \qquad \text{Equation 11}$$

$$NO_2{}^- + 0.5O_2 \rightarrow N0_3{}^- \qquad c)$$

$$NH_4{}^+ + 2O_2 \rightarrow N0_3{}^- + 2H^+ + H_2O \qquad d)$$

The nitritation phase is empirically the rate-limiting step in the nitrification process, and AOB therefore play an important role in ammonia removal in wastewater treatment processes. In a properly functioning plant, oxidation rate of ammonia to nitrite is lower than the oxidation rate of nitrate to nitrites. For this reason, it is very difficult for nitrites to accumulate inside the nitrification tank.

Assuming a cellular composition of the bacteria equal to $C_5H_7O_2N$ and considering the corresponding cell growth rates, the nitrification process can be globally represented in the following terms which express the overall weight ratios for the oxidation and synthesis reactions (equation 12).

$$NH_4{}^+ + 1{,}86\, O_2 + 0{,}1\, CO_2 \rightarrow 0{,}02\, C_5H_7O_2N + 0{,}98\, N0_3{}^- + 1{,}98H^+ + H_2O \qquad \text{Equation 12}$$

from this formulation it is possible to observe that: (1) the synthesis of nitrifying bacteria is very limited compared to the quantity of oxidized ammonia (0,16 g of new cells for each g of oxidized ammonia nitrogen); (2) that alkalinity is required in the ammonia-nitrate oxidation process is equal to 7,14 mg $CaCO_3$ to 1 mg of N-NH_4^+ and that (3) the oxygen consumption for respiration is very high (4,2 g$CaCO_3$ for 1 g of 1 mg of N-NH_4^+ oxidized).

If the concentration of oxygen is too low (<0,5 mg /L), the nitrification process can be inhibited, with a greater effect on oxidizing nitrite bacteria rather than ammonium oxidants.

As also shown before, the nitrification phase implies proton formation and, in consequence, the potential pH decrease, this depends on buffer capacity of wastewater or slurries. The rate of nitrification drops to 50% of the optimum if the pH rises over 9,8 and goes under 7,5 (Kim et al., 2004).

Temperature conditions affect the values of the equilibrium and solubility constants, and kinetics. The influence of temperature on nitrification process can be expressed by Van't Hoff Arrhenius equation.

Nitrate generated during nitritation are reduced to molecular nitrogen (N_2) that leaves the aqueous phase (schematic formula is reported here below (Equation 13), considering as source of C, waste or wastewater) by ***denitrification process***, which involves the anaerobic heterotrophic bacteria (more common are *Pseudomonas*). Denitrification bacteria are both autotrophic and heterotrophic. Most of them are chemio-heterotrophic, and take energy from chemical reactions and use the organic compounds as an electron donor and as a carbon source for cellular synthesis; as electron acceptor, under anoxic or anaerobic conditions, they use nitrates and produce N_2 as refused gaseous catabolite.

In equation 13 is reported reduction equation using as electron donor the internal carbon (yet present in the wastewater/slurry).

$$10NO_3^- + C_{10}H_{19}O_3N \rightarrow 5N_2 + 10CO_2 + NH_3 + 3H_2O + 10\,OH^- \qquad \text{Equation 13}$$

Alternatively, it is possible to add external source of readily biodegradable organic compounds (methanol or acetic acid).

Denitrification therefore leads to the simultaneous elimination of degradable organic matter and nitrates. Denitrification kinetics considered as zero order reaction if the concentration of organic substrate is much higher than the constant of semi-saturation. So, when the carbon is present in excess of the theoretical demand for nitrate removal, the process is independent of both substrate and nitrate concentration.

Denitrification process is effective at anoxic conditions when DO level is below 0,5 mg/l: if DO concentration is higher than 1,0 mg/l, the denitrification process is inhibited. For this process, the optimum pH is between 6,5 and 9 (Dutta & Sarkar, 2015). The two main inhibitors of the nitrification process is nitrous acid HNO_2 and ammonia NH_3, the first more than ammonium (Yan et al., 2013).

3.4. Characterization of raw animal slurries

As also said before (Paragraph 3.2.1), the characteristics of inlet slurry is the first thing to consider for a correct design of SBR operation. Livestock manure have different chemical physical characteristics than civil wastewater. For example, are characterized by concentration of ammonia nitrogen ten times higher and chemical oxygen demand varies from 2 to 51 g/L. Therefore, it is necessary to consider these types of waste as something different in comparison with domestic effluents, resulting in some differentiation in modelling of biological process that are involved.

This can lead also to the need to set SBR operation differently.

IN fact, the physical and chemical characteristics of livestock manure have a key role in their management. Actually, manure characteristics definition is crucial to define, at first the treatment purpose, and then for the proper process definition and application.

It is very difficult to generalize the characteristics of livestock manure, because, as known, animal slurry is composed not only of mixture of urine and faeces, but also of bedding materials and process water. Moreover, animal manure characteristics also depend on the different animals considered (i.e. pigs, cattle, poultry etc.) and for the same animal, those change with countries, farms, alimentation etc. (Boursier et al., 2005; Hjorth et al., 2010).

However, for some parameters, it is possible to do several generalizations, which enables an understanding of the main biochemical processes that will be involved.

Considering biological process, for its proper operation, the pH value must be considered; in fact, the optimal range for bacteria metabolism ranges between 6,5-7,5. Animal manure is buffered to pH 7 – 8,4 (Sommer et al., 2013).

Focusing on activated sludge process, the wastewater/effluent characteristics that must be considered for its correct design can be grouped into the following categories: (1) **carbonaceous constituents**, (2) **nitrogenous compounds**, (3) **phosphorus compounds**, (4) **total and volatile suspended solids** (TSS and VSS), and (5) **alkalinity** (Metcalf & Eddy, 1991).

About animal slurry, organic matter, ranged typically from 70 to 85% of total solids (Burton, 2007; Hjorth et al., 2009), is essential for the biological process, because microorganisms use it for removing and reducing the nutrients from the slurry. Thus, total solids, volatile solids, biological oxygen demand BOD_5 and chemical oxygen demand COD are important parameters for defining organic matter in the slurry.

BOD is an unspecific parameter for carbon that is oxidized by the microorganisms present during standardized incubation (e.g., time, 5 days; temperature, 20°C).

The difference between the BOD_5 and the COD values reflects the difference between the various components: the first is related to the more readily digestible carbonaceous material, COD is the oxygen required to chemically degrade the oxidizable organic substance present in sample.

A large part of COD (more than 50%) can be considered as no biodegradable; this fraction is linked with the coarse material in the slurry (Burton C.H., 1992). In a typical animal raw slurry, the COD contents can vary from 1 g/L to 20 g/L. However, is also reported that can happen that only a part of the total organic substrates (45%) are aerobically biodegradable (BOD_5/BOD=0,88) (Magrí & Flotats, 2008). In the study of Magrì et al. (2008), who obtained results comparable with the study of Boursier et al. 2005 is attested that, although VFAs (volatile fatty acids) concentration, which represent source of readily biodegradable organic substance, in liquid fraction of pig slurry reach values up to 6 gO_2/L, the assessment of readily biodegradable organic substrates through respirometry gave only half of this value: they concluded that VFA which were not directly degraded were supposedly stored inside cells prior to degradation, but cannot be used as carbonaceous source.

The content of total nitrogen is another essential parameter, which must be considered in animal slurry for its treatment: an accurate quantification of different fraction and total N is necessary to select and design the most appropriate treatment to apply. In animal slurry N concentration is related to the protein contents of diet. Farm animals consume a considerable amount of protein and other nitrogen N-substances with their feed. The animal absorption of N contents in dietary is often inefficient and 50 to 80% of the N consumed is excreted. For example, pigs and cattle excrete more than 50 to 60% of the N by urine respectively (70% of the N in the urine is urea-N). Therefore, the contents of N in manure is a variable characteristic.

The liquid total ammonia N (TAN = $NH_3 + NH_4^+$) in pig and cattle urine and faces is lower (trace), but during manure storage urea and organic N or acid uric are transformed to TAN, that is dissolved in liquid (Sommer et al., 2013). Nitrogen as TAN is approximately 2-5 g/Kg_{manure} in the pig slurries (lowest value for sows) and 1,7-5,3 g/Kg_{manure} in the dairy cow slurry. For the latest value, it must be considered that the ammonia contents differ with the variation of housing. Concerning total nitrogen, in pigs' slurry N is about 3,8-6,7 g/Kg_{manure} and regarding dairy cows (depending on housing type) changing from 3,2 g/Kg_{manure} to 28,1 g/Kg_{manure} (Sommer et al., 2013).

Multiple studies have demonstrated the importance of the definition of the content of organic matter and nitrogen in manure and especially of their ratio. In fact, although these two parameters individually are not easy to monitor continuously at real scale, their ratio provides important initial information on the potential of the nitrogen removal treatment, as explain below.

The C/N ratio, expressing carbon contents as COD or BOD in their different fractions, is one of the most important parameters controlling the removal efficiency of nitrogen during biological treatment, and it can be used to help the **definition of the sequence** of different phases in a SBR operation. This because the catabolic reaction of bacterial population, can occur only if ratios between carbon, nitrogen and phosphorus (BOD_5:N:P) are respected. Indicatively, for denitrification this is equal to 6-8 $gCOD_{biod}/gN\text{-}NO_3^-$.

When the carbon already present in the wastewater/slurry is not enough to satisfy the request of the denitrifying bacteria, it is necessary to provide an external carbon source (as methanol, molasses etc.) and this leads to additional expenditure of money. This happens especially when denitrification is expected downstream of nitrification. Han et al., (2006) validated one of possible solution to this problem, applying the step-fed strategy. This scheme consists in separating the feeding load into sub-feeding streams which entering the system at the beginning of each non-aeration phase, which are at least 2. In this way the required carbon source is substituted by the organic compound present in the inlet flow.
However, if the low value for bCOD/N_{nit} (where bCOD means biodegradable COD) ratio implies a problem for the denitrification process, the higher bCOD/N_{nit} values will increase oxygen requirements due to heterotrophic biomass activity, which may even cause an increase in temperature inside the reactor that would be sufficient to inhibit nitrifying organisms. This kind of situation produces a state which can lead to losses up to 50% of the initially contents of total nitrogen due to ammonia stripping (Choi et al., 2005). If these aspects it be considered, coupling AD and NDN process can be not represent the best choice for nutrients removal: nevertheless, each case must be examined correctly before deciding.

Concerning phosphorus (P) in the slurry, 80% is in the orthophosphate form (PO_4^{3-}) (Hjorth et al., 2010) with a concentration ranged from 0,1 to 5 kg/ton with higher values referred to pig slurry. However in the literature, is reported a lower value, as in the study of Magrí & Flotats (2008), where is attested that only the 10% of TP (0,7 g P/L) was in the form of orthophosphate. Béline et al., (2004) reported that in liquid fraction of pig slurry the P concentration ranges between 0,96-1,22 kgP/ton: in that slurry 4–10% of phosphorus is dissolved (0,13-0,39 kgP/ton) while 60–85% is precipitated (4,0-6,0 kgP/ton), and 3–20% linked to the biomass (Beline et al., 2004). The $PO_4^{3-}{}_{(aq)}$ fraction may vary during storage increasing after excretion and then decreasing.

The particle size distribution influences the sedimentation of manure during storage and the choice of the appropriate separation process necessary to remove solid fraction. In general, in pig slurry and in cattle slurry 66-70% and 50-55% of the solids respectively are smaller than 25 µm (Hjorth et al., 2010; Møller et al., 2002; Sommer, et al., 2008). The particle size is correlate also with the nutrient content: in cattle slurry more than 80% of N and P are in the particle fraction below 125 µm, and in slurry more than about 70% of the un-dissolved N and P was in the particle size fraction 0,45–250 µm (Masse et al., 2005).

Ionic strength expresses the concentration of ions in a solution (Sommer et al., 2013). This parameter can be expressed by electrical conductivity (EC), which in the slurry ranges from 3 to 40 mS/cm (Burton C.H., 1992). EC has been observed to be higher than 10 mS/cm in several animal slurry studies (Sommer and Husted, 1995; Christensen et al., 2009, Hjorth et al., 2010; Masse et al., 2010). Liquid fraction of animal slurry typically that have a high ionic strength and high salinity levels, which can affect some treatments.

Obviously, the different treatments that are applied to animal slurries can change some physic-chemical characteristics and consequently influenced the choice of the final disposal of treated effluent or their further treatments. For example, the main variation brought by anaerobic digestion (AD), which is one of the most widespread treatment applied to livestock manure for the energy recover, is the reduction of the dry matter concentration, changes of the particle size distribution and not least on the breakdown of organic matter and nutrients. Into the digestate, there are the non-digested recalcitrant organic fraction, besides micro- and macronutrients. When raw pig slurry undergoes to AD, more than 80% of the total N becomes available as NH_4^+ (Vaneeckhaute et al., 2017). Effluent of anaerobic digestion has a highest concentration of TAN (1000–3000 mg/L as ammonium), smaller carbon to nitrogen ratios (C:N ratios) and reduced viscosities than undigested animal manures (Möller et al., 2012). It can be use directly or, in the most cases, needs further treated before utilization (Sui, et al., 2014). The species $NH_4^+ \leftrightarrow NH_3$, $CO_2 \leftrightarrow HCO_3^- \leftrightarrow CO_3^{2-}$, and $CH_3COOH \leftrightarrow CH_3COO^-$ mainly control the digestate pH value (Möller et al., 2012). During anaerobic digestion, different reactions occurs and some of them (e.g. precipitation of carbonates; precipitation of Fe^{2+}, phosphates releases protons, struvite formation) decrease the pH, and other (e.g. formation of ammonium carbonate ($(NH_4)_2CO_3$) and the removal of CO_2, the rising of concentration of basic cations, mineralization and reduction of multivalent ions in feedstock) increase the pH. Overall, the pH after a digestion process is higher than in an untreated slurry. In comparison with a pH of slurry, the median digestate pH amounts to 8,3 (Vaneeckhaute et al., 2017). AD changes also the contents of the amount of reactive phosphorus and magnesium, which are increased.

3.5. Typical SBRs control parameters

There are also physical-chemical parameters that play a fundamental role not only in the definition of the process but also in its correct functioning.

The first is Oxidation Reduction Potential- **ORP** or RedOx- that generally gives a measure of the oxidative state in aqueous system, gages the electrical potential required to transfer electrons from one compounds or element to another compounds or element. ORP is measured in mill volts: the negative value indicating a tendency to reduce compounds (elements) and positive values indicating a tendency to oxidize compounds (elements).

ORP has been shown to be a **helpful tool** for representing the biological state of a system. It can be used for its correlation with biological reactions between anoxic and anaerobic conditions, or as monitoring parameter to ménage biological nitrogen removal in SBR system (Akin & Ugurlu, 2005). In particular ORP is very useful to detect different phases of the SBR cycle (anaerobic, aerobic and anoxic) as reported in several studies (Dutta & Sarkar, 2015; Obaja et al., 2003; Ra, Lo, & Mavinic, 1999).

The normal range of values of ORP is:

- 0 to 50 mV in aerobic stages;
- 0 to -300 mV in anoxic stages;
- Below -300 mV for anaerobic environment (Dutta & Sarkar, 2015).

In the study of Kishida et al., (2003) the final ORP at the end of the non-aeration phase was within the range from -300 to -400 mV. This value attest the end of denitrification phase, and is very similar to those observed in the study of Han et al., (2008), where the minimal concentrations of N-NO_3^- (< 12 mgN/L) were observed in the ORP range from -320 to -412 mV.

In a biological process applied for nutrients removal, during anoxic condition, ORP has a continuous dropping profile with respect to time. Along this, it is possible to identify a particular point, known as **nitrate knee**, (Figure 9) which corresponds to the end of nitrates removal and so, to the end of denitrification: when this situation occurs, it is safe to stop anoxic phase and start the next step (Dutta & Sarkar, 2015; Kocijan & Hvala, 2013; Zhu et al., 2006).

As explain in the study of Han et al., (2008), the ORP has a significantly decrease throughout the non-aeration phase when microbes have reduced oxides: denitrifies reducing NO_3^- to nitrogen gas and sulphate-reducing bacteria (SRB) reducing SO_4^{2-} to S_2^-. In addition, Han et al., (2008), highlighted as sulphides have a great impact on ORP: a 0,07$mg_{sulfide}$-S/L concentration increase, induces a 100 mV fall of the ORP value. Instead, there is an increase of ORP during the aeration phase due to oxides production catalysed by aerobes in the presence of DO (dissolved oxygen).

Even if the ORP can be a useful tool for monitoring denitrification process, its application can lead to an incorrect result. In fact, as shown in the same study it is possible that the nitrate knee on the ORP profile is missed. This is probably due to a sharp drop in the ORP profile following the feeding, which covers the nitrate knee.

Also during the oxic period, ORP can be useful indicator. On its pattern, it is possible to identify the **nitrogen break point** (Figure 9), which represents the end of nitrification. Even in this case, the appearance of this point is not certain: the absence of ORP nitrogen break point in the case of a complete nitrification could be due to excess aeration for the instantaneous oxygen demand. In the step-fed SBR process (please also see par. 3.6), with the excess of aeration, the DO concentration at the end of aeration before feeding could increase and could lead to an aerobic waste of organic matters contained in the influent.

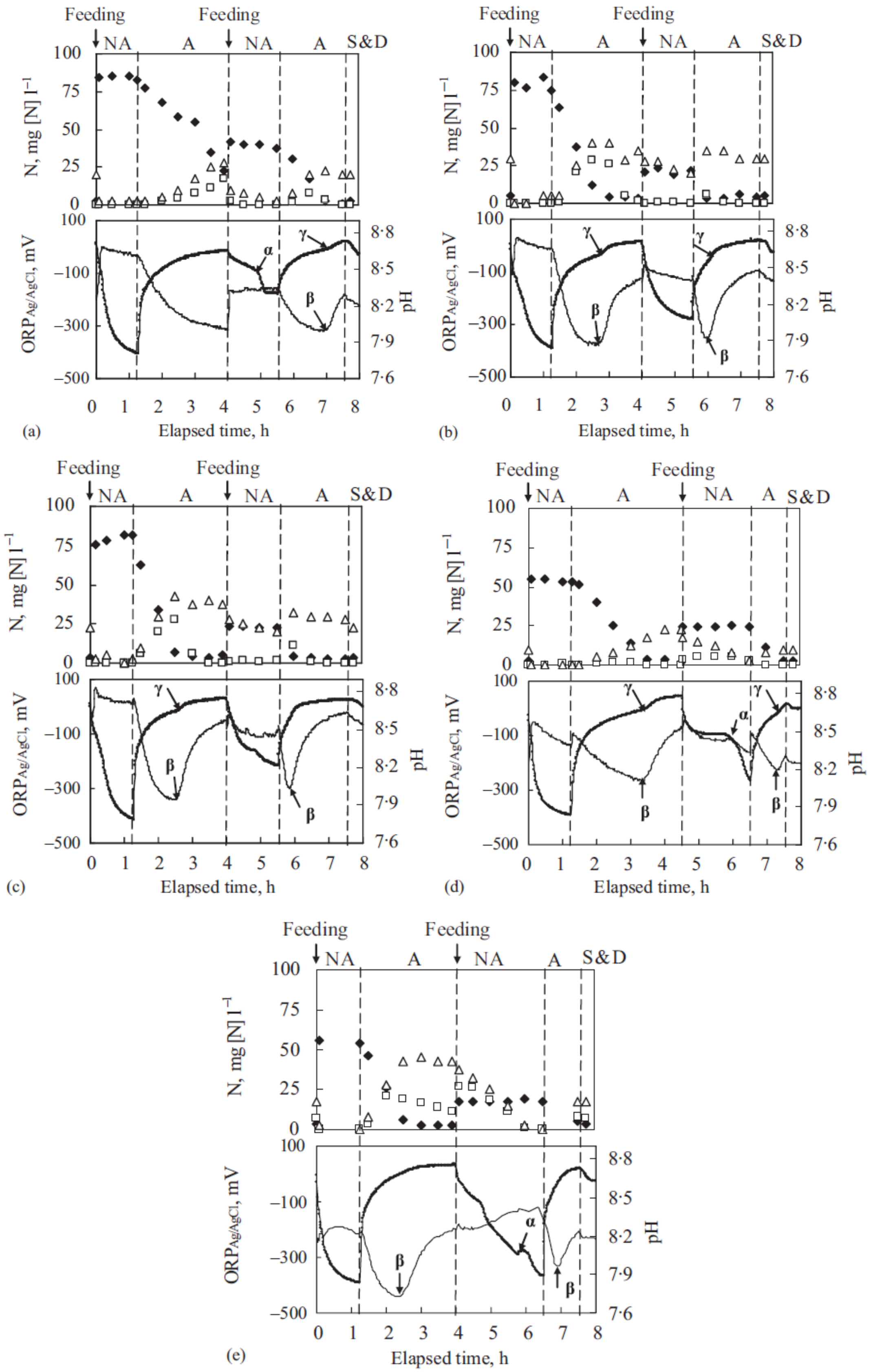

Figure 9 Variations of oxidization–reduction potential (ORP), pH and NH_4^+-N, NO_2^--N, NO_3^--N during one typical cycle in five SETs;(a) SET1; (b) SET2; (c) SET3; (d) SET4;(e) SET5; ♦ NH_4^+-N; □ NO_2^--N; Δ NO_3^--N; ● ORP; — pH. NA, non-aeration phase, A, aeration phase; S&D, settling and discharging; α, nitrate knee; β, ammonia valley; γ, nitrogen break point. Han et al., (2008).

With the aim to have a helpful feedback indicator for regulating the, the appearance of the ORP nitrogen break point can be considered in order to avoid organic matter loss and reduce energy waste.

It is also possible to use the consistent increasing and decreasing of ORP to diagnostic the problem occurring during aeration phase, which represents a critical issue for aerobic bacteria.

The use of ORP can be applied also for monitoring the phosphorus removal, when SBR is configured with this aim. Also a relation between ORP and DO is individuated; in particular, it was found that the ORP value is correlated with the logarithm of the DO concentration in a linear relationship (Akin & Ugurlu, 2005).

The characteristics curve and the abovementioned bending points of ORP's path is highly dependent to the denitrification/nitrification rate of nutrient components in wastewater/slurry. For these reasons the effectiveness of each bending point of ORP functioning as the control point varies in different SBR systems.

Another important parameter connected with biological system that responds to microbial reaction is **pH**. Its variation can be used as indication of the ongoing biological reaction, even if this parameter has a more complex profile than ORP (Han et al., 2008). The optimum pH value is consider to be equal to 8,0 in order to allow biological growth regardless of the kind of involved organism in microbial reaction (Magrí & Flotats, 2008).

As well as the ORP value, also the pH trend can be identified significant points: at the end of the nitrification in the aerobic phase, can be distinguished the pH **ammonia valley** (Figure 9). Furthermore, pH shapes the end of the denitrification in the anoxic phase through the pH **nitrate apex**, which indicates the shift from the anoxic to the anaerobic conditions, necessary for phosphorus removal.

Ammonia valley is an evident point along the pH profile, during aeration phase: its appearance is linked with nitritation (the first step of nitrification), during which acid is produces and pH tends to decline gradually at the beginning of nitrification. When all the ammonia has been oxidized and transformed into nitrite, which concentration produces a peak, the acid production lined with ammonia conversion is stopped and pH profile shows a concomitant minimum known as *ammonia valley*. The complete removal of ammonia indicates also the end of alkalinity consumption in the wastewater (Akin & Ugurlu, 2005; Han et al., 2008; Kim et al., 2004).

During the anoxic stage, contrary, the pH growths and produces a continuously rising profile. The maximum is reached when the total nitrate is converted to nitrogen, indicating an end of denitrification stage: this point is called *nitrate apex* and corresponds exactly with *nitrate knee* as observed through ORP profile (Akin & Ugurlu, 2005).

Although researchers argue that pH profile is the best indicator of the changes in the microbes profile occurring inside a SBR reactor, the alkalinity present in the wastewater often has a robust buffering capacity that contracts noticeable the pH variation (Dutta & Sarkar, 2015).

For example, Kocijan & Hvala (2013), observed in their study that pH *ammonia valley* and the pH *nitrate apex* were difficult or even impossible to observe also for this reason. In addition, they claimed that the chemical background of this characteristic pattern is still unclear, and so the pH was used only as an additional regress or to the DO and ORP signals. Nevertheless, in most studies, pH is used as a powerful indicator; for example, in the study of Han et al., (2008), a correlation between pH and NH_4^+-N is found in a power function during nitrification occurring in the first aeration phases of their experiment. On the contrary, in the same research, it is established that there is not a significant correlation was found between nitrogen oxides and pH. Also in this case, the use of pH trend for controlling SBR operation is useful not only for the nitrogen removal but also for the phosphorus deletion. For example, Akin & Ugurlu, (2004) concluded that through pH path beside the individuation of the end of nitrification, it is possible to recognize significant points the such as phosphate release and completion of phosphate uptake.

The dissolved oxygen **DO** is an important parameter that shall be verified during biological process, since it serves as an electron acceptor in nitrification and acts as a limiting factor in denitrification. Between the various operating conditions that may be changed in SBR systems, the aeration profile it is one of the most important control variables (Spagni & Marsili-Libelli, 2009).

During aerobic phase, in order to guarantee nitrification, a high DO concentration is requested. In this phase, it is possible to use the DO concentration for process control, because the end of nitrification can be deduced from the DO time history. This is achievable because the respiratory activity of the biomass decreases when the nutrients are exhausted in the reactor. Consequently, at the DO breakpoint, DO increases as a result of the process transition from a high to a low oxygen-uptake rate (Kocijan& Hvala, 2013). A significant zero DO level was present at the beginning of the aerobic phase, however, the DO level increased to 2,3 –2,7 mg/L at the end of the same phase. The maximum rate of nitrification has been observed to occur at a DO level greater than 2,0 mg/L and denitrification process is effective at anoxic conditions when DO level falls below 0,5 mg/L and at greater than 1,0 mg/L, the denitrification process is inhibited. As well as other parameters, also DO can be used for monitoring phosphorus elimination. For example, in the study conducted by Akin & Ugurlu, (2005) was observed that it is possible to control phosphorus uptake and nitrification by using DO profile (in aerobic condition), but the same consideration cannot be made during anaerobic phase. During this phase the nitrification rate could be used for aeration control as referenced parameter due to its likeness of the aeration and NH_4^+-N loading rates (Han et al., 2007).

The increase of DO concentration affects phosphorus uptake in the biological phosphorus removal process: excessive aeration may affect the reduction of biological phosphorus removal and can lead to the stop of phosphorus uptake. This fact results of the gradual depletion of poly-β-hydroxybutyrate (PHB). When nitrogen and phosphorus are targeted, biological phosphorus elimination is interfered with the final or intermediary products of nitrification-denitrification, as nitrite, nitrate, and nitrous acid, which are closely associated with the DO level.

It is crucial to consider also the influence of loading rates with the oxygen demand. In some studies, in fact, it was observed that nitrification process is accomplished at DO levels lower than 0,5 mg/L at short sludge ages in activated sludge system operates with low organic loading rates.

The intensity of aeration during aerobic phases has an important role for definition the proper DO concentration and significantly influences the performance of the overall process (Han et al., 2007). In addition, DO concentration is a very important parameter for controlling the SBR operational costs (Han et al., 2007). The aeration phase, in fact, require the greatest part of energy.

Concerning other process parameters, SBR correct operations can be carried out without any special concern at **temperatures** higher than 16 C° (Obaja et al., 2003). However, temperature parameter is fundamental for a proper development of microbiological metabolism and often imposes some limitations for reaching acceptable nitrification rate.

Also the knowledge about microbial population involved in activated sludge processes has an important role for understanding the biological process and to handle correctly the treatment plant. The chemical-physical approach for analysing the waste composition does not allow evaluating the activity of the biomass and the various kinetic parameters necessary for modelling the process. The chemical and physical methods have little use for the application of simulation models and process control unless combined with biological measures.

The Oxygen Uptake Rate OUR, ammonium nitrogen concentration AUR, the Nitrate Utilization Rate NUR tests are efficient tools for characterizing biomass in sludge in SBR systems. Routine performance of the AUR and NUR tests can be a useful tool for estimating any problem arising from the process (for instance, low temperatures). In addition, it is observed that values of AUR, NUR and OUR increase with substrate concentration and that is possible to identify a correlation between these parameters and average nutrient removal rates observed during SBR operation (Obaja et al., 2003).

The ammonium-oxidizing bacteria –AOB- community structure, which are responsible of the ammonia transformation, is influenced by the ammonium nitrogen concentration, pH, DO (dissolved oxygen), C/N (carbon-nitrogen) ratio, organic carbon concentration and inorganic carbon concentration (Sui et al., 2014).

AUR (Ammonium Utilization Rate) allows studying autotrophic bacteria activity, measuring the consumption of ammonium made by bacteria, during the time.

NUR (Nitrate Utilization Rate) allows to study denitrifying bacteria activity, measures the consumption of ammonium made by bacteria, during the time.

OUR (Oxygen Uptake Rate) represents the oxygen utilization speed by microorganism community. The measurement of OUR, which can be measured with respirometry techniques, is commonly used for the compositional typification of waste water and the calibration of bio-kinetic models (Zonta et al., 2014).

3.6. Operational strategies of SBRs applied to livestock manure for nitrogen and phosphorus removal

SBR, compared with other system, has attracted even more attention for its high efficiency in removing nutrients from animal slurries (Bernet et al., 2000; Bortone C.H. 1992; Obaja , Mace et al., 2003; Obaja, 2005; Zhu, et al., 2006). Since 1980s, scientific articles attest the interest relating to the use of SBR for pig slurry treatment (Choi et al., 2004; Deng et al., 2008; Poo et al., 2005). The earliest experiments reported in the literature about the treatment of livestock manure using a laboratory-scale SBR are dated from the late 1980s (Magrí & Flotats, 2008). Nowadays, there is evidence of operative and serviceable SBR reactors for treatment of liquid fraction of pig slurry in several countries, at both pilot-scale (Poo et al., 2005) and full-scale (Deng et al., 2007; Lo & Liao, 2007, Magrí & Flotats, 2008).

As say before (paragraphs 3.4), the characteristics of animal slurry are very different from the other type of wastewater. In Table 5 are reported data about the characteristics of livestock slurries, which are used as substrates in that several scientific researches. Most of them used as substrate for experiments pre-treated pig slurries or anaerobic digested pig slurries; in some studies, the substrate undergone to liquid solid separation and/or dilution before to be used in the experiments. The pH of substrates range is between 7,1 and 9. The carbon content (as COD) varied from 1 g/L to 50 g/L. The concentration of nitrogen ranged between 1,0-3,5 g/L as TKN and 0,16-3,8 g/L as N-NH_4^+. Nevertheless, the content of ammonium nitrogen in most of considered researches were lower than 1 g/L. This parameter is also important because the reachable removal rate of nitrogen depend also by the initial concentration of nitrogen in the slurry. For example, as observed by Obaja et al., (2005), the maximum initial NH_4^+-N concentration that could be treated in SBR was 1500 mg NH_4^+ -N/l day.

Then, in the following, deeper analysis about the studies and researches concerning nitrification and denitrification process and biological phosphorus removal carried out in SBR system for treating livestock manure is done.

In Table 6, 7, 8 and 9 the main characteristics and results obtained by different studies are reported.

Among the studies analyzed, the values of nitrogen removal yields (such as TAN) are very high both when the treated wastewater was a liquid fraction of pig slurry and when it was a liquid fraction of digestate. Nevertheless, for these substrates the lowest removal values are attested (71% for a wastewater pre-treated in a UASB reactor and 68% for an anaerobically pre-treated slurry).

Regarding COD removal rate, the smallest removal yields were observed for a liquid fraction of digested and for a swine effluent pre-treated through a coagulation/flocculation treatment. However, as will be explained in the previous paragraphs, the characteristics of the treated wastewater are fundamental for the success of the removal process, but the operational parameters of the process are equally important.

Since the characteristics of the animal manure produce by farms are very inconstant, the first problem addressed and analysed here concerns HRT.

Studies affirmed that the fixed hydraulic retention time (HRT) used in traditional treatment processes frequently results in poor control over the quality of the effluent and in extended treatment that is inefficient in terms of energy consumption. In fact, the high nitrogen contents of animal slurry leads to the application of long hydraulic residence times (HRT) for reaching the removal required result, even if this problem could be solved, in some cases, with the addition of external carbon source (Obaja et al., 2003). Anaerobically digested swine slurries (or wastewater), had high concentration of ammonium nitrogen and a low C/N ratio and so, also for this kind of substrate it is necessary to apply a long hydraulic retention time (HRT) in order to reach the required removal rate (Obaja et al., 2003, 2005; Sui et al., 2014). Nevertheless, in the analysed studies (Table 6,7,8, and 9) that used SBR lab-scale prototype, and applied fix HRT, this parameter ranged between 0,1 and 3 days. Among the studies analyzed, only in one case that studied at real scale the nitrification/denitrification process applied in one tank, the fixed HRT is equal to 9,4 days (Riano et al., 2014).

In fact, as known, another process parameter defines the permanence of biomass in the reactor: the sludge retention time (SRT). The SRT, besides its importance in correct process definition, is strictly connected with the biomass activity. Regarding the SBR applied to treatment of livestock manure, Akin & Ugurlu, 2004 observed significant changes in the effluent concentrations of phosphate, ammonium, and oxide nitrogen with changing SRT. In particular, through their experiment, they attested that SRT could influence phosphorus uptake and release and the nitrogen removal rate. They detected that concentration of ammonium nitrogen in the effluent increased with decreasing SRT. Besides, in experiment of Akin & Ugurlu, (2005) it was observed that there is discrepancy between the optimum SRT applied for P removal and for N removal: the highest efficiency occurred with a SRT of 10 d and 25 d for P removal and nitrification rate achievement respectively. In fact, with the growing of SRT, attained by increasing the mixed liquor suspended solids (MLSS) concentration in the reactor, it is possible to increase concentration of nitrifiers and consequently the nitrification rate.

For modifying SRT another possibility is the application of low inlet flow rate. Among the investigated researchers, only in one case the SRT was lower than 11 d (Akin et al., 2003). In the other studies, the N-NH_4^+ removal rate is higher than 68% regardless of the applied SRT.

Actually, as also indicated in the paragraph 3, not only HRT and SRT influenced the removal yields achievable. In the study of Akin et al., (2003), in which was applied constant SRT, the obtained removal rate changed a lot varying the way of influent feeding.

Also the duration of each phase as an important influence on removal rates: in the study of Han et al., (2008), it was observed that the duration of the non-aeration phase played a key role in improving denitrification efficiency. In fact, a prolonged non-aeration phase enables denitrifies to use its cellular-stored carbon to accomplish a endogenous denitrification. Akin & Ugurlu, (2004) attested that phosphate release (about 6%) occurs at the end of the settling phase. This fact shows as a long settling period kept for their experiment (1,5 h), resulted in the establishment of anaerobic conditions. In order to reduce the phosphorus release and to improve sludge settling characteristics a shorter settling period (30 min) was applied in their experiment.

In the studies considered, the operation setting, and in particular the duration of each phase, were very changeable. When an SBR is used for nutrients removal, the most applied time cycle is equal to 8 hours; in few cases the cycle's duration is higher than 24 h or lower than 4 h. In many application cases, the denitrification phase was preceded to the phase of the need to exploit the naturally occurring carbon in the wastewater/slurries used. About that, considering livestock manure slurries and wastewater, but also for other type of wastewater, the definition of the of organic matter and nitrogen contents and especially of their ratio (paragraph 3.4) is essential. Considering the analysed studies, COD/TKN, BOD/TKN, BOD/COD and BOD/TAN are defined (where possible) (Table 5). COD/TKN is always higher than 4,2 and only in one case TOC/TN ration is lower than 2 (1,2). Anyway, in this specific case, the aim of the research was to develop a real-time control strategy to respond to cases in which the ratio between carbon and nitrogen was not enough to allow complete denitrification of NO_3^- products. The removal yields achieved was 94,7% for the TOC and 96% for the TN (Kim et al., 2004). Obaja et al. (2003) support that C/N ratio as (C–acetic/NO_x–N), must be equal to or higher than 1,7 to obtain complete denitrification to molecular nitrogen; the data found by other researches, even expressed in a different way, were different. Henze et al. (1995), identified minimum values of 5–6 to ensure complete denitrification, as supported also by Choi et al., (2004), which indicated that COD/TN rate greater than 6 and the Alk/TN ratio higher than 3,6 are required to have higher removal efficiency. As shown in the Tables 6,7,8,9, the lowest observed removal rate for TAN (68%)

correspond to a BOD/TN=2,09. In the study of Mees et al., 2014, whose characteristics were not reported in Tables (6, 7, 8 and 9), SBR was applied for treating bird slaughterhouse and cold storage processing wastewater (135 mg/L of NH_4^+-N), the ratio value C/N (COD/N-NO_2^-+ N−NO_3^-) shall be ranged from 6 to 9 for obtaining the highest value of inorganic nitrogen efficiency removal.

In the study of Magrí & Flotats, (2008), was indicated that a lack of biodegradable organic carbon, represented by a BCOD/TKN ratio equal to 3,9, may limit denitrification potential. The authors suggested considering carefully the adoption of other ratios that do not contemplate the biodegradability of organic carbon, such as the COD/TKN ratio (8,8), as they may lead to erroneous interpretations.

Some of analysed researches were focused on acting on the way of reactor feeding, or on the possible modification of the inlet sludge composition for changing the ratio between biodegradable chemical oxygen demand and nitrificable nitrogen (BCOD/N_{nit}). For increasing the BCOD/N_{nit} ratio due to the lack of internal carbon source, Lee et al., (1997) and Obaja et al., (2005) proposed to use as external carbon source matter already available on farms (e.g. Pig Raw Slurry). This allowed to not increase system operating costs, obtaining the desired removal yields. Otherwise, Zhu et al., (2006) analyzed the variation of C/N value operating on addition of synthetic organic supplements and concluded that, contrary to other researches, the global TKN removal rates, with or without acetate addition, were similar, indicating that most of TKN could be decomposed by microorganism using the carbon presents in the slurry.

Further studies are focused on optimizing the addition of the external carbon source under low C/N load cycles in order to enhance nitrogen removal (Kim et al., 2004).

Other observations regard the relation between the C/N ratio and the nature of reactor’s influent. In fact, differently from some types of manure (as pig slurry), BCOD/N_{nit} ratio is lower if the sludge was previous subjected to anaerobic digestion (Deng et al., 2007; Kim et al., 2004; Magrí & Flotats, 2008). As reported in the study of Otawa et al.,(2006), in the anaerobically digested swine wastewater, COD/TN ratio is equal to 1–3, and in their study this resulted insufficient for TN removal.

In many studies, the operation of reactors occurred at controlled temperature to optimize bacterial activity (above 20°C). Appling simplified Van't Hoff Arrhenius equation (equation 14), where μ_T is the maximum growth rate at the operating temperature T and $\mu_{20°}$ the reference speed assumed equal to 20°C, it is evident that the bacterial growth increases with temperature. Θ is the constant of adjustment, that assumes different values depending on the biological processes considered. In an activated sludge system θ is worth 1,06-1,12 for the nitrification process and 1,03-1,12 for the denitrification process.

$$\mu_T = \mu_{20°} * \theta^{(T-20)}$$

Equation 14

Nevertheless, some studies analyzed deeper how the removal rate changed varying the operational temperature applied. In the study of Deng et al., (2007) is observed that, although the air temperature fell below 10°C in winter, with water temperature ranging from 10 to 15 °C, SBR still showed good removal of COD and NH_4^+-N.

To establish the temperature limits, Obaja et al., (2003) done a series of experiments applying nine different temperatures from 8 to 25°C. At the lower temperatures tested, substantial reduction in removal percentage occurred. This phenomenon was especially evident below 16°C. Under 16°C the ammonium removal yields was unacceptable in most of the practical situations (Figure 10) but a signify decrease in the yield of the process did not occur until 8–10 °C (Dutta & Sarkar, 2015). Contrary, temperature does not affect denitrification reaction.

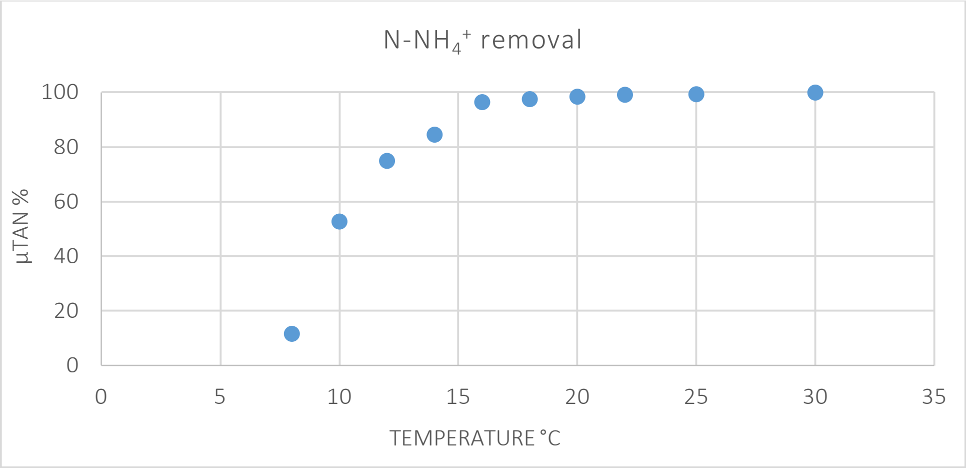

Figure 10 Removal rate of TAN varying the temperature (Data elaboration from Obaja et al., 2003).

The content of dissolved oxygen in the reactor during nitrification and consequently the aeration flow has an important role in the reaching the desired nutrients removal yields, as also explained in the previous paragraphs. Han et al., (2007), for example, observed that, using swine wastewater with a COD concentration of 5,9 g/L and COD/TN/TP ratio of 100/21/1, the suitable aeration intensity was 4,2 L/m^3/s for the twice-fed SBR tested (HRT: 3,3 d; SRT: 27 d) in order to achieve optimal nitrogen and carbon removal rate and prevent a phosphorus accumulation with minimal energy consumption. Wu et al., (2015) indicated that an anoxic stage in SBR reactor could be maintained during aeration phase when air was supplied at 0,06 $L_{AIR}/L_{SLUDGE}/min^{-1}$.

In real SBR installations, the on line control of direct parameters such as COD or BOD, TSS, and various forms of nitrate and phosphate (NO_3^-, NH_4^+, PO_4^{3-}) is often expensive and difficult to achieve with the current available technology (Akin & Ugurlu, 2005). These direct measurements cannot be frequently performed through an on-line monitoring system mainly for the economic costs. For this reason, the process control operation, as happen for the development of algorithm used for this scope, are often based on indirect process variables, which usually comprise pH, ORP and DO. These parameters, are explained in the previous paragraphs, are well linked to the path of biodegradation processes, which can be inferred from these measurements (Ga & Ra, 2009; Gu, Wang, Yang, Yang, & Peng, 2012; Kishida et al., 2003; Won & Ra, 2011) and can be can be used for developing a proficiently process and a correct control strategy for livestock manure treatment.

Generally, the control strategy plays a key role in the process optimization not only for the nutrient removal, but also for the limitation of energy and operational cost. Is reported that SBR process automation, can also afford to save more than 60% of the operating cost required in a conventional activated sludge process applied to wastewater and guarantees high effluent quality with a shorter aeration time (Dutta & Sarkar, 2015; Yang et al., 2010).

A lot of scientific researches, both in the field of livestock manure treatment both and wastewater treatment, are aimed to study the application of adjustment of SBR cycle length or other process parameters by using real-time control and common probes (Kim et al., 2004; Kishida et al., 2003; Won & Ra, 2011; Poo et al., 2005; Han et al., 2008; Han et al., 2007).

In general, control strategy applied in that specific case to SBR systems, can be divided into fixed-time control strategies, based on the set “fixed” duration of each phase, and real-time control strategies, which adjust the duration of the phase on-line, during process. In the latter case, each operational period should be concluded when the desired biodegradation processes in the considered phase are finished. For example, the aerobic phase should be stopped when all the ammonia is converted into nitrate, while the anoxic phase should be completed when the nitrate has been denitrified and all the available organic carbon has been used in the process. For this goal, the use of process parameter is necessary.

Won & Ra, 2011 at the SBR farm-scale studied a process with newly designed real-time control strategy using ORP and pH (mV)-time profiles: they evaluated its practicality and analysed the operational characteristics of the real-time control. Using this strategy, the duration of the anoxic and aerobic phases was flexible from cycle to cycle. As attested in their study, through this control, a very high nitrogen removal efficiency was achieved (100% NH_4^+-N and 98% soluble N) despite high N loading rate variations. They underline that the most evident advantage of real-time process control over a fixed-time cycle mode of operation is the complete removal of contaminants in the wastewater by the achievement of a flexible hydraulic retention time (HRT). In particular among their test, they reached the higher removal rates with cycle duration time average equal to 14,6 h, having 4,01 h of air-OFF (mixer-ON) and 8 h of air-ON.

Comparing that result with ones obtained with the fixed-time operation (24 h cycle) they fund an energy usage saving about 39%.

Using as a control parameter the pH value, Akin & Ugurlu, (2004) observed that during the shorter settle phase set in their experiment (0,5 h), a decrease of pH value occurred due to CO_2 accumulation when oxygen was used up in respiration. Therefore, pH can be a useful tool for defining the correct duration of sedimentation phase. In addition, the same authors, in other study (Akin & Ugurlu, 2005) observed that, during a P-removal process the oxic phase can be ended after the achievement of pH plateau: a confirmation of the potentiality of this parameter for controlling the process.

There are also studies focused on establish operating strategies suitable for the fluctuations of influent loads; for example the research of Kim et al., (2004) aimed to study an integrated strategy of real-time control. They set an integrated real-time control strategy, feeding the reactor pulsing swine waste only when the nitrate breakpoint occurred in the ORP-time profile. This approach enables the optimization of swine waste addition.

Other studies, even though do not concern livestock slurries, are focused on the optimization of a particular phase of SBR cycle as in the study of Flores-Tlacuahuac & Pedraza-Segura, (2016) where the nonlinear continuous optimization formulation is proposed.

Recently, computational intelligence methods for SBR control and decision-making have proved to be very useful for on- line control. Some researchers studied different methods like artificial neural networks, fuzzy logic, neural fuzzy model and similar used for the inference of the operation-phase ends (Hong et al., 2007; Dias and Ferreira, 2009; Huang et al., 2010; Marsili-Libelli, 2006; Marsili-Libelli et al., 2008; Sottara et al., 2009; Sottara et al., 2007). Statistical methods and probabilistic methods are used too (Villez et al. 2010, Sottara et al. 2009; Zonta et al. 2014). For example, Poo et al., (2005) in their study presented the local control techniques such as the diagnosis of influent loading rate and determination of aeration period centred on ORP and DO profile. In addition, they described the remote management system that can be accomplished by the fuzzy inferred system and artificial neural network (ANN) that allows reaching the complete removal of TAN.

There are no doubts that the best removal yields, and the best saving of energy and economic resources is obtained with the application of real-time control of the duration of the process and the phases. In this strategy, an HRT that is not pre-established but varies over time optimized using ORP or pH, and DO.

Concerning not only the process control strategy, but also the modelling of biological process, despite the numerous studies relating to the modelling of urban wastewater treatment (for example, the ASM1 model that have been developed and edited by the International Water Association, and their following variation-ASM2, ASM2d, ASM3), there are not many researches focused on modelling of livestock waste treatment using SBR system. Furthermore, for the correct utilization of ASM models it is necessary to know a lot of input parameters besides kinetics and stoichiometric parameters. As explain before, standardization of livestock slurry characteristics is very difficult as well as summed up this in synthetic parameters for applying it to different cases. Also for these reasons the direct use of these models for simulating the treatment of concentrated streams as piggery wastewaters, may not be a suitable option (Magrí & Flotats, 2008).

Nevertheless, there are studies that purpose to establish and calibrate a model for the simulation of the NDN process applied for the treatment of the liquid fraction of piggery sludge in an SBR. In that model, developed by Magrí & Flotats, 2008, nitrification and denitrification were considered as two-step processes, and pH was considered a state variable. Sensitivity analysis was also conducted to identify those parameters that affects mostly the response of the model under test conditions.

It is interesting to note that among the parameter considered for the model, the average of values of the parameters related to aerobic hydrolysis (kh, hydrolysis rate constant and K_X, particulate COD) were estimated to be 4,13 1/d and 0,17 mgCOD/mg COD, respectively. Typical values given in ASM1/2 for urban wastewaters treatment are lower than estimated one [kh= 3,00, K_X =0,03/0,10]. Indeed, parameter values may vary significantly according to the origin of the wastewaters.

Béline et al., (2007) have studied and proposed a version of the ASM1, which uses data from a pilot-scale SBR for calibration procedure. The whole objective of their work was to develop and to calibrate a mathematical model for the simulation of the modified SBR, treating piggery wastewaters. That latter evolution introduces an intermediate step: nitrite was considered as an intermediate in both the nitrification and denitrification processes. During nitrification, temperature and DO concentration were recognized as significant factors, which induce nitrite accumulation during nitrification.

It is therefore clear that modelling the process can be a powerful tool; however, it must be used taking into account that the input parameters and variables must be related with livestock manure and wastewater and that therefore the default values normally used may not represent exhaustively any specific case.

Table 5 Characteristics of substrates used in several research. SDP: Supernatant of digested piggery wastewater; SP: Supernatant of NON-digested piggery slurry; SDPC: Supernatant of digested piggery wastewater after centrifugation SW: Swine pre-treated Wastewater; SWSS: Swine Wastewater Screened/Sieved; DSW: Digested (UASB) Swine Wastewater; DRS: Diluted Raw Slurry; SRS: Swine Raw Slurry.

Parameters	U.M.	Magrì et al., 2008	Magrì et al., 2009		Obaja et al., 2003	Obaja et al. 2005		Kimshida et al. 2003		Kim et al., 2004		Poo et al., 2005	Han et al. 2007	Han et al. 2008	Wu et al. 2015	Deng et al. 2008	Zhu et al. 2006	Qing et al., 2011	Beline et al., 2007	
Substrate	-	SDP	SP	SP	SDPC	SDPC	SP	SW	SRS	SW	SRS	SW	SP	SW	SWSS	DSW	DRS	-	SWSS (1)	SWSS (2)
pH	-	8,0			8,4	8,42	8,45	8,9	-	-		8,7	-	-	7,1	-	7,4	7,6	-	-
EC	dS/m	18,2			-	-	-	-	-	-		-	-	-	-	-	-	-	-	-
TKN	g/L	3,4	2,7	3,4	-	-	-	-	-	-		-	1,3	1,2	-	-	1,22	-	-	-
TN	gN/L	-			1,7	1,65	0,79	1,13	3,7	0,72	4,53	-	-	-	1,2	0,83	-	-	4,6	4,3
NH_4^+-N/TAN	gN/L	2,6	2,0	2,6	1,6	1,60	0,72	1,12	-	0,589		3,8	1,2	1,2	0,9	0,75	0,86	0,17	3,0	3,4
NO_2^-	mgN/L	-			0,0	0,0	0,0	<0,1	-	-		-	0	n.d	-	-	0,0	0,06	0,0	0,0
NO_3^-	mgN/L	-			0,0	0,0	0,0	<0,1	-	n.d		-	0	n.d	-	-	0,0	0,65	0,0	0,0
BOD	gN/L	11,4			-	-	-	-	-	-		-	-	-	3,7	-	3,66	-	-	-
BOD5	gN/L	-			1,73	1,73	3,25	2,96	97,3	3,2	90,28	8,0	2,98	3,57	-	0,26	-	-	-	-
COD ($^{45\ \mu m}$)	gN/L	30,1	18,7	30,1	3,97	3,09	7,45	-	-	-		-	5,94	6,13	8,8	1,19	8,80	1,5-2	50,60	39,40
CODs	gN/L	8,0			-	-	-	-	-	-		11,0	-	-	-	-	-	-	21,80	14,80
TOC	mg/L	-			-	-	-	1230	-	864	26,17	-	-	-	-	-	-	-	-	-
DP	mg/L	-			-	-	-	-	-	-		-	46,7	44,7	56	-	39,9	-	-	-
TP	mg/L	722	1100	700	171	-	-	20,8	2780	46,0	2,6	20,0	59,1	59,2	601	46,4	601	-	-	-
PO_4^{3-} -P	mg/L	75,0	200	100	147	147	120	4,6	-	18,0		-	-	-	-	-	-	-	-	-
TS	g/L	24,9	26,5	24,9	11,2	11,2	13,4	-	-	-		-	-	-	10,5	-	10,5	-	54,0	31,1
TSS	g/L	17,7			2,58	2,58	3,10	0,31	115,7	0,92	0,917	1,5	-	-	-	-	7,66	-	-	-
VS	g/L	15,3			5,35	5,35	6,23	-	-	-		-	-	-	-	-	5,40	-	42,50	20,10
VSS	g/L	-			1,96	1,96	2,20	-	-	-		-	-	-	-	-	4,42	-	-	-
Alkal3,53	gN/L	-			5,23	5,23	6,53	-	-	-		-	-	-	-	-	-	-	-	-
Alkal7,74	gN/L	-			1,18	1,18	1,45	-	-	-		13	-	-	-	-	-	-	-	-
VFA	gN/L	-	2,1	6,4	-	1,05	5,28	-		-		-	-	-	-	-	-	-	10,70	6,20

Table 6 The summary of considered studies (W: wastewater, SW: synthetic wastewater; DPS: digested pig slurry; P: pig slurry; PDPS: partially pig digested; BS: bench scale; SRPS: supernatant of raw manure finishing barn pigs; SP: sown; AD: anaerobic digestion; at: anaerobic treatment; S/L :solid liquid separation; C/F coagulation/flocculation; DI: Dilution; OPC: operational conditions.

	SLURRY TYPE	PRE-TREATMENT	TREATMENT	SCALE	SCOPE
Tilche A., 2001	P	S/L	SBR NDN	REAL	MONITORING- MODEL VALIDATION CALIBRATION
Wu et al., 2017	P	S/L+ D	SBR NDN+P	LAB 1,2	STUDY STEP FEED INFLUENCE ON C,N,P REMOVAL EFFICIENCY AND INFLUNECE OF N(IN) ON C/NTOT (ORG, NO3-, NO2-, NH4+) AND CORRELATION BETWEEN C/N AND N REDUCTION
Riaño et al., 2014	SP	S/L+ C/F	NDN IN 1 TANK	REAL	STUDY INFLUENCE OF DIFFERENT PROCESS ON N-, OM-,ME- AND PATOGENS REMOVAL RATE. COST'S ANALYSIS
Deng et al., 2008	DPS	S/L +AT (UASB)	SBR NDN	LAB 1,2, 3	STUDY DIIFERENT WAY TO ADD CARBON SOURCE FOR THE TREATMENT OF DIGESTED SLURRY
	DPS +ALK				
	DPS+ RW				
Deng et al., 2007	PDPS			REAL	ANARWIA PROCESS DEMOSTRATION
Deng et al., 2006	PDPS	S/L + AT (UASB)	IC-SBR NDN	LAB	STUDY THE APPLICATIONOF ANAEROBIC DIGESTION FOLLOWING BY IC-SBR FOR TREATING SWINE WASTEWATER
Akin et al., 2003	SW	-	SBR NDN +P	BS1	STUDY SBR (ANAEROBIC-ANOXIC-AEROBIC) FOR REMOVING NUTRIENTS (N & P); TEST DIFFERENT SRT
			SBR NDN +P DIFFERENT FEEDING TYPE	BS2	STUDY SBR (ANAEROBIC-ANOXIC-AEROBIC) FOR REMOVING NUTRIENTS (N & P); TEST DIFFERENT SRT
			SBR NDN +P DIFFERENT OPC	BS3	
			SBR NDN +P DIFFERENT OPC	BS4	
Han et al., 2008	SRPS	S/L	SBR FED STEP	BS 1, 2, 3, 4	STUDY ORP-AND PH-TIME PROFILES FOR STEP-STRATEGY FOR OPTIMIZATION N-REMOVAL
Obaja et al., 2003	DPS	AD+ S/L+ D (1:10)	SBR	LAB 1, 2, 3, 4	STUDY THE POSSIBILITY TO TREAT ANAEROBICALLY DIGESTED PIGGERY WASTEWATER. STUDY INFLUENCE OF TEMPERATURE ON NUTRIENT REMOVAL RATE
Obaja et al., 2002	DPS	AD+ S/L+ D (1:10)	SBR	LAB 1, 2, 3, 4	STUDY THE INFLUENCE OF CYCLE LENGHT ON NITRIFICATION RATE REACHABLE
Obaja et al., 2005	DPS	AD+ S/L+ D (1:10)	SBR	LAB 5,6,7	STUDY THE POSSIBILITY TO REMOVING NUTRIENTS FROM DIGESTED PIG SLURRY ISING AS CAROBON SOURCE THE UN-DIGESTED PIG SLURRY
Kim et al., 2004	P (+W)	S/L + D(C/N: 1,22-1,53)	SBR	LAB 1,2	STUDY IF IT POSIBLE TO ESTABILISH AN INTEGRATED SWINE WASTEWATER TREATMNT SYSTEM AND OPERATING STRATEGIES SUITABLE FOR INFLUENT FLUCTUATIONS
	P (+W)	S/L + C/F (C/N 0,45-0,79)+ D			
Magrì et al., 2008	P	S/L	SBR	LAB 1,2	TO ESTABLISH AND CALIBRATE A MODEL FOR THE SIMULATION OF THE NDN PROCESS APPLIED FOR THE TREATMENT OF THE LFPS IN A SBR TYPE REACTOR.
Zhu et al., 2006	P	S/L + D1% TS	$(AN/AX)^2$ SBR	LAB1, 2	STUDY NEW PROCES APPLIED IN SBR WITH USE OF EXTERNAL USE OF CARBON SOURCE
Poo et al. 2005	PW	-	SBR (INTERMITTENT FEED)	REAL	STUDY 3 DIFFERENT CONTROL METHODS BASED ON OXIDATION REDUCTION POTENTIAL (ORP) AND DISSOLVED OXYGEN (DO) FOR DETERMINING AERATION TIME
Lo & Liao, 2007	DPS	AD+ S/L	SBR (AEROBIC ONLY)	REAL	THE TREATMENT RESULTS AND IMPLICATIONS OF THIS FULL-SCALE SBR SYSTEM IN TREATING SWINE WASTEWATER
Won et al. 2011	PW	-	SBR	FARM (PILOT)	STUDY THE OPERATION OF A FARM SCALE SBR OPERATING WITHA REAL TIME CONTROL
Han et al., 2007	SRPS	S/L	SBR FEED STEP	LAB 1, 2, 3	STUDY ORP-AND PH-TIME PROFILES FOR STEP-STRATEGY FOR OPTIMIZATION N-REMOVAL

Table 7 characteristics of substrate used in the studies.

	SLURRY TYPE	TS,i (g/L)	VS,i (g/L)	TSS,i (g/L)	BOD_5,i (g/L)	COD,i (g/L)	TKNi (g/L)	TAN,i (g/L)	TN,i (g/L)	Ptot,i (mg/L)	CODsol,i (g/L)	TOC,i (g/L)	Psol,i (mg/L)	NO_3^-,i (mg/L)	NO_2^-,i (mg/L)	COD/ TKN	BOD/ TKN	BOD/ COD	COD/ TAN	BOD/ TN	BOD/ TAN
Tilche A., 2001	P	6,0	2,6	1,9		6,02	0,68	0,52		91						8,83			11,5		
Wu et al., 2017	P	4,3	3,6					0,68			3,56		31,4	0,00	0,00						
Wu et al., 2017	P	4,3	3,5					0,70			3,39		30,8	0,00	0,00						
Riaño et al., 2014	SP	5,3	2,5			4,5	1,05	1,01		46	3,6		44			4,28			4,47		
Deng et al., 2008	DPS				0,26	1,2		0,75	0,83	46,4								0,22	1,59	0,32	0,35
Deng et al., 2008	DPS +Alk				0,26	1,2		0,75	0,83	46,4								0,22	1,59	0,32	0,35
Deng et al., 2008	DPS+ RW				0,26	1,2		0,75	0,83	46,4								0,22	1,59	0,32	0,35
Deng et al., 2007	PDPS			0,74	0,22	1,2		0,609	0,75									0,19	1,89	0,30	0,37
Deng et al., 2006	PDPS																				
Akin et al., 2003	SW					0,4		0,5						20					0,80		
Akin et al., 2003	SW					0,4		0,5						20					0,80		
Akin et al., 2003	SW					0,4		0,5						20					0,80		
Akin et al., 2003	SW					0,4		0,5						20					0,80		
Han et al., 2008	SRPS				3,0	6,03	1,25	1,2		60,9			44,1	N.D	N.D	4,81	2,41	0,50	5,03		2,52
Han et al., 2008	SRPS				3,0	5,92	1,26	1,21		56,8			45,4	N.D	N.D	4,70	2,37	0,50	4,91		2,47
Han et al., 2008	SRPS				2,9	5,86	1,26	1,22		53,6			50,7	N.D	N.D	4,66	2,33	0,50	4,80		2,40
Han et al., 2008	SRPS				4,1	6,35	1,21	1,06		53			23,2	N.D	N.D	5,25	3,39	0,65	5,99		3,87
Han et al., 2008	SRPS				4,8	6,48	1,22	1,20		71			60,1	N.D	N.D	5,32	3,96	0,74	5,39		4,02
Obaja et al., 2003	DPS					2,26		0,91					88,7						2,48		
Obaja et al., 2003	DPS					2,96		1,19					117,3						2,48		
Obaja et al., 2003	DPS					3,74		1,51					144						2,48		
Obaja et al., 2003	DPS					3,74		1,51					144						2,48		
Obaja et al., 2005	DPS							0,3													
Obaja et al., 2005	DPS							0,3													
Obaja et al., 2005	DPS							0,3													
Kim et al., 2004	P (+W)			0,9	3,2			0,59	0,72	46		0,86	18			1,20		3,70		4,44	5,43
Kim et al., 2004	P (+W)			0,9	3,2			0,59	0,72	46		0,86	18			1,20		3,70		4,44	5,43
Magrì et al., 2008	P	24,9	15,3	17,7	11,4		3,4	2,60		722	8,00		75								4,38
Magrì et al., 2008	P	24,9	15,3	17,7	11,4		3,4	2,60		722	8,00		75								4,38
Zhu et al., 2006	P	10,5	5,4	7,6	3,7	8,8	1,22	0,86		601			39,9	0,00	0,00	7,21	3,00	0,42	10,23		4,26
Zhu et al., 2006	P	10,5	5,4	7,6	3,7	8,8	1,22	0,86		601			39,9	0,00	0,00	7,21	3,00	0,42	10,23		4,26
Poo et al. 2005	PW			1,5	8,0			3,80		20	11,00										2,11
Lo & Liao, 2007	DPS			4,4	3,2		2.1			600							1.52				2,09
Won et al. 2011	PW	5,1	2,5	0,95				1,19				1,62	23,5	1,20							0,00
Han et al., 2007	SRPS				3,0	6,03	1,25	1,20		60,9			44,1	0,00	0,00	4,81	2,41	0,50	5,03		2,52
Han et al., 2007	SRPS				3,0	5,92	1,26	1,21		56,8			45,4	0,00	0,00	4,70	2,37	0,50	4,91		2,47
Han et al., 2007	SRPS				2,9	5,86	1,26	1,22		59,6			50,7	0,00	0,00	4,66	2,33	0,50	4,80		2,40

Table 8 Settings of reactor operation used in the researches considered. W_L: working volume; O: oxic; ANA: anaerobic; ANOx: anoxic; Sed: sedimentation; R: in the reactor; W: raw wastewater; RT: real time; VA: variable; +: at the beginning 1h, after Oxic phase adding external carbon source (25% internal 75% external for exp 1, 50% internal 50% external for exp. 2, 100% internal 0% external for exp. 3), after this phase each 30 min. 6 times addition of external carbon source.

	SLURRY TYPE	V m^3	W_V L	Q_feed L/d	O_2 (Air) L/min	O_2 SET mg/L	HRT d	SRT d	No. Cycle /d	No-Reaction period/cycle	No- ANA/ reaction period	No- O /reaction period	No- ANOx/reaction period	ANA h	Oxic h	ANOx h	Sed h	SST (R) g/L
Tilche A., 2001	P	1250		315000						5,0	-	1,0	1,0	-	2,00	2,00		
Wu et al., 2017	P		8,0	0,6+0,2	0,5		3,3	23,0	3,0	2,0	2,0		2,0	1,25 (1) 0,5 (2)	2,75 (1) 2(2)			7,50
Wu et al., 2017	P		8,0	0,4+0,4	0,5		3,3	23,0	3,0	2,0	2,0		2,0	1,25 (1) 0,5 (2)	2,75 (1) 2(2)			7,50
Riaño et al., 2014	SP	350		37248			9,4		n.a	-	-	12,0	12,0		1,5	0,67		3
Deng et al., 2008	DPS		10,0	3,0	15	3,0	3,3		3,0	1,0		1,0	2 (FILL + IDLE)		4	2	1	3-5
Deng et al., 2008	DPS +Alk		10,0	3,0	15		3,3		3,0	1,0		1,0	2 (FILL + IDLE)		4	2	1	3-5
Deng et al., 2008	DPS+ RW		10,0	3,0	15		3,3		3,0	1,0		1,0	2 (FILL + IDLE)		4	2	1	3-8
Deng et al., 2007	PDPS	1800							3,0	1,0		1,0	2 (FILL + IDLE)		4	2	1	
Deng et al., 2006	PDPS		18,0				3,0	30,0	2,0	1,0		2,0	2 (FILL + IDLE)		3			
Akin et al., 2003	SW		10,0	5,0		>2	2,0	25,0	2,0	1,0	1,0	1,0	2,0	4+1,5 (o/ana)	6	0,5	1,5	3,81
Akin et al., 2003	SW		10,0	5,0		>2	2,0	25,0	2,0	1,0	1,0	1,0	2,0	4 + 1,5 (o/ana)	6	0,5	1,5	3,81
Akin et al., 2003	SW		10,0	5,0		>2	2,0	15,0	2,0	1,0	1,0	1,0	2,0	4 + 1,5 (o/ana)	7	0,5	0,5	3,81
Akin et al., 2003	SW		10,0	5,0		>2	2,0	10,0	2,0	1,0	1,0	1,0	2,0	4 + 1,5 (o/ana)	7	0,5	0,5	3,81
Han et al., 2008	SRPS		8,0	1,8+0,6	1,0		3,3	25,0	3,0	1,0	-	2,0	2,0	-	2,75+2	1,25+1,5		14,7
Han et al., 2008	SRPS		8,0	1,8+0,6	2,0		3,3	25,0	3,0	1,0	-	2,0	2,0	-	2,75+2	1,25+1,5		16,6
Han et al., 2008	SRPS		8,0	1,8+0,6	3,0		3,3	25,0	3,0	1,0	-	2,0	2,0	-	2,75+2	1,25+1,5		14,1
Han et al., 2008	SRPS		8,0	1,2+0,6	1,5		4,4	25,0	3,0	1,0	-	2,0	2,0	-	3,25+2	1,25+2		12,6
Han et al., 2008	SRPS		8,0	1,1+0,4	2,0		5,6	25,0	3,0	1,0	-	2,0	2,0	-	2,75+2,5	1,25+2,5		15,7
Obaja et al., 2003	DPS		3,0	3,0			1,0	11,0	3,0	1,0	-	1,0	1 +1 acetic acid 1,3 g/L	-	4	2+1	<1	2,16
Obaja et al., 2003	DPS		3,0	3,0			1,0	11,0	3,0	1,0	-	1,0	1 +1 acetic acid 1,7 g/L	-	4	2+1	<1	3,39
Obaja et al., 2003	DPS		3,0	3,0			1,0	11,0	3,0	1,0	-	1,0	1 +1 acetic acid 2,16 g/L	-	4	2+1	<1	2,42
Obaja et al., 2003	DPS		3,0	3,0			1,0	11,0	3,0	1,0	-	1,0	1 +1 acetic acid 2,16 g/L	-	4	2+1	<1	2,42
Obaja et al., 2005	DPS		3,0	3,4			0,9	11,0	3,4	1,0			8*		2	1+1+ (6*0,5)	<1	
Obaja et al., 2005	DPS		3,0	3,4			0,9	11,0	3,4	1,0			8*		2	1+1+ (6*0,5)	<1	
Obaja et al., 2005	DPS		3,0	3,4			0,9	11,0	3,4	1,0			8*		2	1+1+ (6*0,5)	<1	
Kim et al., 2004	P (+W)		9,0		2,4		VA	32,0	VA	AUTOMATICALLY CONTROLLED BY THE COMPUTER DEPENDING ON THE VARIABLE PROCESS							0,83	7
Kim et al., 2004	P (+W)		9,0		2,40		VA	32,00	VA	AUTOMATICALLY CONTROLLED BY THE COMPUTER DEPENDING ON THE VARIABLE PROCESS							0,83	7,00
Magrì et al., 2008	P		20,0															
Magrì et al., 2008	P		20,0															
Zhu et al., 2006	P	0,01	8,00		0,72		3,30	20,80	3,00	1,00	1,00	-	2 (with aeration)	1,50	-	2,75+2	0,50	
Zhu et al., 2006	P	0,01	8,00		0,72		3,30	20,80	3,00	1,00	1,00	-	2 (with aeration)	1,50	-	2,75+2	0,50	
Poo et al. 2005	PW		20000		3640				VA	5,00	-	1,00	1,00	-	RT	1,00		
Lo & Liao, 2007	DPS	130,2	108800	22000			4,90		4,00	1,00	-	1,00	-	-	4,50	-	0,75	
Won et al. 2011	PW	24,2	18000				VA			1,00	1,00	1,00	1,00	3,00	pH_RT	ORP_RT	2,00	
Han et al., 2007	SRPS		8,00	0,6+0,2	1,00		3,30	27,00	3,00	1,00	-	2,00	2,00	-	2,75+2	1,25+1,5	0,25	10,00
Han et al., 2007	SRPS		8,00	0,6+0,2	2,00		3,30	27,00	3,00	1,00	-	2,00	2,00	-	2,75+2	1,25+1,5	0,25	10,00
Han et al., 2007	SRPS		8,00	0,6+0,2	3,00		3,30	27,00	3,00	1,00	-	2,00	2,00	-	2,75+2	1,25+1,5	0,25	10,00

Table 9 The removal rate obtained for different studies. W: wastewater, SW: synthetic wastewater; DPS: digested pig slurry; P: pig slurry; PDPS: partially pig digested; BS: bench scale; SRPS: supernatant of raw manure finishing barn pigs; SP: sown; AD: anaerobic digestion; at: anaerobic treatment; S/L: solid liquid separation.

	SLURRY TYPE	%TAN	%TKN	%COD	%BOD	%P	%TSS	%SOL_ COD	%SOL _P	%ST	%SV	%TN
Tilche A., 2001	P	98,8	95,7	94,1		75,8	84,5					
Wu et al., 2017	P	98,7						95,0	95,2	36,2	90,7	
Wu et al., 2017	P	98,3						98,3	68,5	41,2	88,4	
Riaño et al., 2014	SP	98,0	95,5	88,9		15,0		88,9	13,6	11,3	40,0	
Deng et al., 2008	DPS	71,5		86,9		48,8						
Deng et al., 2008	DPS +Alk	92,0		89,4		62,6						
Deng et al., 2008	DPS+RW	98,8		94,3		70,6						
Deng et al., 2007	PDPS	98,3		73,7	93		87,8					
Deng et al., 2006	PDPS											
Akin et al., 2003	SW	69-78							37-58			
Akin et al., 2003	SW	94-96							72-76			
Akin et al., 2003	SW	91-93							95-99,5			
Akin et al., 2003	SW	92-94							92-97			
Obaja et al., 2003	DPS	99,7		64,1					97,8			
Obaja et al., 2003	DPS	98,7		70,3					97,5			
Obaja et al., 2003	DPS	99,7		70,2					97,3			
Obaja et al., 2003	DPS	99,7		70,2					97,3			
Obaja et al., 2002	DPS	99,6							98,4			
Obaja et al., 2002	DPS	99,7							97,3			
Obaja et al., 2002	DPS	99,8							98,6			
Obaja et al., 2002	DPS	99,6							98			
Obaja et al., 2005	DPS	99,9							98,1			
Obaja et al., 2005	DPS	99,8							97,8			
Obaja et al., 2005	DPS	99,8							97,8			
Kim et al., 2004	P (+W)			94,7* as TOC	99,6	50	98,9					96,2
Kim et al., 2004	P (+W)			94,7* as TOC	99,6	50,00	98,90					96,2
Magrì et al., 2008	P			98,00					67			98
Magrì et al., 2008	P			95,00					45			98
Zhu et al., 2006	P		98,70	97,40	100	98,70	99,90			77,50		
Zhu et al., 2006	P		96,30	97,70	100	96,10	99,30			78,80		
Poo et al. 2005	PW	100										
Lo & Liao, 2007	DPS	68,00	66,67		82	50,00		88,64				
Won et al. 2011	PW	100					20,40	90,10	20,10	50,50	66,7	
Han et al., 2007	SRPS		97,00	96,00					78,00			
Han et al., 2007	SRPS											
Han et al., 2007	SRPS											

3.7. Discussion and observations

The wide application of SBR for treating livestock manure and wastewater in general is attested.

SBR is a versatile system that, if well applied, can lead to high nutrient removal efficiency.

Generally, it is very difficult to define which are the optimal process conditions and how characteristics of the "*perfect*" livestock manure must be to ensure the optimal removal of nutrients.

However, analysing the researches on this topic in the scientific literature, it is possible to identify some successful settings and general indications. In the list here below the most important outcomes of the analysis are reported.

- First, the TAN removal yield is always higher than 68% and in the in most searches, reaches value above 98%.

  The COD-removal yields are always high than 65%. The lowest removal rate is observed for solids content.
- P-removal rates can be very high when the biological process is set to achieve the anaerobic conditions.
- Is possible to model biological process applied in SBR even if the substrate considered has very variable characteristics; however, it is necessary to calibrate correctly the model for adapting it to the considered influent. Nevertheless, the inputs needed to get a good output are numerous.
- Applying the fixed-time control strategies, based on the set duration of each phase, the most applied time cycle is 8h.
- Applying the fixed-time control strategies, the denitrification process is applied before the aeration one in order to exploit the content of carbon content in the slurry.
- The feed step strategy resulted a good fixed control strategy for improving removal rate reachable.
- The real time control strategy based on ORP and pH allows reaching the complete removal of nitrogen with the minimal waste of energy.
- All slurries used in the analysed researches before to be fed the biological reactor are undergone to pre-treatment process. In fact, is necessary to remove solid, mostly the inert part of them to not overload biological system.

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4. Monitoring of real scale treatment plants for nitrogen removal and for the enhancement of effluents

Different treatment plants have been monitored in order to attest their operational state and their achieved removal efficiency.

The aim of monitoring campaigns was mainly finalized to individuate possible improvements for NDN (nitrification/denitrification) plants.

In the following paragraphs monitoring activities, main results and observations of the monitoring campaign are reported.

4.1. Description of different plants

4.1.1. Plant TP

The TP plant's farm is located in Torre Pallavicina, in the province of Bergamo. In the farm are bred about 8000 piglets and 3100 sows divided into various structures. In fact, as shown in Figure 11, the farm is portioned in different sheds where different categories of pigs are separated for health reasons: the yellow line delimited sows' area and blue line delimited the area where grower pigs and are located.

Figure 11 Plant overview with different zone (Google maps ®).

The farm owns its biological treatment plant (indicated in Figure 11 with red line), that works with a standard NDN (nitrification-denitrification) process. The flow scheme of the plant is represented in Figure 12.

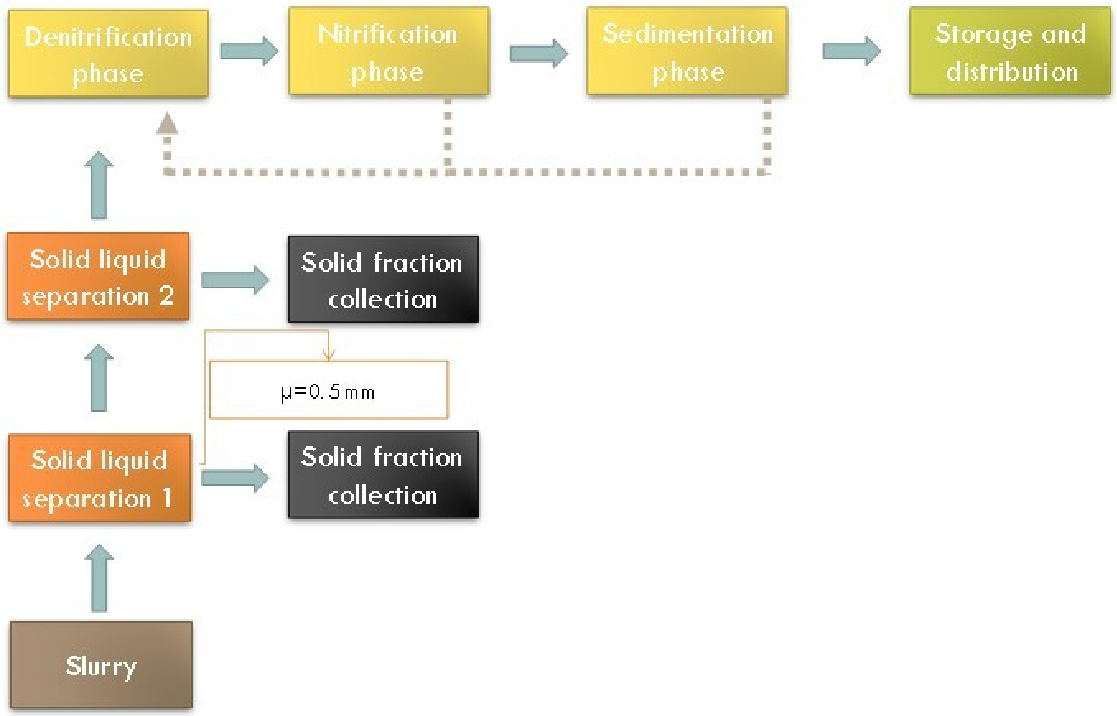

Figure 12 Flow scheme of TP plant. μ represents the theoretical removal rate.

The plant's influent, collected from various sheds, is made of a mixture of sewage, manure, water, litter and other components.

The first step of slurry treatment consists in slurry's transfer from a collection channel to a collection tank; from here, an installed pump conveys stored slurry to mechanical treatment that consists in a solid liquid separation through a rotary sieve aim to removing coarse solids. The liquid fraction obtained, using pump is send in another tank that has a volume equal to 57,4 m^3. Then, liquid fraction that is continuously mixed, is pumped by submerging pump to the further flotation treatment (Figure 13). This kind of chemical-physical process allows removing the smallest particles size from slurry using different chemical products and an air bubble system. In this plant, aluminium sulfate ($Al_2(SO_4)_3$) as coagulant and polyelectrolyte as flocculants are used. Mixing the inlet slurry with $Al_2(SO_4)_3$ the negative charges of colloidal particles are destabilized and can be re-aggregated, thanks to polyelectrolyte, in bigger light flocks that can be separate through air bubble. In TP plant, coagulant and flocculants are added to the slurry before Venturi's system that mixed the blend of slurry and chemicals with air bubbles; the aerated slurry mixed with chemicals enters subsequent tank, where the flakes that have formed bringing to the surface and are removed by superficial chain scraper. The floating material accumulated on the surface of the tank is removed (Figure 13,d), while the liquid fraction continues to the following biological treatment.

The effluent of floating system enters through a concrete sluice gate the first biological phase: denitrification phase occurs in a bigger concrete tank equipped with 3 mixers, positioned in 3 different central points in the middle of the tank. Mixers blend and homogenize the inlet flow with the activated sludge already present in the tank, allowing the transformation of nitrates into a N_2 (denitrification phase).

Figure 13 Floatation system of plant TP: a) chemical dosing, b) superficial chain scraper, c) liquid fraction obtained and d) the “solid” fraction obtained.

Nitrates arrive in denitrification thank through a recirculation flow (Figure 14a). As usual, the denitrification phase occurs before the nitrification phase in order to allow to denitrifying bacteria (that are heterotrophic) to use the readily degradable organic matter as source of carbon. In fact, if that phase will be worsened next to nitrification phase, an external source of carbon is requested.

The hydraulic retention time of denitrification (and whole biological process) is imposed by the operation of the floatation system and by the gate used for re-circulate the flow: in fact, the flow entering the biological treatment is constant but can be adjust through the volume of sludge pumping at in the separation treatment and so, to the duration of flotation system phase and the quantity of re-circulated flow is imposed by the high of the weir (Figure 14a).

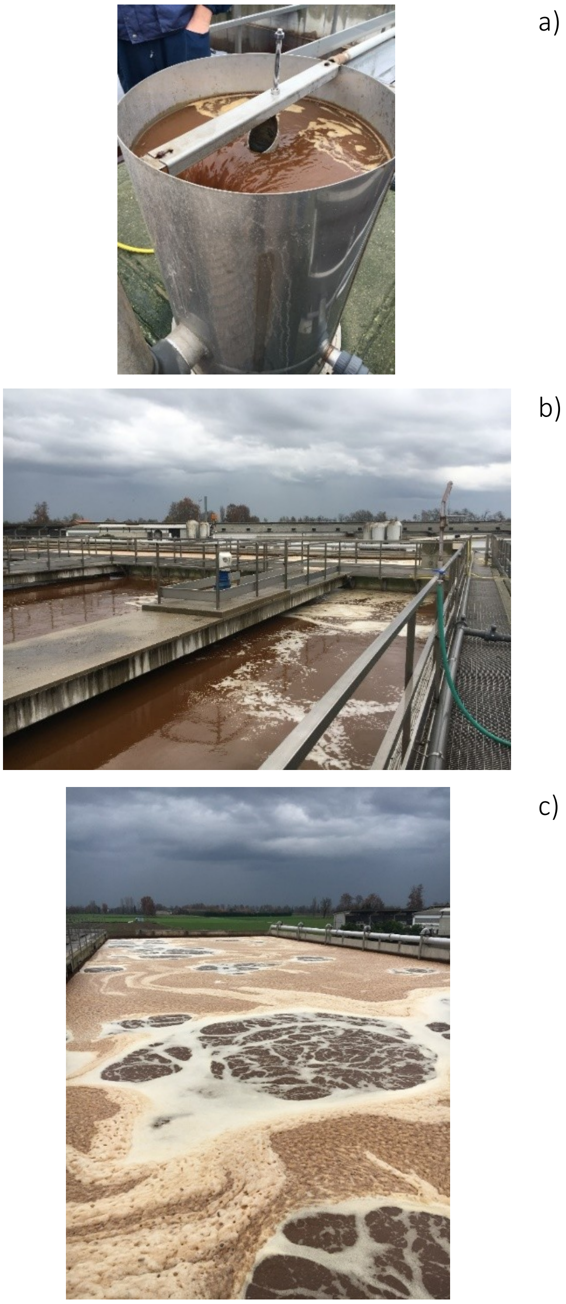

Figure 14 a) the recirculation apparatus, b) mixing tanks where denitrification occurs and b) aeration phase for nitrification process.

Unfortunately, the control system of the plant has a lot of problem, and even if the pump flow enters the biological phase, the working time of the system is not correctly managed and controlled.

Finally, the denitrified effluent passes into the following concrete tank through a weir. Here, through a blower, air diffusion system dissolves the necessary amount of oxygen in the water in order to establish nitrification of the ammonia nitrogen present in the effluent (Figure 14,c), which is oxidized to nitrite and then to nitrate that are re-circulated to the denitrification phase. Also in this case, the automation system does not foresee any control on airflow furnished.

The treated effluent than arrives into the sedimentation thank, always by gravity, where the solid and activated sludge are separated. The clarified effluent where finally accumulated in a storage tank and will be used and distribute to the land as fertilizer according to imposed regulation.

Plant TP, as also explained before, does not have a well-appointed and complete control system. Despite all the process is automated, tanks are not equipped with any type of probe (e.g. pH, RedOx, O_2 etc.) and in any point of the piping system is not present the flow control instrumentation. This fact makes process control difficult to monitoring and not allow a correct operation of all system. This plant, given its continuous operation, has been selected to monitor its performance, and, if possible, it was chosen for trying to individuate some possible improvements to apply.

4.1.2. Plant TO

Farm TO is a fatteners pig farm located in the province of Brescia. In this farm, the pig slurry is treated in a SBR (sequencing batch reactor) system for biological nitrogen removal before its reuse in agriculture.

In the Figure 15 a flow scheme of the plant is reported.

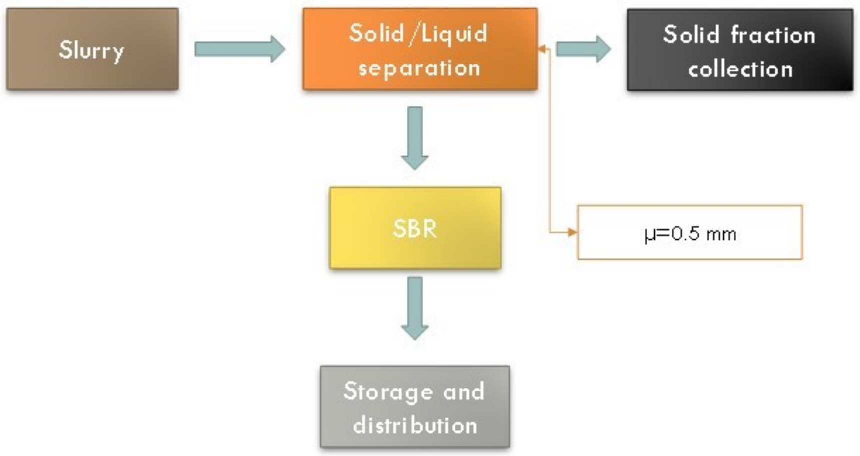

Figure 15 Block Scheme of TO plant. μ represents the theoretical removal rate.


a)

b)

Figure 16 SBR tank. In the Figure a) it is possible to see the pipe that connect the SBR’s outlet with a storage tank, where the effluent is collected and stored for further utilization. In Figure b) a particular of inlet and aeration system.

The clarified slurry enters the reactor during the filling phase without any type of mixing or aeration (static filling) since the required level. Then, alternatively, aeration phase and mixing phase are provided. Also for this plant, the usual operation of biological treatment foresees a pre-denitrification phase, and a subsequent nitrification phase. Aeration, which allows ammonia nitrification, occurs through a Venturi system (Figure 17a) that transferred 460,0 Nm^3/h of air to the sludge, ensuring an oxygen transfer of 21,0%. Two blowers of 540,0 m^3/h of total capacity, guarantee the air supply. Before entering in the tank, the installed system provides the well mixed of air bubble and slurry.

The mixing phase instead occurs thought the same system of aeration, with the difference that in this case, the air is not mixed with the sludge, but the slurry is re-circulated through injection system to ensure a good homogenization of the active sludge present in the tank and the fresh clarified slurry.

Finally, is foreseen a sedimentation phase that allows the separation of the activated sludge and heavy flocks to reach the bottom of the tank from the clarified slurry. At the end of the sedimentation phase, the clarified sewage is discharged through the overflow (Figure 17b) in the final accumulation tank. That mechanism is one of the most delicate points of the system, because is necessary guarantee the good quality of the effluent avoiding the discharge of biomass or the dragging of suspended solids.

The removal of the activated sludge accumulated in the bottom of the tank, is carried out at the same time of the loading of other fresh slurry.

In the plant TO the control of operations is completely automated, using PLC (programmable logic controller), with whom the duration of the various treatment phases and the operation of all electromechanical equipment (compressors, mixers, pumps etc.) are managed. Biological tank is also provided with a level transmitter (ultrasonic type), an instrument for foam level control, a RedOx probe and dissolved oxygen control probe.

All these instrumentations allow managing all the phases of operation and, in case of malfunction, intervening for correction. This plant, given its continuous operation and its useful control system, has been selected to monitor its performance.


a) b)

Figure 17 Particular of SBR aeration system Venturi a) and the overflow for effluent's outlet b).

4.1.3. Plant NA

That plant, located in the municipality of Dovera (CR), is managed by an S.r.l. composed by six associate companies, of which four produce cereals and two deal with animal husbandry. That plant is more complex (e.g. foresees more stages of treatment) in comparison with the TP and TO plants. In fact, all slurries produced in the different farms are treated for energy recover in an anaerobic digester before to be treat for nitrogen removing. That solution is very widespread in Po valley also for the economic support received from the Italian state for biogas production (D.Lgs 387/03 and D.Lgs 28/2011).

A scheme of plant is reported in Figure 18 and in Figure 19 the block scheme of plant is shown.

Besides the slurry produce by the associated farms, approximately 67 m^3/d of external slurry (predominantly cattle and pigs' slurries) and 27 t/d of biomass (corn and shredded silo) are fed to the plant by means of truck. The inlet slurry and other biomass entering the plant are treated for energy recovery in a 630 kW Thöni plant. The first part of the plant consists of two anaerobic digesters: the feeds of anaerobic digester 1 is delivered into a pre-tank (if necessary, the feed of the tank can be ground with the use of a shredder). In that digester, biomass is also supplied. Digester 2 only receives bovine / swine (mainly bovine) sewage.

After a digestion process, the digestate from the digester 1 is sent to the digester 2. The digestate from the energy recovery (digester 2) is further treated for nitrogen removal before to be spread. The treatment provides, as a first step, the mechanical separation of coarse material with helical separator (0,75 mm): the resulting liquid fraction is sent to intermediate storage tank and then should undergo further separation by centrifugation.

Figure 18 Overview of Plant NA: 1 and 2 are two inlet points, where the slurry is charge and enters in the plant; 1,2: are two inlets points; 3: storage of digestate after AD process; 4: liquid fraction of digestate; 5:solid fraction of digestate; 6 SBR; 7 final storage tank.

This kind of treatment is focused to removal of fine particles allowing sending to biological nitrogen removal a slurry with a low load of carbonaceous compound that would over-load the biological process unnecessarily.

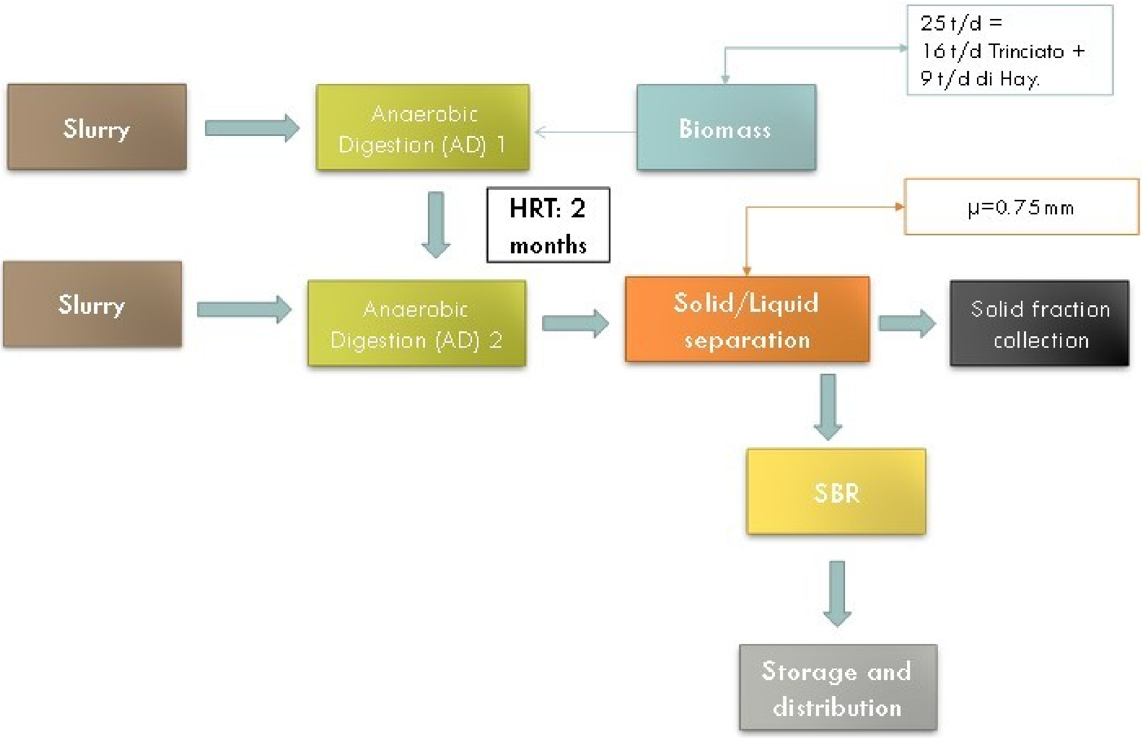

Figure 19 Blok Scheme of NA plant. μ represents the theoretical removal rate.

Regrettably, during a period of monitoring, centrifuge was not in use.

The solid fraction separated by helical separator is accumulated in storage slab and the liquid fraction in a collection tank (point 4 in Figure 18), from which it is drawn to feed the SBR where nitrification-denitrification process occurs. Concrete tank for biological treatment has a diameter and high equal to 12,5 m and 6,0 m respectively, to which correspond a geometrical volume of 736,3 m^3 and an operative volume equal to 674,9 m^3. During monitoring period, SBR does not operate at full capacity. As explain in paragraph 3.1 also in this SBR an alternative phase of mixing and aeration occur for transform nitrogen in a non-polluting form. For the duration of biological nitrogen removal (nitrification-denitrification), ORP, O_2, pH and temperature are monitored using different probes. The plant does not have an autonomous data acquisition system. In Table 10 is reported fluxes data of plant NA, furnished by the plant's owner.

Table 10 Data of flows processed in plant NA.

Date	m^3 TQ/d	m^3 SBR/d	m^3 solid fraction/d
October	65,70	30,95	-
November	63,89	56,98	6,90
December	62,96	59,00	3,95
January	67,32	62,61	4,7
February	70,46	64,67	5,58
March	67,62	62,33	5,27

This system, although ready to be used, in the period designated for monitoring, was not 100% functional for various management and performance problems. For this reason, the monitoring was aimed not at verifying its performance, but also at detecting which critical issues it presents and how its correct functioning could be assisted.

4.1.4. Plant VA

The plant VA treats pig slurry from a fattening pigs' farm. It is located in Camisano (Cremona); in Figure 20 is reported the overview of the plant, and in Figure 21 the flux scheme of that plant.

The plant does not provide energy recovery (i.e. anaerobic digestion, AD), but consists in a mechanical separation of solid fraction (medium and big ones) and subsequent nitrogen removal applied in sequencing batch reactor (SBR), where a nitrification/denitrification reaction takes place.

Figure 20 The overview of the plant VA. 1: inlet points; 2: tank for preliminary storage; 3: liquid fraction after solid/liquid separation; 4: solid fraction; 5 SBR plant; 6,7,8: storage tanks.

In particular, the raw slurry collected from the different sheds arrives by gravity in a pre-storage tank (Figure 22,a) that is equipped with a pump and a mixer. That pump is manually switch on and feeds a vertical helical separator (0,5 mm) (Figure 22,b) that allows a separation of liquid fraction from solid particles. Solid fraction is accumulated in storage slab; instead, the liquid fraction is accumulated in a storage tank before to be send to the SBR plant. The flow rate fed to the separator will be equal to 2,4 m^3/min (from data read on the flow meter installed on the pipe that feeds the separator).

SBR has a diameter of 13,5 m and high of 5 m that correspond to a geometrical volume equal to 715,7 m^3 and an operational volume equal to 644,1 m^3

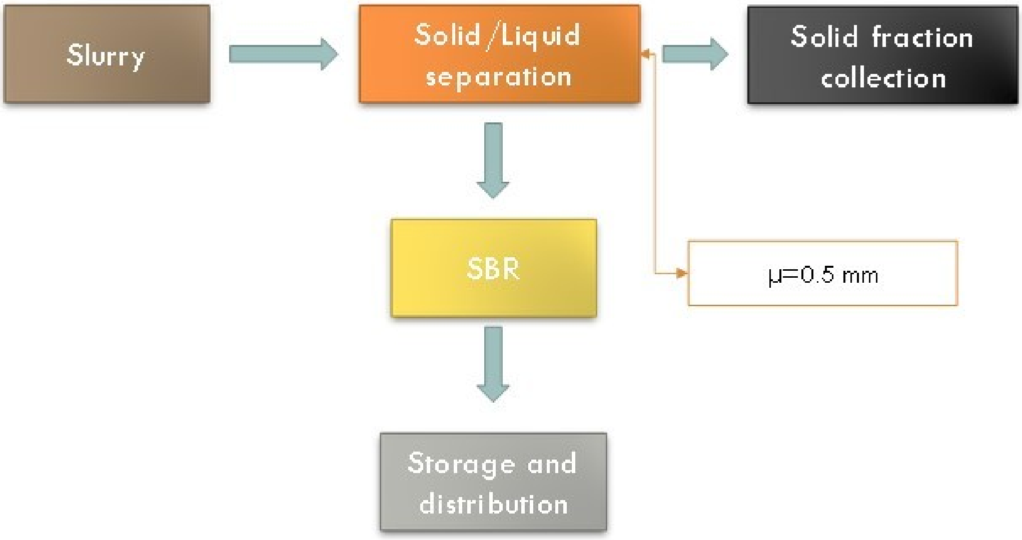

Figure 21 Scheme of VA plant. µ represents the theoretical removal rate.

SBR system, although ready to be used, in the designated period for monitoring was not 100% functional for various management and performance problems. For this reason, also for that plant, the monitoring was aimed not at verifying its performance, but at identifying which critical issues it presents and in how correct functioning could be reached.

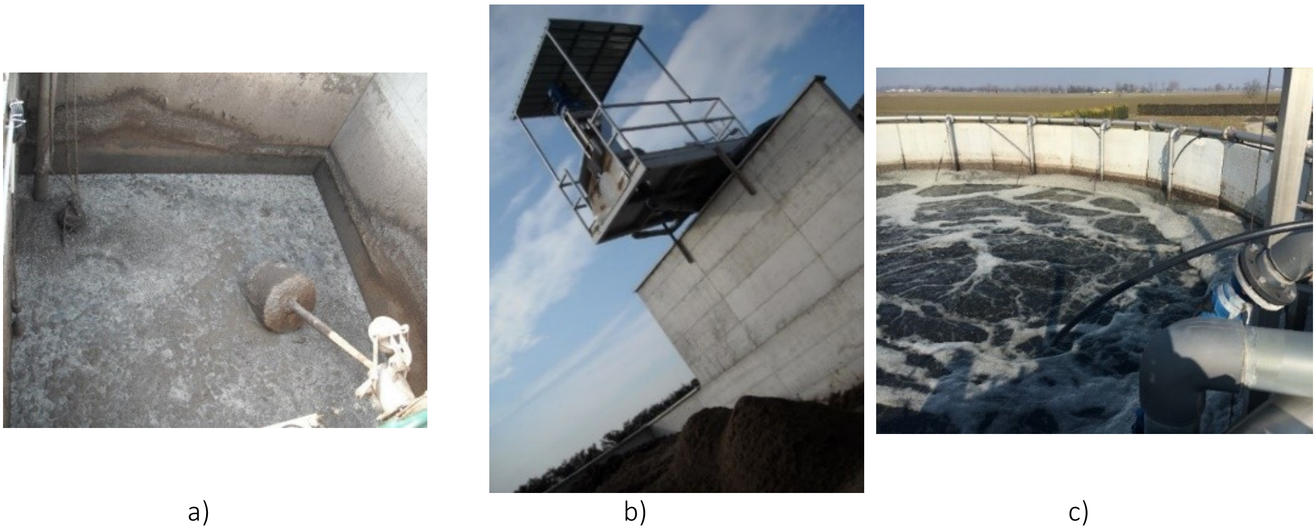
a) b) c)

Figure 22 a) the pre-storage tank of VA plant; b: the solid/liquid separation system; c: SBR tank, during aeration phase.

4.1.5. Plant RR

Plant RR is located in Credera Rubbiano (Cremona). That plant treat slurry from dairy farms (sewage and manure). In Figure 23 is reported the overview of the plant while in Figure 24 is reported a flow scheme of the plant. The particularity of that plant is that all slurry that enter in arrived through a system of underground pipes and only a small part (solid fraction) arrive to the plant by trucks. This allows to reduce consumption of fuel and, consequently, environmental impact.

Theoretically, 80 m^3 of slurry and 13 m^3 of manure should be conferred to the plant; however, only 5 m^3 of manure are treated during the monitoring period.

Also in that case, as for the plant NA, the plant is not only focused on the nitrogen removal but also to energy recovery.

First, the slurry is treated in anaerobic digester. The scheme provides that, at the exit of the digester, (that has not had an overflow), a pump brings the slurry directly to the solid/liquid separator. The pipe that collects digester with solid/liquid separation unit is furnished with a flow meter.

After the solid fraction is removed from the digestate, the liquid fraction is fed to a sequencing batch reactor system equipped with a level sensor (piezoelectric sensors), an ultrasonic sensor, probe for monitoring O_2, and RedOx.

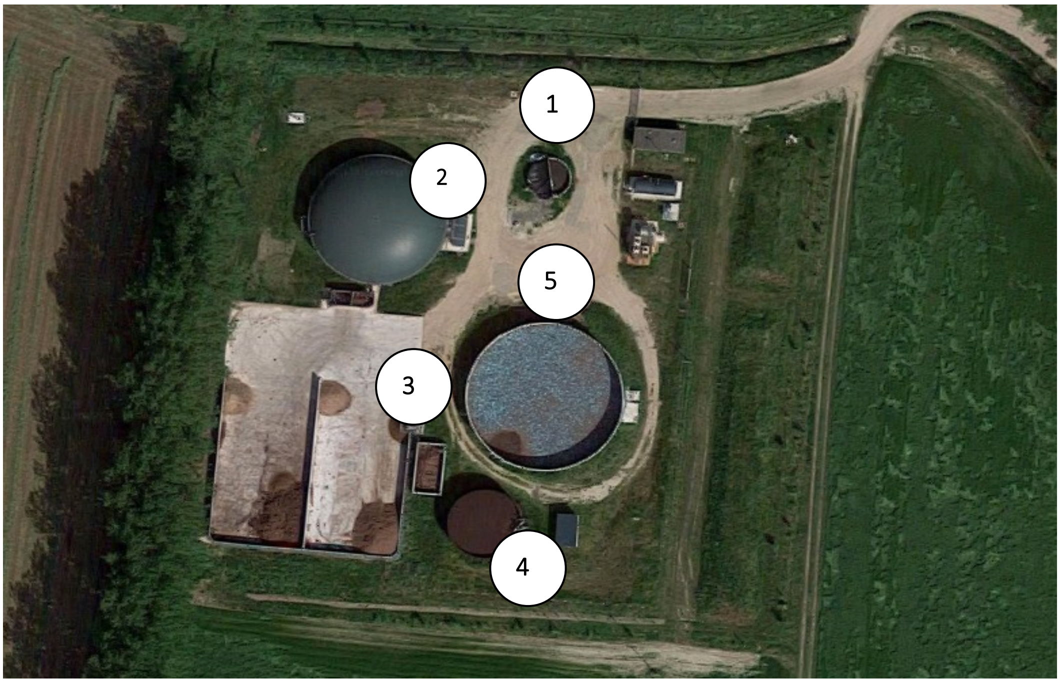

Figure 23 The overview of the plant RR. 1: inlet points and preliminary storage; 2: anaerobic digester; 3: S/L separation and accumulation; 4: SBR plant; 5: final storage tanks.

The SBR cycle length varying based on slurry collected: an aeration and mixing phase are always foreseen for nitrification/denitrification aims. The plant does not have an autonomous data acquisition system. For this plant, we do not have dimensional parameter. This plant, given its continuous operation, has been selected to monitor its performance.

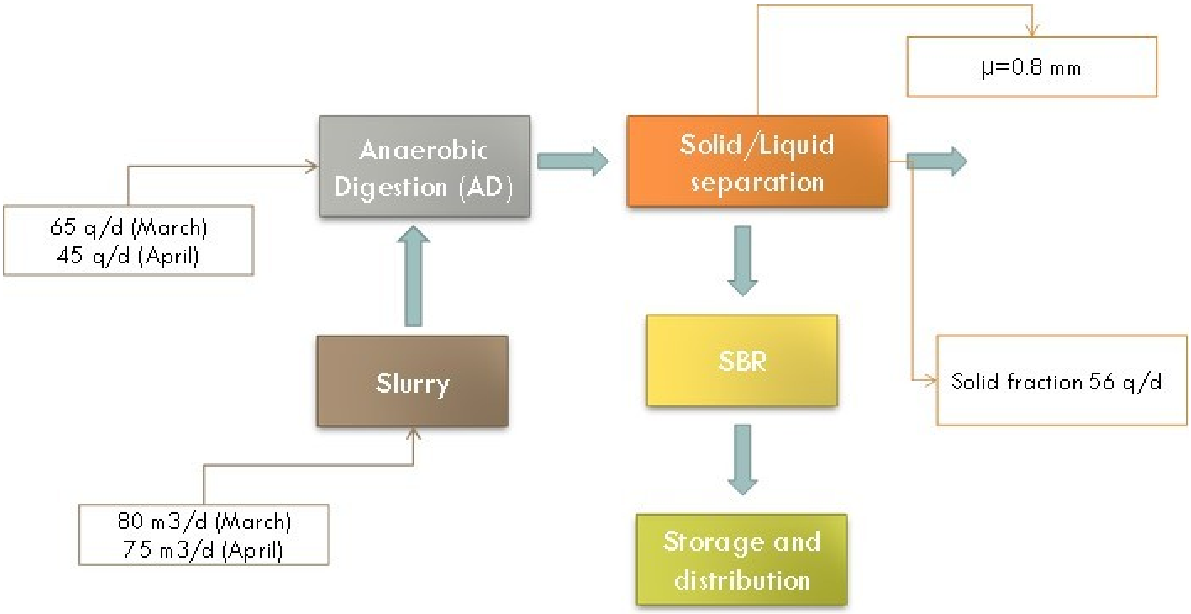

Figure 24 Scheme of Plant RR. μ represents the theoretical removal rate.

4.2. Materials and methods

4.2.1. Monitoring plan for all treatment plants

In Table 11 are reported data about monitoring plan for all plants: the length of monitoring period, the selected sampling points for each plant, and analysis carried out for each sampling point.
The sampling points have been chosen in order to have an a

Where possible and foreseen, also the parameter logged by the plants are recorded and analysed. The short period of monitoring for plant NA, VA, RR is also linked with a lot of management problems and many malfunctions that forced the plant operators to stop them and try to solve the problems.

4.2.2. Analytical methods

Each sample was characterized for Total Kjeldahl Nitrogen (TKN), electrical conductivity (EC), total ammonia nitrogen (TAN), pH, dry matter content (TS) and volatile solid (VS), which were determined using Standard procedures (APHA, 2014). For plant TO also RedOx potential have been measured. For plant TP and TO also NO_2^- + NO_3^- are measured for sampling withdrawn from biological process (APHA, 2014).

Table 11 Characteristics of monitoring plan for all considered plants.

	Sampling period	Sampling points	Lab Analyses
TP Plant	5 months: from January 2017 to June 2017	- INLET: the liquid fraction after the first treatment finalized to solids removal; - LIQUID FRACTION: the liquid fraction after removal of small particles (flotation); - SOLID FRACTION: the solid fraction of flotation treatment; - DENITRIFICATION: an homogenize sample of denitrification slurry (sampled in 3 different points in the tank); - NITRIFICATION: an homogenize sample of nitrification slurry (sampled in 3 different points in the tank); - OUTLET: the treated slurry from the sedimentation tank.	- pH; - EC Electrical conductivity; - TKN Total Kjeldahl Nitrogen; - TAN total ammonia nitrogen; - ST total solid; - SV volatile solid; - RedOx (for plant TO)
TO Plant	5 months: from June 2017- to September 2017	- INLET: the liquid fraction after the first treatment finalized to solids removal; - OUTLET: the treated slurry.	
NA Plant	March-April 2016	- IN 1: INLET 1 (RAW SLURRY); - IN 1: INLET 2 (RAW SLURRY); - DIG: outlet of anaerobic digestion 2; - SLIQU: liquid fraction after solid/liquid separation; - SSOL: solid fraction after solid/liquid separation; - SBR: sample taken from the biological reactor; - OUT: treated slurry, withdraw from a final storage tank	
VA Plant		- IN: Inlet point 1, slurry withdraw from the sluice gate of the piggery. - IN*: Inlet point 2, withdraw from the pre-storage tank - SLIQU: liquid fraction after solid/liquid separation; - SSOL: solid fraction after solid/liquid separation; - OUT: treated slurry, withdraw from a final storage tank	
RR plant		- IN: raw slurry; - DIG: DIG: outlet of anaerobic digestion 2; - SLIQU: liquid fraction after solid/liquid separation; - SSOL: solid fraction after solid/liquid separation; - SBR: sample taken from the biological reactor; - OUT: treated slurry, withdraw from a final storage tank	

4.3. Results of monitoring campaigns

4.3.1. Plant TP

Regarding TP plant is necessary to know that during all monitoring period, regardless the unstopped plant operation, a lot of management problem occurred: as the blocking of some equipment, the malfunction of scum dosing system etc.

As explain, the absence of any acquisition system or PLC, is makes almost impossible know all events and problems that happened in the plant and the quantity of treated influent.

Nevertheless, thanks to preliminary tests, the pump flow that feeds separation treatment was calculated, and resulted equal to 31 m^3/h. However, due to the lack of information about other data's flow, it would not be possible to do a correct mass balance of the plant, to measure and check the flow of the plant and, consequently, to calculate the removal rate reachable.

The only things that it can be done is monitored the concentration of the nutrients along the treatment line and calculate the potential removal treatment reachable considering the project flow (Table 12).

Table 12 Data of flows processed in plant TP (project data).

SAMPLING POINT	VOULME (m^3)	FLOW (m^3/d)
RAW SLURRY	28400	77,81
INLET	27200	74,52
LIQUID FRACTION	21200	58,08
DENITRIFICATION	21200	58,08
NITRIFICATION	21200	58,08
OUTLET	21200	58,08
SOLID FRACTION	6000	16,44

In Table 13, is reported average and standard deviation of chemical-physical characteristics of each sample withdrawn for each sampling point in plant TP.

Table 13 Monitoring result obtained for each sampling points of plant TP.

Sample		NH_4 (g/kg)	TKN (g/kg)	ST%	SV/ST%	EC (mS/cm)	pH
TP_INLET	A	0,69	1,14	1,00	67,76	7,03	7,45
	SD	0,24	0,37	0,36	4,36	1,84	0,23
TP_LIQUID FRACTION	A	0,63	0,72	0,38	45,40	6,70	7,26
	SD	0,22	0,33	0,17	9,86	1,74	0,28
TP_DENITRIFICATION	A	0,11	0,21	0,48	44,28	4,41	7,00
	SD	0,06	0,07	0,23	14,07	0,65	0,71
TP_NITRIFICATION	A	0,10	0,29	0,60	53,33	4,35	6,88
	SD	0,06	0,08	0,10	3,94	0,65	0,73
TP_OUTLET	A	0,11	0,17	0,34	30,77	4,42	7,24
	SD	0,06	0,04	0,06	5,03	0,65	0,56
TP_SOLID FRACTION	A	1,04	3,67	6,98	75,03	8,77	7,12
	SD	0,38	1,33	1,61	3,48	8,11	0,30

In addition, in Figure 25, are reported the graphs representing the trend of value observed during the sampling period. The same consideration has been done for solid content (Figure 26). Regarding pH, for all sampling points, the value is around 7, an optimal pH for the for the establishment of biological processes (Sommer et al., 2013).

The concentration of nitrogen compounds in the inlet flow has an inconstant trend (for both the ammonia and TKN). This trend can be explained by the non-uniformity of the characteristics of faces: in fact, the sewage comes from a breeding of sows and fattening pigs. The latter, due to the variation of the feed during the fattening period, produce different waste with the consequence that the sewage entering the plant does not have constant characteristics. Nevertheless, the most plausible explanation of these variations is connected to precipitation: the homogenization tank from which the slurry is taken before being sent to the separator, receives rainwater, which causes a dilution effect on the treated sewage. Moreover, at the TP plant, a considerable amount of water is used to wash some areas of the plant or with the purpose of defusing the sample: all this quantity of water is poured into the slurry accumulation tank and results in an alteration of the characteristics of the slurry itself.

Downstream of the solid/liquid separation treatment, as expected, the concentration of nitrogen content of both TKN and ammonia decreases, even if the latter has a lower drop. In fact, assuming that there are no nitrites and nitrates (since the conditions for which nitrification processes cannot occur), the separation process allows removing fine and coarse particles in which organic nitrogen is concentrated, and that therefore through this treatment removed; small particles are often associated to nutrients, especially N and P. In particular, about the 70,0% of un-dissolved N and P is related to particle size 0,45-250 µm (Masse et al., 2005).

Considering the concentrations' values obtain during sampling campaign, the removal rate reachable (based on average concentration measured and project flow) through solid/liquid separation, regarding the solid content and nitrogen content are respectively 28,2% (TAN), 50,8% (TKN) and 70,5% (ST), 80,2% (SV).

Lacking the analysis of the specific content of nitrates and nitrites (the conducted analysis regarding the oxidized nitrogen returned a total value and not specified) as well as of suspended solids (index of the biomass present in the tank), it is not possible to elaborate deep considerations regarding the performance of the nitrification and denitrification phase. In addition, no analyses are been conducted for the recirculation flow. Nevertheless, it is possible to calculate the removal rate reachable always considered the average concentration value. The solid content in denitrification and nitrification samples are higher than that one observed in the liquid fraction: this is due to the sampling; in fact, due to mixing and aeration of the tank, the influent, with a low solid content is mixed with activated sludge, that rises this parameter. The removal rates reachable through biological process is equal to 87,6% (TAN) and 59,1% (TKN).

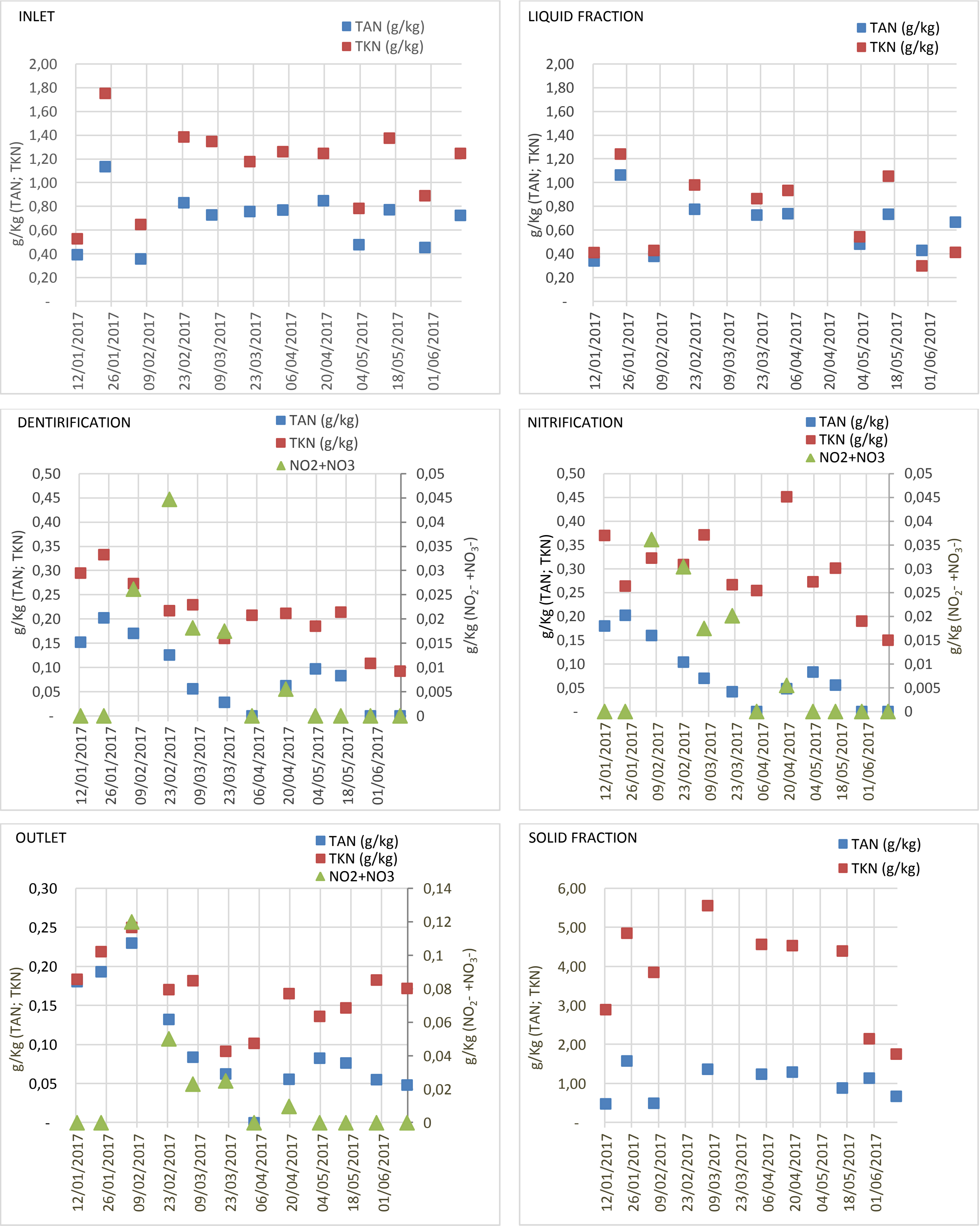

Figure 25 Patterns of ammonia and TKN content during sampling campaign for each sampling point in TP plant.

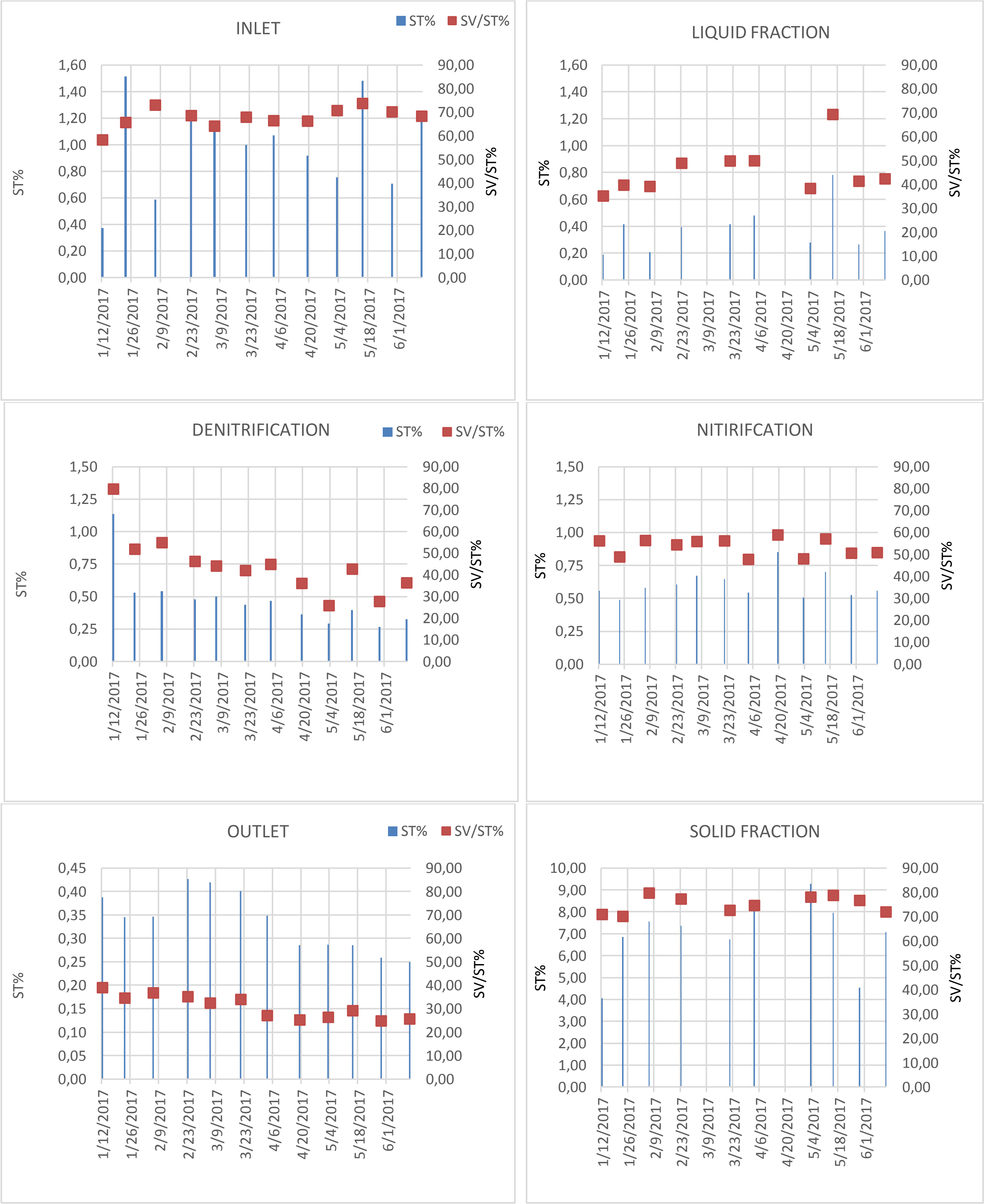

Figure 26 Pattern of total solids content and the percentage of volatile solids on total solids, content during sampling campaign for each sampling point in TP plant.

The absence of nitrite and nitrate in the nitrification tank it may be due to the absence of a correct nitrification process; in fact, that process should transform the ammonia nitrogen entering the system into nitrites and nitrates, which then feed, through the recirculation flow, the denitrification tank.

Overall, considering all the process the removal rates reachable are equal to 28,2% (TAN), 50,8% (TKN) and 70,5% (ST), 80,2% (SV).

Considering TAN and TKN parameters, the characteristics of the outlet is comparable with those one observed in nitrification tank; regarding solid content, since the effluent is withdrawn after sedimentation, the concentration is the lowest observed.

Regarding the characteristics of solid fraction, it is interested to underline that, right now, in the plant, the solid fraction (ST= 7%) (combined with the solid fraction comes from the pre-treatment of separation-please see paragraph 4.1.1), doesn’t be used and needs to be sent for further treatment.

The solid fraction obtained downstream of the flotation cannot be defined solid, but may be pumped to the anaerobic digestion for energy recovering or to a phosphorus recovering treatment, in order to move on the positivity, the energy balance and cost of the plant.

Although the data obtained seems to attest high efficiency of the denitrification/nitrification treatment plant TP, it is necessary to consider that the influent is much diluted and that a part of ammonia can be leave the tank through ammonia emissions and not thanks to biological removal. In addition, the continuous management problems of the plant and the lack of conscious management of the system, make these results only indicative and not representative. Therefore, it is necessary to install a control system and to control always the correct operation of the plant.

4.3.2. Plant TO

The plant TO was on operation during all monitored period. In Table 14 are reported the average and the standard deviation of concentration data collected for that plant.

The point “RIC” represent characteristics of the recirculation flow that is not monitored always during sampling campaign.

Regarding this plant, the PLC installed allows checking and measuring not only RedOx and DO but also an inlet flow. The inlet flow changing and decreasing during the monitoring campaign. In the Figure 27 is possible to individuate 3 periods: during the first one (red line) the inlet flow was on average equal to 36,8 m^3/d; during the second period (yellow line) was 18,7 m^3/d and it was characterized by a high variability, and, during the last period, equal to 5,01 m^3/d. That last phase (26/6/2017 - 25/9/2017) coincides with the end of the production cycle of pigs and their sale and to the consequent drop of slurry production.

Table 14 Monitoring result obtained for each sampling points of plant TO.

Sample		N-NH_4 (g/kg)	TKN (g/kg)	ST%	SV/ST	pH	EC (mS/cm)	RedOx
IN	A	1,40	1,89	1,17	41,44	7,89	15,97	-202,75
	SD	0,80	1,02	0,78	10,91	0,28	6,13	189,81
RIC	A	0,21	0,33	0,58	40,46	6,95	7,84	59,60
	SD	0,16	0,22	0,21	6,11	0,69	3,28	124,72
OUT	A	0,51	0,68	0,90	40,18	7,34	10,64	-52,90
	SD	0,56	0,56	0,32	6,46	0,93	4,33	154,73

The so imposing variation of the flow fed to the plant, that have been design for treat 45 m^3/d, certainly has repercussions on the performance and consumption of the same; despite the versatility of these types of SBR systems, if the volume in the tank remains the same, feeding a lower flow rate will affect SRT and HRT. When the inlet flow is equal to 5 m^3/d the system was not be considered to working correctly. In addition, since level in the tank is very low during this period, the oxygen probe was not immersed in the sewage; this, in addition to returning false data (with the oxygen data> of 9 mg/L), can compromise the functioning of the probe. Due to its operational setting, it is possible to consider that the inlet flow is equal to outlet flow; so, the removal rate during the monitored period can be calculated.

Concerning O_2 content, is interesting to underline the differences between the 3 periods. In fact, as shown in Figure 27, the concentration of dissolved oxygen during first period concentration of O_2 is lower always than 2 mg/L (maximum value registered in the plant). Usually, in order to guarantee nitrification process at least concentration 1,5-2 mg/L of dissolved oxygen is request.

In some studies, the request oxygen concentration reaches also 3 mg/L (please see Chapter 3). Therefore, with this dissolved oxygen concentration it is possible that a correct nitrogen oxidation did not occur. Nevertheless, it also probable that, since RedOx potential assumed variable values, the probe is dirty and for this reason does not allow reading the correct values. Other option is that the rising in RedOx potential observed is related to an absence of proper biomass in the sludge able to nitrify (SRT not adequate). In that same period, in fact, the RedOx potential had a “conventional trend”, with minimum value observed below zero, corresponding to the non-aeration phase, and higher than zero when blower is turned on.

During the second period, the value has a different behaviour: in fact, the maximum value of oxygen concentration is higher. Nevertheless, due to the discontinuity plant operation, with strongly variable flow rates, the detected data cannot be considered as an average operating condition.

Figure 27 The inlet flow measured by plant's instrument (flow meter) (a), the DO concentration measured by on oxygen probe (b) and the RedOx registered during sampling campaign (c).

The same conclusions can also be reached in period 3, where, as mentioned, the level of operation of the tank was lower than that one observed during normal operation. In fact, since the oxygen probe was not correctly immersed in the liquid (for the lower level) the recorded oxygen values are always higher than 5 mg/L. Regarding the RedOx potential, the maximum and minimum values never fall below the value 0, since the quantity of air supplied is the same and the volume of slurry in the tank is lower, the oxygen transfer was more efficient. However, the energy consumption of the plant grew a lot, making inefficient and expensive a system that, if properly managed, allows reaching the required removal rates with a limited cost.

In order to better evaluation of the behaviour of oxygen concentration, RedOx and results obtained, it is necessary to analyse a single SBR's cycle. In Figure 28 is reported an example of pattern of RedOx and dissolved oxygen during one day (20/05/2017) belong to first period: in particular is reported the parameters' variation during 6 hours.

The trend of RedOx and oxygen are not comparable with the conventional graphs obtained in a sequencing batch reactor (please see Chapter 3). Nevertheless, the minimum in oxygen graph, correspond to a lowest RedOx potential value. It is possible to individuate the duration of cycle equal to 40 minute, very short time compare with those one applied usually in that type of plant (see Chapter 4).

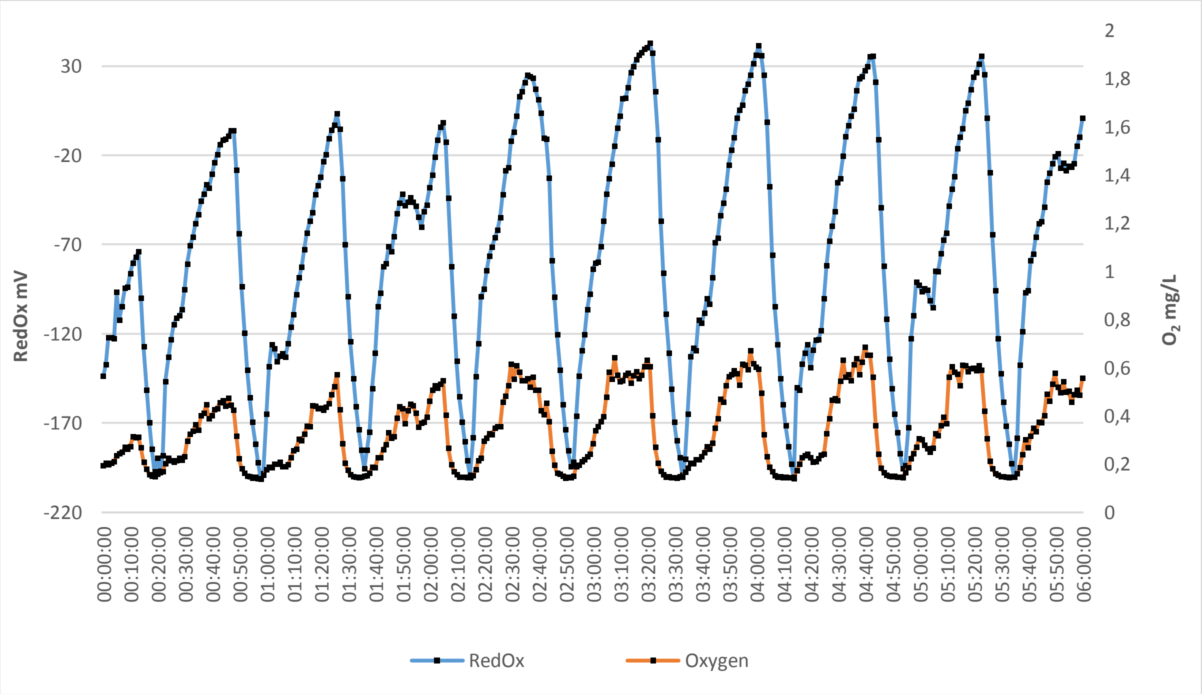

Figure 28 Pattern of RedOx and oxygen detected during one operation day (20/05/2017) in TO plant.

In Figure 29, is reported the concentrations of nitrogen compounds and solid compounds along all monitored period in plant TO.

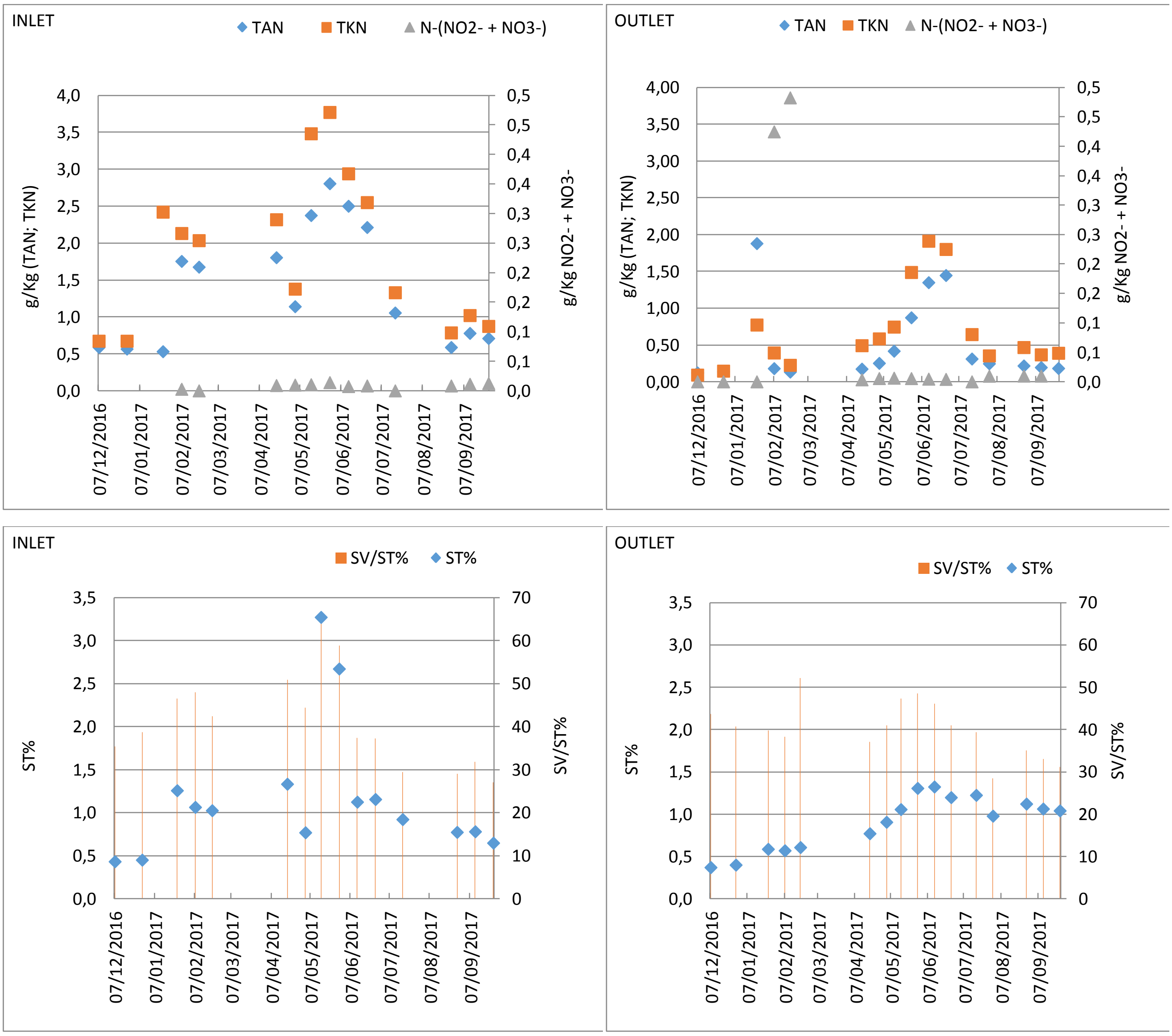

Figure 29 Path of TAN and TKN concentration and solids content during sampling campaign for each sampling point in plant TO.

The inlet concentration of nitrogen compounds varying between 0,53 g/L and 2,8 g/L (TAN) and 0,67 g/L and 3,77 g/L concerning TKN. Relating to solid content the minimum value observed for total solid and volatile solid were 4,3 g/L and 1,5 g/L and the maximum are 32,7 g/L and 23,8 g/L respectively. Also in this case, the faces of the pig changing during the fettering period. In addition, also in this plant, there is not a cover for the different tanks, and so the rain (copious in some days) may diluted the slurry. At the output, the concentration observed of solids has a more constant trend, and the nitrogen content did not show a constant trend. This can be a result of the change in the inlet flow quantity and quality.

The mass balance and the removal rate definition were done for 3 periods. In Table 15 and 16 are reported results of detected concentrations and removal rates obtained.

Average removal rates are calculated using an average flow 21,4 m^3/d.
As well in that plant, data about the activated sludge was not be present. For this reason, it cannot be possible to understand if the denitrification process occurs correctly.

Table 15 Chemical-physical characteristics for three different periods.

		N-NH_4^+ (g/kg)	N ($N0_2^-$+NO_3^-) (g/kg)	TKN (g/kg)	pH	EC (mS/cm)	T°C	ST%	SV/ST
PERIOD 1	IN	1,57	0,01	2,18	7,81	16,60	8,34	1,34	46,69
		0,84	0,01	1,06	0,27	6,59	8,91	0,92	9,28
	OUT	0,55	0,13	0,68	7,65	8,53	8,21	0,79	43,48
		0,62	0,22	0,59	0,80	4,01	8,82	0,35	4,94
PERIOD 2	IN	2,21	0,01	2,55	8,37	24,05	0,00	1,15	37,29
		-	-	-	-	-	-	-	-
	OUT	1,44	0,00	1,80	8,71	17,78	0,00	1,19	40,99
		-	-	-	-	-	-	-	-
PERIOD 3	IN	0,78	0,01	1,00	7,97	12,40	0,00	0,78	29,34
		0,20	0,00	0,24	0,18	2,21	0,00	0,11	1,98
	OUT	0,24	0,01	0,46	6,53	13,71	0,00	1,09	33,98
		0,05	0,00	0,13	0,35	0,36	0,00	0,10	4,54

Also in this case, the nitrogen removal rate is higher than 60% but, considering that part of ammonia can leave the system as ammonia emissions, it can be useful to check the amount of ammonia leave the system and how is biologically removed.

Table 16 Removal rates obtain in each period, and the average one.

	N-NH_4^+ REMOVAL RATE%	TKN REMOVAL RATE %	ST REMOVAL RATE %
PERIOD 1	65,0	68,6	41,2
PERIOD 2	34,7	29,4	-3,6
PERIOD 3	69,1	54,3	-40,6
AVERAGE	64,0	64,1	23,1

For this plant, that is well furnished of control system and any type of probe, optimal operation would be expected.

4.3.3. Plant NA

In Table 17 are reported concentration data (average and standard deviation) obtained for plant NA.

As first step and where all data are available, a mass balance is conducted (Figure 30).

For plant NA, it was hypothesized that all the produced digestate (NA-DIG) is sent to the following phase aimed at biological nitrogen removing (NA-SBR).

Table 17 Monitoring results obtained for plant NA.

Sample (A)		NH_4^+ (g/kg)	TKN (g/kg)	ST (%)	SV (%ST)	pH	EC
NA-IN1	A	1,19	2,99	7,37	80,56	7,05	12,64
	SD	0,11	0,47	1,07	0,21	0,06	2,25
NA-IN2	A	1,23	2,96	7,53	81,08	7,09	16,03
	SD	0,08	0,13	0,11	0,63	0,13	0,69
NA-DIG	A	2,01	4,06	6,49	70,37	7,93	13,65
	SD	0,01	0,07	0,04	1,57	0,03	0,37
NA-SLIQ	A	1,99	3,94	5,15	67,00	8,13	22,57
	SD	0,03	0,13	0,11	0,13	0,08	0,53
NA-SBR	A	2,05	3,94	5,00	65,25	8,10	22,20
	SD	0,00	0,06	0,03	1,84	0,01	2,62
NA-OUT (STOC)	A	1,91	3,49	4,78	64,91	7,84	22,32
	SD	0,11	0,06	0,03	0,30	0,01	0,58
NA-SSOL	A	1,62	5,94	22,64	77,82	-	-
	SD	0,04	0,32	0,56	7,99	-	-

The data provided by plant operators attest that 67,6 m^3/d of slurry are fed into the plant (NA-IN) and that 62,3 m^3/d of digested slurry reaches the SBR tank. If the input data (NA-IN) represents the total flow of both the treated slurry and the added biomass, anaerobic digestion feed was equal to 67,6 m^3/d.

Considering that, the 62,2 m^3/d treated by the SBR came from mechanical separator (NA-SLIQU), the separate liquid of the helical separator is equal to 62,3 m^3/d.

A mass balance for nitrogen content was also conducted (Figure 30). It was supposed that N concentration (as TKN) in silage and hay were equal to 8,5 gTKN/L and 2,0 gTKN/L respectively. For TKN balance, data collected in March are used, because represent a correct operation of the plant.

The samples collected from two different inputs of the NA plant have similar characteristics in terms of dry matter and volatile solids, as well as for the nitrogenous components. The concentration of ammonia after the anaerobic digestion increases but remains almost unchanged after the SBR treatment until storage, before distribution. As observed for TKN, an increase in concentration downstream of anaerobic digestion was observed for the addition of biomass.

The plant's operators attested that on average 25 m^3/d of chopped and hay (as also reported in the mass balance) entered the plant. The data about concentration of the TKN related to storage, compared with the characterization of the slurry taken in the SBR, were slightly lower for probable phenomena of emissions (losses) that occurred. Samples of solid fraction had ammonia nitrogen and TKN values in line with the characteristic data of separate solid. The total solids concentration shows how the process of anaerobic digestion causes a decrease in the percentage of total solids (hydrolysis phase, and catabolic reactions).

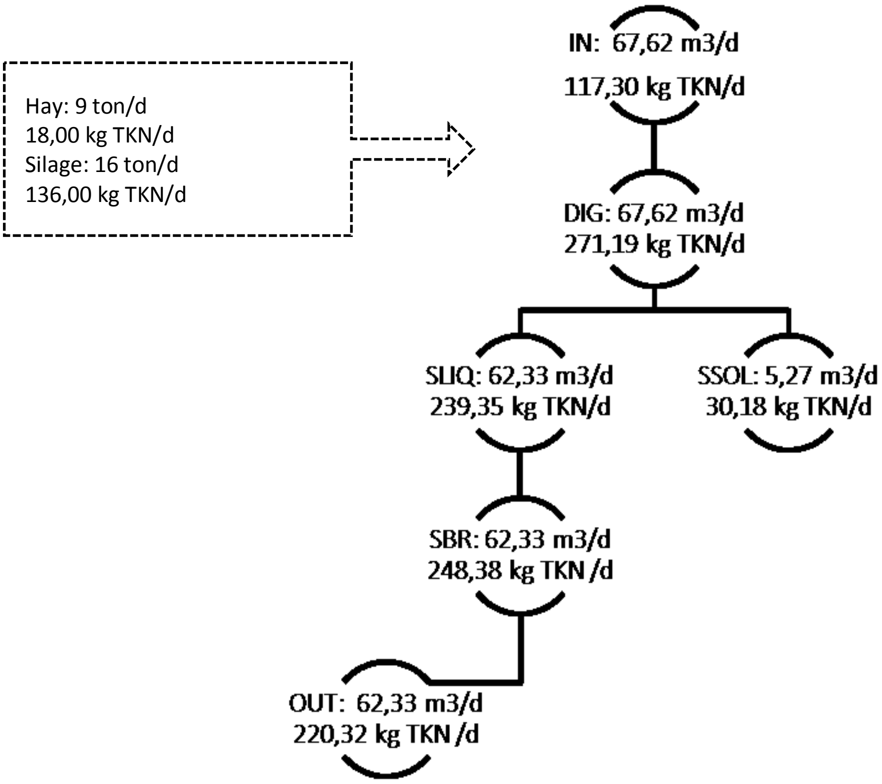

Figure 30 Mass balance and N (TKN) balance in March for plant NA.

For the plant NA, a verification of the separation efficiency in terms of total solids was determined. Considering the concentrations of solids detected with laboratory analysis and the flow rates declared by the plant, it is possible to verify the percentage of solids entering the SBR plant. The results obtained are reported in Table 18.

Table 18 Verification of solid separation efficiency for Plant NA.

SEPARATION PHASE VERIFICATION	Qin,S/L separator [m^3/d]	67,6
	Cin,ST concentration in the feed of separator [%]	6,5
	Cout,ST concentration in the outlet of separator [%]	5,2
	Cout,sol,concentration in the outlet of separator (solid fraction) [%]	22,6
	Qout,liq [m^3/d]	62,3
	Qout, sol [m^3/d]	5,27
	µ vol %	7,8%
	µ solid %	26,9%
	Min,ST [kg/d]	4388,5
	Mout,sol ST [kg/d]	1192,9
	Mout, liqu ST [kg/d]	3210,0

The separation efficiency in terms of volume was equal to 8%. This data is not so high but in line with those ones reported in literature (5-20%). Nevertheless, the total solids content in the separator effluent was very high, and this fact can affect negatively the proper functioning of the SBR. It would be advisable to start to use, as also foresees by the plant operators, the installation of a machine that removes the total solids as the centrifuge. Moreover, with some simplifying hypotheses, the aeration system has been sized necessary to reach nitrogen removal levels > 60%. The results obtained are shown in Table 19.

Table 19 Dimension of air supply for Plant NA.

Energy consumption for air supply	Qin inflow of SBR [m3/d]	62,3
	Cin,N TKN concentration [gN/kg]	3,9
	Cin,N TAN concentration[gN/kg]	2,0
	Qin,TKN [kg/d]	245,6
	Qin,TAN [kg/d]	124,0
	TAN/TKN	0,5
	µ nitrogen removal required [%]	60%
	Qout, TKN [kg/d]	98,2
	TAN that is necessary remove in the SBR [kg/d] **	147,3
	$kgO_2/g\ N\text{-}NH_4^+$;	4,3
	O2 required for ammonification [kgO_2/d]	626,2
	Oxygenation yield for a bubble system kgO_2/kWh (0,8-1,3)	1,1
	kWh	596,4
	kW machine for a functioning of 14 h day	42,6

**hypotheses: due to the TAN/TKN rate in the slurry, is < 60%, the hypotheses is that part of N_{org} in the sludge is mineralize during the process.

It is essential to consider that the nitrogen to be removed for nitrification is ammonia, since it is this component that can be oxidized to nitrates and subsequently transformed into molecular nitrogen.
This consideration is done for all plants here described.

The data recorded in the phase sequence reactor (SBR) confirm what the system has declared: the SBR is not working. In fact, the chemical / physical parameters upstream and downstream have not changed.

4.3.4. Plant VA

The Table 20 here below shows the data collected for the different samples for plant VA.
The only data about the flow treated slurry by the plant is calculated starting from the number of pigs owned and the total amount of slurry produces during a pervious observed period. In fact, considering that in 6 months, the slurry production was about 6000 m^3 TQ.
Considering a volume separation efficiency of 10% (hypothesis), the SBR system treats 5400 m^3 TQ. Considering 30 days per month, an average of 30,0 m^3/d can be obtained. Since the reading of the flow meter that feeds the helical separator, was equal to 2,4 m^3/min (from data read on the flow meter), it is possible to calculate the time of operation of the feed pump equal to 12,5 min/ day.

Table 20 characteristics of slurry during monitoring period of plant VA.

Sample		pH	EC	NH_4^+ (g/kg)	TKN (g/kg)	ST (%)	SV (%ST)
VA-IN	A	7,81	35,23	3,17	3,99	2,83	50,45
	SD	0,12	0,35	0,28	0,39	0,57	2,80
VA-IN*	A	7,61	33,25	3,41	5,44	6,92	63,90
	SD	-	-	-	-	-	-
VA-SILQ	A	7,75	33,45	3,53	5,19	5,56	59,49
	SD	0,21	1,37	0,18	0,28	0,81	0,01
VA-OUT	A	8,26	24,07	1,57	1,82	1,48	36,51
	SD	-	-	-	-	-	-
VA-SSOL	A	-	-	2,06	8,29	30,90	74,71
	SD	-	-	0,74	0,99	2,66	6,54

In the plant VA, there is a discrepancy of the definition points “INLET” considered for sampling. In fact, having detected during first campaign an inequality between the characteristics of the incoming slurry (B-IN) (especially in terms of total solids) taken downstream of the pig-shade and the separate liquid (B-SLIQU) sampled at downstream of the solid/liquid separator, it was decided to define as “INLET” the slurry withdrawn from the accumulation tank located upstream of the separator (B-IN *). Actually, due to longest accumulation period, the characteristics of total solids and volatile solids can change during storage.

Moreover, during the second sampling it was decided to not take the “outlet” sample (B-OUT) because the prolonged storage of the wastewater (>4 months) meant that the chemical-physical characteristics were no longer representative of that point.

For the plant NA, a verification of the separation efficiency in terms of total solids was determined. Considering the concentrations of solids detected with laboratory analyses and the flow rates declared by the plant, it is possible to verify the percentage of solids entering the SBR plant.
The results obtained are reported in Table 21.

Table 21 Verification of solid separation efficiency for Plant VA

SEPARATION PHASE VERIFICATION	Qin,S/L separator [m^3/d]	33,3
	Cin,ST concentration in the feed of separator [%]	6,92
	Cout,ST concentration in the outlet of separator [%]	5,56
	Cout,sol, concentration in the outlet of separator (solid fraction) [%]	30,9
	Qout,liq [m3/d]	29,97
	Qout, sol [m3/d]	3,33
	µ vol %**	10,0%
	µ solid %	28%
	Min,ST [kg/d]	2304,36
	Mout,sol ST [kg/d]	1028,97
	Mout, liqu ST [kg/d]	1664,83

** The value was hypothesized, having no flow data available for the two fractions (solid / liquid)

The total solids content in the effluent of the helical separator can be decreased with the use of a more efficient separator. This device can also meliorate the quality of slurry enters SBR, and in that way it could raise the removal yields of nitrogen.

Also for this plant, with some simplifying hypotheses, the aeration system has been sized necessary to reach nitrogen removal levels> 60%.
The results obtained are shown in Table 22.

In this case, the SBR in this system was not in operation. Samples of separate solid have values for ammonia nitrogen and TKN in line with the characteristic data of separate solid.

Table 22 Sizing (preliminary) of air supply for Plant NA.

Energy consumption for air supply	Qin inflow of SBR [m^3/d]	30,00
	Cin,N TKN concentration [gN/kg]	5,19
	Cin,N TAN concentration[gN/kg]	3,53
	Qin,TKN [kg/d]	155,70
	Qin,TAN [kg/d]	105,90
	TAN/TKN	68%
	h nitrogen removal required [%]	60%
	Qout,TKN [kg/d]	62,28
	TAN that is necessary remove in the SBR [kg/d]	93,42
	$kgO_2/g\ N\text{-}NH_4^+$;	4,25
	O2 required for ammonification [kgO_2/d]	397,04
	Oxygenation yield for a bubble system kgO_2/kWh (0,8-1,3)	1,05
	kWh	378,13
	kW machine for a functioning of 14 h day	27,01

4.3.5. Plant RR

The Table 23 here below shows the concentration’s data collected for the different samples for plant RR.

Observing the trend of ammonia concentration downstream of anaerobic digestion there is an increase of 0,5 g/L, and undergoes a sharp decrease during the SBR process. However, in the sample taken from the storage tank, the concentration rises again. This observation leads to consider a possible by-pass of slurry for the separation treatment to the storage tank. This deduction can also be corroborated by TKN data, and relative to solids, for which, in storage, an increase in concentration can be noted. The SBR process, as mentioned, seems to bring about a substantial reduction of TAN. However, since there is no information regarding the phase in which the sampling was carried out no detailed considerations can be made. In addition, as for plant TP, TO, NA and VA, no data about activity of activated sludge are defined and verified. Also for this reason, any consideration done about nitrifying and denitrifying bacteria activity is not possible. Visually, it was understood that the sampling was withdrawn during the sedimentation or denitrification phase, since there was no air blowing.

Since in plant RR the flow rates have changed during the monitoring period, the reported mass balances are distinguished for the 2 months considered (Figure 31).

In order to make the mass balance of the plant, a nitrogen concentration (TKN) of 2,0 g/was assumed for the manure.

Table 23 Characteristics of slurry during monitoring period of plant RR.

Sample		pH	EC	NH_4^+ (g/kg)	TKN (g/kg)	ST (%)	SV (%ST)
RR-IN	A	7,02	14,00	1,52	3,53	8,87	80,81
	SD	0,10	4,70	0,06	0,14	0,12	2,61
RR-DIG	A	7,96	16,55	1,94	3,46	5,31	69,85
	SD	0,17	4,73	0,18	0,20	0,48	1,36
RR-SLIQ	A	8,02	21,38	1,93	3,40	4,17	62,84
	SD	0,08	2,05	0,13	0,25	0,31	4,31
RR-SBR	A	8,19	13,06	0,45	1,52	3,22	57,36
	SD	0,42	0,83	0,18	0,45	0,67	6,17
RR-OUT (STOC)	A	7,65	17,64	1,54	2,85	4,04	60,01
	SD	0,17	0,88	0,38	0,40	0,11	5,40
RR-SSOL	A	-	-	1,67	5,54	26,37	89,72
	SD	-	-	0,04	0,22	1,62	0,29

In April, in fact, as the flow rates were lowered, the mass balance is different (Figure 31), and with it also the efficiency of the SBR that was equal to 67%.

In April, during the plant's monitoring, it was considered the possibility that the slurry inside the SBR was settling for a long time, being the level quite low and the foam completely absent. Moreover, always during sampling, it was clear that there was a displacement of slurry's flow starting from an intermediate storage tank to an unknown point, since the level in the SBR did not change during the handling of that liquid flow (encountered by the noises). So, it was plausible to think that the digested slurry (or the liquid separated), was partly by-passed. From the obtained data, it is possible to calculate a nitrogen removal efficiency (as TKN) of the RR plant in the month of March: this is equal to 43,2%. Although the data obtained seems to attest to a high efficiency of the SBR, it is necessary to consider that at the output of the plant we find an effluent having TKN much higher than that detected in the SBR. Moreover, as already stated, although the efficiency of the plant seems high, considering storage as a final point, the efficiency drops considerably from the "theoretical" 43% and 67% (in the months of March April-respectively), to a real 20,2% 12,3% (in the months of March April-respectively). The variability of the data, which is more marked for the samples taken downstream of the liquid separator, in the SBR and downstream of the same, can be traced back to the different feeding of the plant in the two months in which the sampling was carried out.

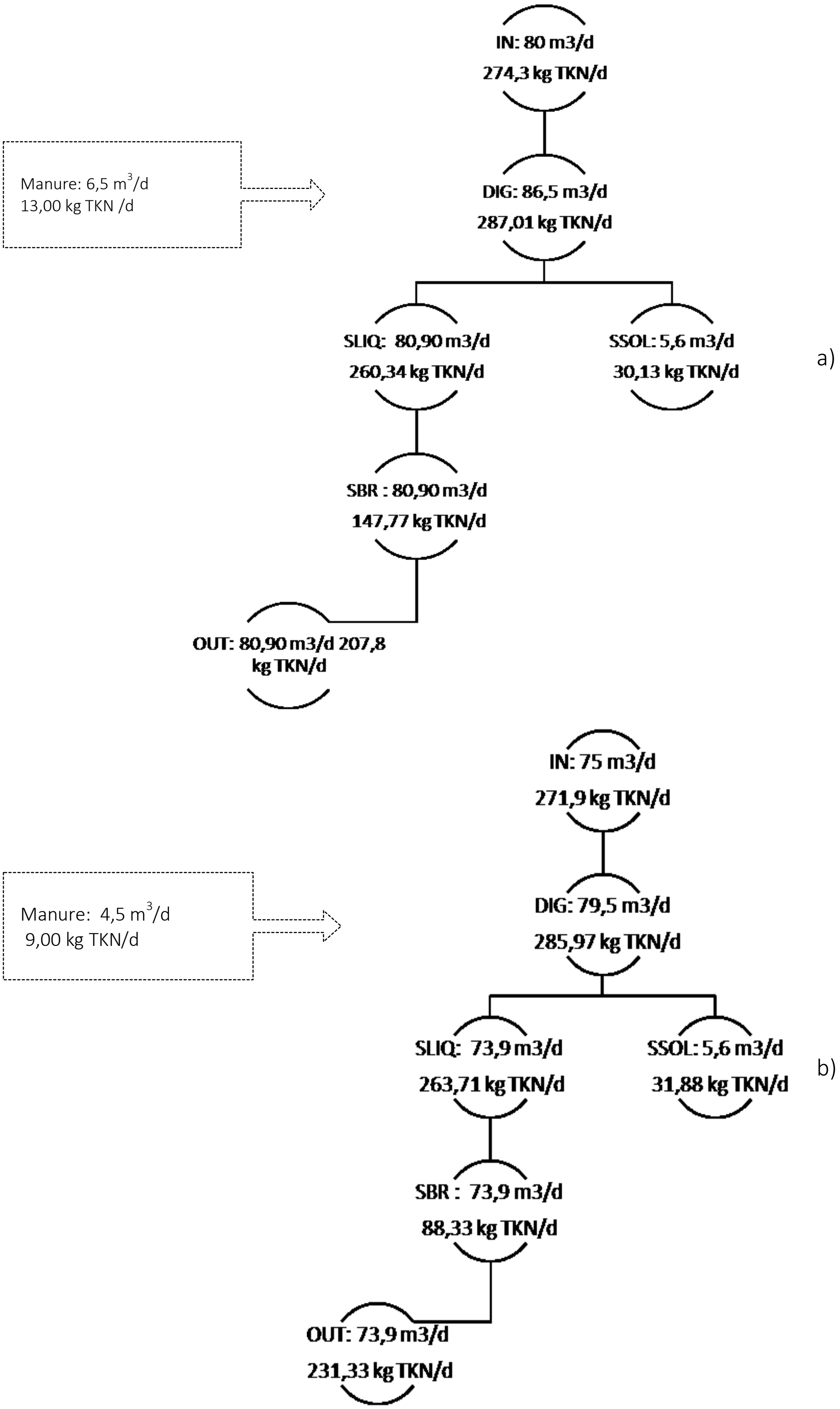

Figure 31 Mass balance and N (TKN) balance in March (a) and April (b) for plant RR.

Considering the concentrations of solids detected and the flow rates declared by the plant, it is also possible to verify the percentage of solids entering the SBR plant as be done for other plants. The results obtained are reported in Table 24.

Also for this plant, with some simplifying hypotheses, the aeration system has been sized necessary to reach nitrogen removal levels> 60%. The results obtained are shown in Table 25.

Table 24 Verification of solid separation efficiency for Plant RR

SEPARATION PHASE VERIFICATION	Qin,S/L separator [m3/d]	86,5
	Cin,ST concentration in the feed of separator [%]	5,3
	Cout,ST concentration in the outlet of separator [%]	4,2
	Cout,sol, concentration in the outlet of separator (solid fraction) [%]	26,4
	Qout,liq [m3/d]	80,9
	Qout, sol [m3/d]	5,6
	µ vol %**	6,5%
	µ solid %	27,0%
	Min,ST [kg/d]	4593,2
	Mout,sol ST [kg/d]	1476,4
	Mout, liqu ST [kg/d]	3373,5

Table 25 Dimension of air supply for Plant RR.

Energy consumption for air supply	Qin inflow of SBR [m3/d]	80,90
	Cin,N TKN concentration [gN/kg]	3,40
	Cin,N TAN concentration[gN/kg]	1,93
	Qin,TKN [kg/d]	275,06
	Qin,TAN [kg/d]	156,14
	TAN/TKN	57%
	µ nitrogen removal required [%]	60%
	Qout,TKN [kg/d]	110,02
	TAN that is necessary remove in the SBR [kg/d]	165,04
	kgO$_2$/g N-NH$_4^+$;	4,25
	O2 required for ammonification [kgO$_2$/d]	701,40
	Oxygenation yield for a bubble system kgO$_2$/kWh (0,8-1,3)	1,05
	kWh	668,00
	kW machine for a functioning of 14 h day	47,71

4.4. Treatment plants' evaluation and possible improvements

The general conclusion provided by these monitoring campaigns is that in each plant it is possible to introduce some improvements in order to meliorate the achievable nutrients removal results, the optimization of energy consume and consequently, the economic saving.

In Table 26 are reported a summary of main features of monitored plants.

Table 26 Main features of monitored plants.

				BIOLOGICAL PROCESS		
PLANT	Type of System	CONSIDERED PERIOD	PRE-TREATMENT	TAN %	TKN %	ST %
RR	SBR	ALL PERIOD	Anaerobic Digestion + Screw press separator	20,2	16,2	3,1
VA	SBR	ALL PERIOD	Screw press separator	54,0	66,5	78,6
NA	SBR	ALL PERIOD	Anaerobic Digestion + Screw press separator + Centrifuge (under construction)	4,0	11,4	7,2
TP	NDN	ALL PERIOD	Screw press separator + Coagulation/Flotation	82,5	76,4	10,5
TO	SBR	PERIOD 1	Screw press separator	65,0	68,6	41,2
		PERIOD 2		34,7	29,4	-3,6
		PERIOD 3		69,1	54,3	-40,6

Specifically, regarding the plant TP, is evident the need to upgrade the civil structures; in addition, plant TP needs the installation of proper instrumentation that consent process control.

The quality of the inlet flow reached through the solid/liquid separation process (flotation) and detected during sampling campaign are similar to some case of study described in the literature (Chapter 3) where an appropriate removal rates are reached. In that case the biggest problem regards the management of the plant: with a proper investment (such as the installation of at least 2 flow meter in-line on the pipe that feeds the accumulation tank and on the outlet pipe of the pump that feeds separation unit – e.g. magnetic flow meters) the flow evaluation can be done. In addition, at least 2 probes for each tank of biological process have to be installed, as a pH probe coupled with a DO probe, or RedOx probe. Moreover, mainly in that kind of process, where the foam formation can produce many problems, also an installation of level sensor, as a low frequency radar level meter can be useful for process controlling (Figure 32).

Furthermore, it would be advisable to provide for a revamping of the installed machines, where these are obsolete. For the monitoring of the system, it would be advisable to provide a continuous and programmed control of the characteristics of the samples taken at different points of the plant.

The situation in the plant TO is different. This plant, in fact, has all the necessary equipment for a process control. For a cost reduction and for optimization of the process it is possible to propose the application, for example, of the real-time control of the aeration provided based on the parameters detected or the control of the biological process based on RedOx and oxygen trend or on pH and DO trends. Also for this plant, the results obtained lead to the conclusion that it is not sufficient to have a furnished and well-designed plant to obtain good results, but it is necessary to manage it correctly.

Figure 32 Example of a low frequency radar meter (Siemens ®)

In this plant, with the purpose of improve the desired yields, tests could be carried out to optimize the duration of the operating cycles; in addition, also an installation of more advanced pre-treatments that allow the entry of nutrient-laden slurry, but poor in inert components. An example of a very good S/L treatment is represented by a centrifuge (Figure 33).

Figure 33 An example of applicable machinery for solid/liquid separation (Pieralisi S.p.A).

The plant NA, VA and RR have a lot of functioning problem. The first two plants are to be considered practically inoperative and, even if the observed period is very short, the evidence for managing problems occurred.

The SBR plant of the NA farm is new and has all the features necessary for its proper functioning. Improvements aimed at the removal of solid ends using a centrifuge will certainly bring about a noticeable improvement in the quality of the wastewater. Also in this plant, the highest step to overcome is correct plant management and operational optimization to limit energy waste and costs.

For plant VA, as observed for plant TP, is necessary to foresee at first a plant revamping. One of the peculiarities of this plant is the large number of tanks for slurry storage. However, the SBR plant receives a total solids-rich slurry, that may represent a limit in achieving good nitrogen yields, as demonstrated also by the collected data. To improve the performance of the SBR, and without making too much impact on the system, it was proposed to prepare a still pipe inside the storage tank of the separated liquid. In particular, it was planned to insert a 1,5 m height concrete pipe inside the tank in which the submerged pump will be installed. In that way, the slurry with a lower solid content, which will go to accumulate in the lower area of the tank, enter this calm zone, and be sent to the SBR with an inferior load of total solids. Also in the VA plant, the probes installed in the SBR tank are damaged and would need to be replaced. Finally, as regards the VA system, it is necessary to set a correct operating cycle of the SBR, and, perhaps, to provide for the start-up of the system using an inoculum of activated sludge coming from a functioning plant.

The RR plant is the most modern and new. In this case, in order to achieve the required improvements, it is necessary to focus on the actual functioning of the SBR system. This is the only system where there is a person employed in the management and control of the plant. This could represent the keystone for obtaining the desired removal yields.

General, for all considered plants, once put in proper function, the further sampling campaign have to be focused on the deepest characterization of inlet flow and on the biological biomass present in the tanks. These can help the comprehension of the real operation of biological process, whose “heart” is in the activated biomass. It is not enough to have a good-quality influent but is necessary to establish the right microbiological activity. For all these reasons, an experimental activity has to be started in order to try to understand if the common NDN and SBR plant’s management for livestock treatment allows reaching the wanted nitrogen and nutrients removal rates, or is indispensable to change and meliorate it.

4.5. References

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5. Biological process for nitrogen removal in SBR applied at laboratory scale

According to the nitrate directive (91/676/CEE), in Italy and in particular in Po valley, many of existing farms need to treat livestock manure prior to its land spreading.

Besides the solid liquid separation and other possible treatments and despite the diffusion of new technology focused on the recovery of nutrient rather than their simple removal, the existing plants in Lombardy mostly apply the nitrification-denitrification process for nutrients removal. This kind of treatment at real scale mostly occurs both in a "classical" scheme (NDN plants) and in sequencing batch reactor systems (SBRs).

As also attested by real monitoring plant activities (please see Chapter 4), in that kind of treatment plants management problems occurs and often the required nutrients removal rate cannot be reached.

For these reasons, NDN process applied in SBR system at lab scale for nutrient removal from livestock waste was analysed in order to study how the nitrogen removal rate changing with the variation of inlet characteristics and operational settings. Regarding the state of the art related to the topic discussed below, please refer to Chapter 3.

5.1. Aim of the experiments

The process performance of biological nitrogen removal was investigated in a sequencing batch reactor (SBR) at lab scale. The general aim is to understand how the operating settings applied in the SBRs systems can influence the desired nutrient removal yields and how the characteristics of the different types of slurry can influence the obtainable quality of the final effluent. For doing that, 4 experiments were set up.

In particular:

- The first batch of experiments ***Test 1*** was aimed to the identification of the SBR systems' responses using different types of slurry (pig slurries form different farms after separation process) and applying the same nitrogen loading rate (gTAN/$L_{reactor}$/d). In that case, the duration of the whole cycle was the same for all reactors but loading and discharging times and consequently the hydraulic retention time (HRT) change with the varying of slurry's type in order to maintain constant the nitrogen load. Accordingly, loading and discharging time is different for each type of slurry.
- *Test 2*, was aimed to the identification of the SBR systems' responses using different inlet sludge (pig slurries form different farms after separation process and liquid fraction of digestate), but with the

application of the same cycle duration, included the length of sedimentation phase, loading and discharging times and consequently the application of the same HRT.

- **Test 3** was aimed to study the variation of nitrogen removal efficiency changing the process parameter and using the same type of slurry for all available tanks.
- **Test 4** consisted in an analysis of the system performances during the aerobic phase, in particular we wanted to analyse if the biomass in the system, subjected to continuous aeration, was able to nitrify.

5.2. Materials and methods

5.2.1. Laboratory-scale prototypes construction and operation

All that experiments are carried out using sequencing batch reactor prototypes that are project, design and building in the DiSAA (Dipartimento di Scienze Agrarie ed Ambientali) laboratory (Figure 34, 35).

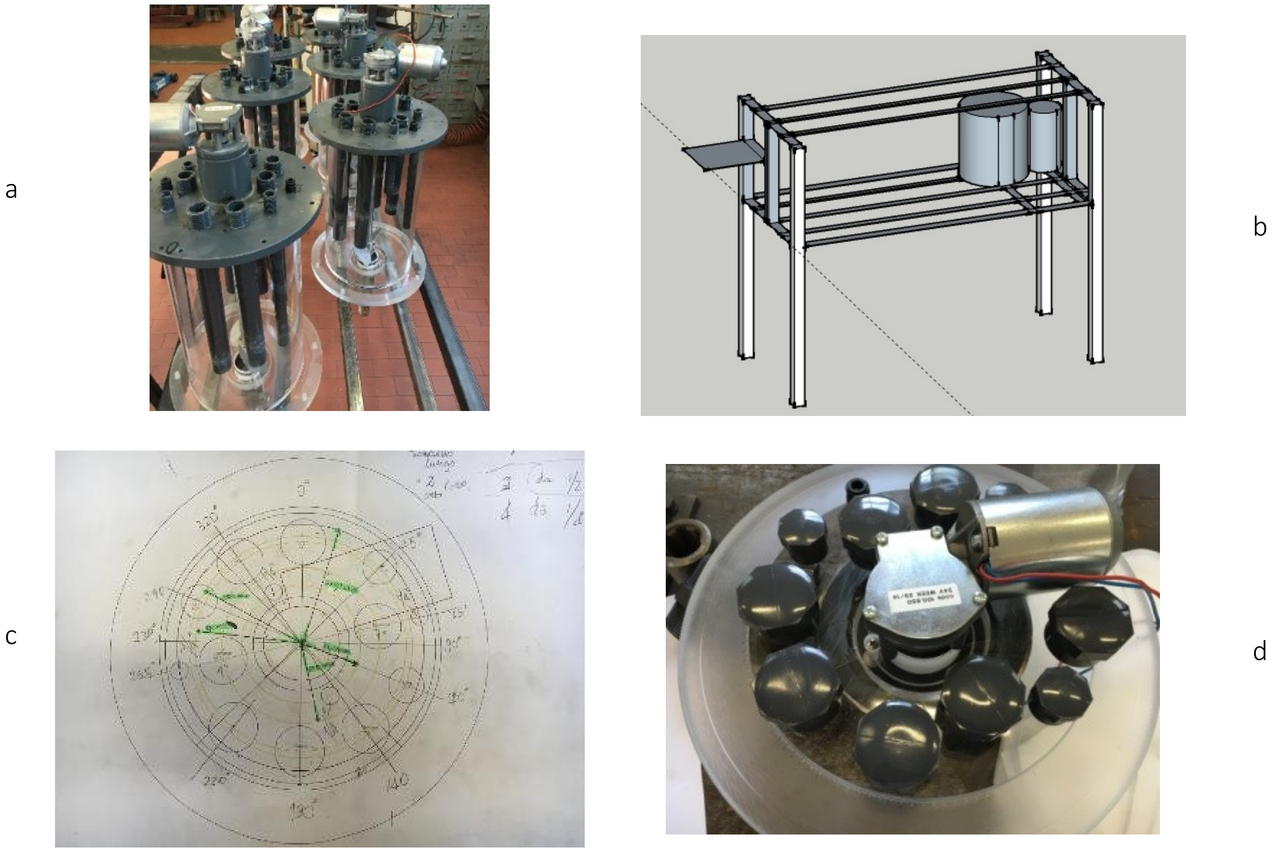

Figure 34 Design and construction phase of SBR prototypes, test and the project of bearing structure, the project and realization of the top cover of SBR.

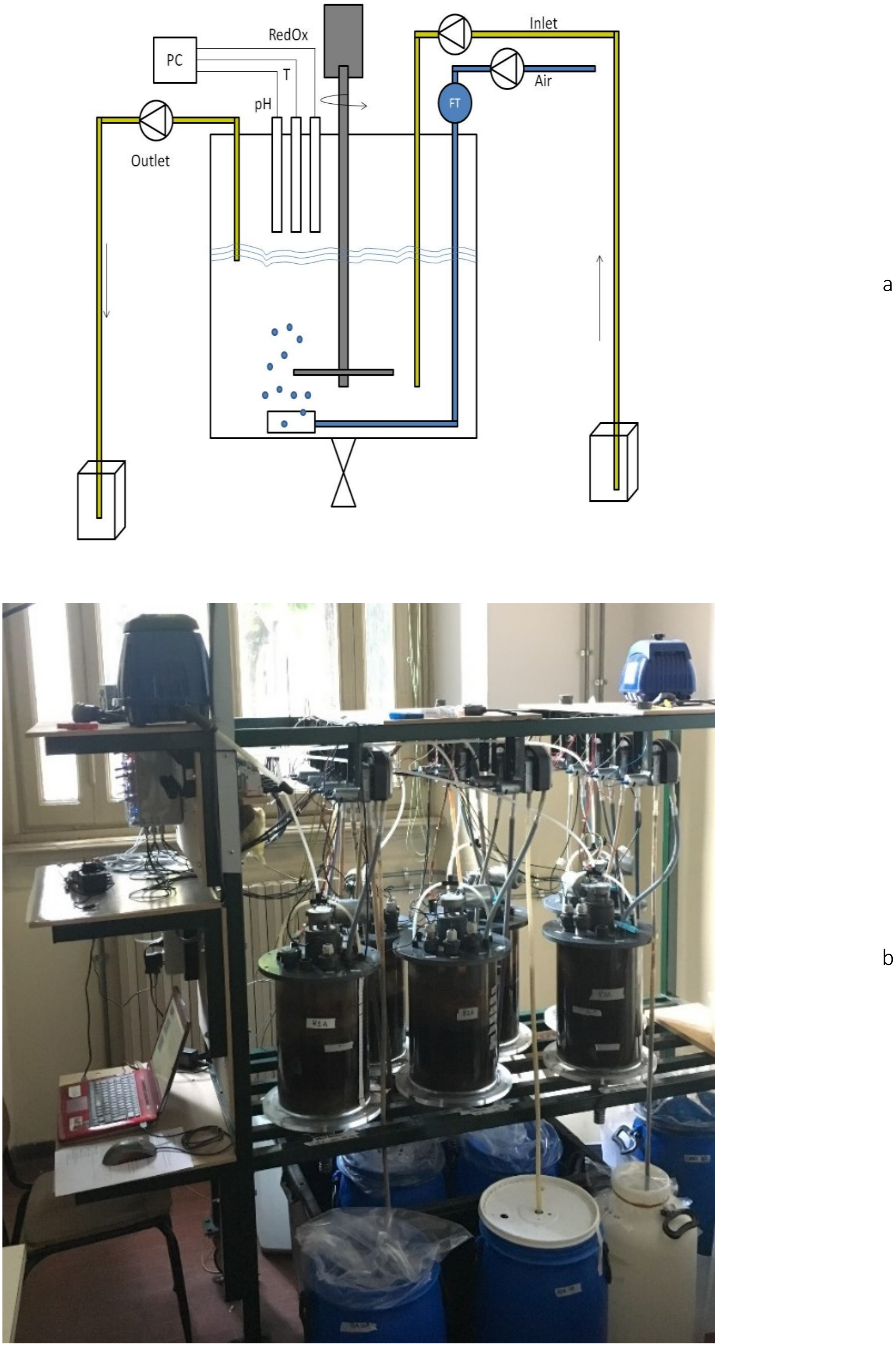

Figure 35 Schematization of the reactor and its components (a); functioning system of six reactors (b) operating with 3 different inlets.

In Table 27 are reported sizing data of one reactor.

Table 27 Sizing data of one reactor (first hypothesis).

DESIGN Reactor- Rev. 01

	U.M.	
Geometrical high (Hg)	cm	35,0
Internal diameter	cm	23,0
Area (Ab)	cm2	415,5
I; Ab/Hg	-	0,7
Internal radius	cm	11,5
Geometrical volume	L	14,5
Net volume	L	10,0
Net high	cm	24,1
SRT (first hypothesis)	d	20,0
HRT (to be defined)	d	15,0
Qin-Qout	L/d	0,5
Cycle time	h	8,0
Number of cycles	cycle/d	3,0
Qin/cycle	L/cycle	0,18
Overflow level	cm	0,43
Q l/ week	L/week	3,7
Q l/2 weeks	L/week	7,5
	Pumps	
Qin/cycle	L	0,18
Q pump	L/min	0,56
Pump operation time	min	0,32

The lab scale treatment system used in the study consists of six Poly(methyl methacrylate) reactors, each with a geometric volume of 14,5 L (35 cm height and 23 cm internal diameter).

Prototypes were provided with a fitting polyvinylchloride lid (Figure 34) with ports for sampling and inserting probes.

Each reactor has an air inlet regulated by a flow meter (2–30 L/min, Key Instruments, Trevose, PA, USA), 2 outlet holes for promoting the air exchange and other 2 air connections in order to permits to measure air gas outlet, if necessary. All the holes, as also shown in the Figure 34, are provided with a cap in case the gas measurement reactor is to be sealed.

Aeration was performed using an air membrane pump (EVO30 series, ELECTRO A.D., Barcelona, Spain) that was set in order to ensure the level of required dissolved oxygen concentration.

Air entered the reactor through a SS316L tube, screwed with two-gas diffuser stones (Scubla S.r.l) placed at the bottom of each reactor that allowed blowing and spreading fine air bubbles in the sludge.

The pH (SZ161, B&C Electronics, Italy) and Redox potential (SZ251S B&C Electronics, Italy) were continuously monitored using a computer coupled with LabView Software (National Instruments, Austin, USA) (Figure 36).

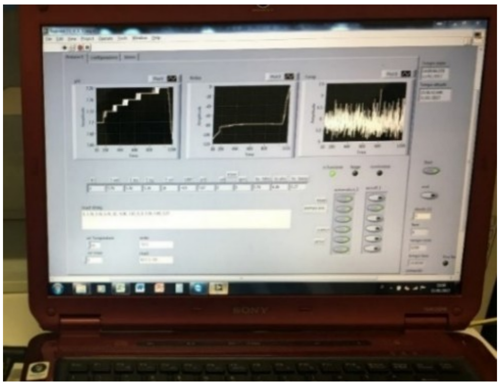

Figure 36 An output window of acquisition system

All the probes were positioned inside a protection PVC tube (Figure 37,b) approximately 10 cm beneath the surface of the reactor contents and connected to pH and RedOx circuits (Atlas Scientific LLC, New York, USA and tentacle card, 4 channel) in order to communicate with software. With the same software, the dissolved oxygen in the sludge was spotted measured using oxygen sensor (VisiWater DO sensors, Hamilton). The temperature of reactors was monitored by a temperature probe (DS 18 B 20) immersed in the liquid’s reactor.

Each reactor was also provided with a mechanical stirrer consists of two horizontal blades mount on a vertical shaft (SS316L) and oriented to push down the liquid. The mixer was driven by an electrical motor (Elvi S.r.l) with a gearbox and with a shaft rotation of 60 rpm.

The feed and discharge of different livestock manure occurred with 2 peristaltic pumps (24VDC pump, Verdeflex, 82 rpm, 8X2,4 mm Verderprene piping, flow rate approximately equal to 0,8 L/ min) installed on each reactor. One of the pump, during some preliminary test for setting the operation of the geared motor are shown in Figure 37,a. Also the mixing, feeding, and discharging of the reactor were automatically controlled through Arduino® mega and management with a computer program developed by LabView Software. The six reactors named R1A, R1B, R2A, R2B, R3A, and R3B were placed on an iron structure, as also shown in Figure 35. The reactors worked in pairs in order to have double experimental units contextually. When working with different type of slurry, each couple of reactors had its own inlet tank, from which the peristaltic pumps withdrawn. Excess sludge discharging occurs manually in order to maintain the required SRT. The reactors are equipped with a 2 1/2" PVC ball valve to be able to empty them easily.

During the experimental tests, ***3*** and ***4***, the system of charge and discharge of manure was implemented.

A balance system is mounted under each SBR tank in order to manage the inlet and the outlet of the slurry based on the weight of each tank. For doing this, a program was implemented with LabView.

a) b)

Figure 37 Peristaltic pumps that were chosen and installed for feeding and discharging of sludge during a preliminary functioning test.

In addition, in order to allow the correct homogenization of the inlet slurry (that in the previous experiments was do manually), a system for automatic mixing (mixer in one tank) was added; before to charge the inlet flow, the mixer was turned on. In the Figure 38 is reported the implemented system used for the experiments.

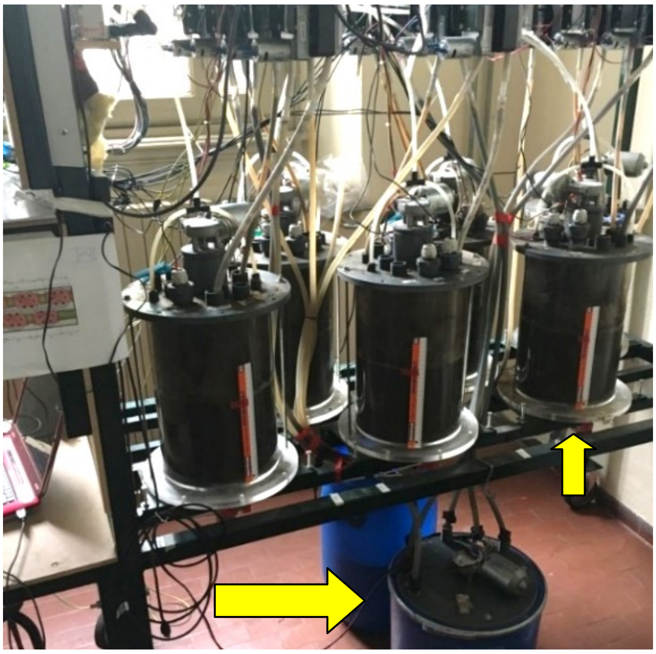

Figure 38 Operative system of six reactors after the installation of weight system and mixing system for the inlet vessel (indicated with yellow arrows).

5.2.2. Experimental plan

First, a series of preliminary tests with water and then with real sludge were performed for attesting the correct operation of the systems, the hardware and the software. This period of settlement lasted 2 months

In order to accomplish the SBR tests, activated sludge needed for starting biological activity *rapidly* was collected by an operative NDN plant ("PLANT 0") located in Casaletto di Sopra (CR). That decision had been made because this plant discharges the effluent in surface water body respecting the stringent laws of D.Lgs. 152/06 (see also paragraph 1.3.2). Therefore, the observance of these limits leads to assume that the plant has a correct functioning and therefore that the activated sludge has a good quality.
Some of the main characteristics of the plant 0 are resumed in Table 28.

Table 28 Main characteristics of plant 0.

Plant 0 Characteristics					
SRT (Sludge Retention Time)_Bioprocess	7	-	15	d	NOTE
SRT (Sludge Retention Time)_Nitrification	5,83			d	
x (biomass concentration in the tank)	4	-	6	gSST/L	
x	2	-	4	gSSV/L	1
CF (carbon load)	0,09			kgBOD$_5$/KgSST/d	
CF	0,15			kgBOD$_5$/KgSSV/d	1
CN (nitrogen load)	0,03			kgN/KgSST/d	
CN (nitrogen load)	0,05			kgN/KgSSv/d	1
CODin	3150			mg/L	
TANin (as N)	850			mg/L	

NOTE

1) Empirical calculation considering SSV/SST = 0,6

After the correct starting of the systems, all six reactors were filled with 7 L of diluted activated sludge: 5 L of activated sludge from the plant 0 and 2 L of tap water. Overall, the concentration of total solids (ST) in the tank was equal to 4,5 g/L. During preliminary test, the fed of the system was done with the influent of the same plant 0. This choice was made to attest the lab-system's responses thanks to compare obtained results with those observed at the real plant. After 15 days of tests, some problems related with sedimentation phase occurred: the level of sediment sludge is very high, and the clarified sludge was not visible in the reactor due to high mixed liquor concentration. For these reasons, reactor's content was diluted with distilled water in order to have a mixed liquor concentration equal to 4 g/L (for 6 reactors: R1A: 4,1 g/L, R1B: 3,99 g/L, R2A: 3,99 g/L, R2B: 3,99 g/L, R3A: 4,1 g/L, R3B: 4,0 g/L).

In Table 29 and 30 are reported the main data regarding all experiments that we carried out.

Table 29 General information regarding the experiments.

Experiment	Number of used inlet slurry	Type of sludge	Variable parameters	Duration of experiments
Test 1	3	Pig slurries pre-treated with different S/L	HRT+ Qin+ type of slurry	from 04/2017 to 07/2017
Test 2	3	Pig slurries pre-treated with different S/L + digestate	Type of slurry	from 07/2017 to 10/2017
Test 3	1	Pig slurry	Number of cycle/day + setting of the cycle	from 12/2017 to 02/2018
Test 4	1	Pig slurry	All reactors work equally	from 02/2018 to 04/2018

Table 30 Experiments general settings.

		TAN loading rate (gN/Lreactor/d)	Working Volume (L)	Number of daily cycles	HRT (d)	SRT (d)	Volume Load (L/cycle)	Duration anaerobic phase (h)	Duration of aerobic phase (h)	Air insufflations (L/min)	Duration of sedimentation phase (h)
TEST 1 preliminary	R1AB R2AB R3AB	0,1	7	3 [*]	Variable, depending on the nitrogen load	25	Variable, depending on the nitrogen load	3	3,5	10	<1
TEST 1	R1AB R2AB R3AB	0,2 [†]	7	3		25		3	3,5	12	≈1,5
Test 2	R1AB R2AB R3AB	0,2	7	3	15	25	0,47	3	4,3	Air insufflations: based on the measured O_2 concentration	<1
Test 3	R1AB	Variable	7	3	8	35	0,29	3	4,3[‡]		0,69
	R2AB		7	3	8	35	0,29	2.5	5,3		0,2
	R3AB		7	2	8	35	0,44	2.6	7,4		2

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[*] Tilche et al., 1999; Ra et al., 2000

[††] Poo et al., 2005

[‡] Also with mixing

For all reactors and all the experiments carried out, pre-denitrification process (and not post-denitrification) was applied, in order to be able to provide the denitrifying bacteria with the COD present in the incoming slurries and avoid the use of external sources of carbon.

The specific features of those experiments are reported in the following paragraphs.

For all experiments, whenever a new batch of waste was used, it was chemically and physically characterized. Furthermore, once a week the inlet and outlet of each reactor were analyzed. Monitoring of RedOx and pH was continuous for all reactors.

5.2.3. Analytical methods

All inlet samples were analysed for TKN (Total Kjeldhal Nitrogen), total ammonia nitrogen (TAN), nitrates and nitrites (NO_3^-, NO_2^-), pH, dry matter content (total solids, TS), volatile solids content (VS) and electrical conductivity (EC), which were determined using standard procedures (APHA, 2014).

Additionally, COD -chemical oxygen demand- was measured in the inlet flow and outlet flow by the reactor digestion method using COD digestion vials (Nanocolor®, MACHEREY-NAGEL GmbH & Co. KG). During the experimental period, weekly a sample of inlet and outlet (treated effluent) from each reactor was collected and analysed for TAN, EC, and pH to identify the transformation of slurry over time. Reached the stability of operation system (e.g. TAN value constant) all chemical-physical analysis were performed.

Biological oxygen demand (BOD) was defined using BOD Sensor BOD Analysis (Velp Scientifica).

The sludge sedimentation was evaluated taking a sludge sample from each reactor and check how the level of the activated sludge changes within a graduated cylinder over time (30 min).

5.3. Test 1 and Test 2 (influence of nitrogen loading rate and slurry characteristics on the nitrogen removal efficiency)

In order to study and evaluate the relationship between nitrogen loading rate, type of influent and nitrogen removal rate achievable, the nitrogen load imposed for this part of test is equal to **0,1 gN (TAN)/L/d** (Magrì et al., 2008). Three different influent sludge A, B, C were used:

- Liquid fraction of pig **slurry A** obtained after a separation treatment achieved through a rotating rotary screen and a floating system, (*Farm 1*); fed in reactorR1A and R1B;

- Liquid fraction of pig **slurry B** obtained after a separation treatment achieved through a rotating rotary screen and a flotation system, (*Farm 2*); fed in reactorR2A and R2B;
- Liquid fraction of pig **slurry C** obtained after a separation treatment achieved through a rotating rotary screen (*Farm 2*), fed in reactor R3A and R3B.

After a short operation period, it was decided to replace two of the three inlets used with others with higher nitrogen concentration and with the most representative characteristics of the livestock slurry (see also chapters 3). Then, **slurry A** and **slurry B** were substituted respectively with other two types of sludge:

- Liquid fraction of pig **slurry E** obtained after a separation treatment achieved through a rotating rotary screen (*Farm 4*) fed to reactor R1A and R1B;
- Liquid fraction of pig **slurry D** obtained after a separation treatment achieved through a rotating rotary screen (*Farm 3*), fed to reactor R2A and R2B;

The imposed nitrogen loading rate **0,1 gN(TAN)/L /d** has led to continuous variations in the duration of the cycles (in particular loading and unloading time) since it was necessary to maintain that parameter constant despite variations in the inlet sewage.

The removal yields achieved with the application of 0,7 gTAN/d were high and, furthermore, the nitrogen detected in the output sample was low and did not respect the detection limit of analytical method used. Therefore, we decided to increase the nitrogen load enters the SBR systems, acting on the duration of the flow rate charged and discharged by each reactor. In particular, nitrogen loading is imposed to **0,2 gN(TAN)/L/d**. Also for this experiment slurries C, D, E was used.

At the university, it is impossible to stock a large amount of sludge in proper barrels, so was necessary to withdraw each 15 days new sludge from each farm. Every time that new sludge was sampled to be added to the inlet tank, the operational set of each SBR was changed in order to maintain the desired nitrogen load.

The *Test 2* started after a period of stand-by for all reactors: during 10 days, a “pause” cycle was imposed and foresee only a fed step, a discharge step, a period of 1 h of aeration and 0,5 h of mixing.

Concerning the type of sludge, for these experiments were used:

- Liquid fraction of pig **slurry E** obtained after a separation treatment achieved through a rotating rotary screen (*Farm 4*);
- Liquid fraction of pig **slurry D** obtained after a separation treatment achieved through a rotating rotary screen (*Farm 3*);
- Liquid fraction of co-digested pig slurry and corn silage (**slurry F**) after a separation treatment achieved through a rotating rotary screen (*Farm 5*).

During these tests, an operational change was done: after a period of monitoring of dissolved oxygen, the airflow rate insufflate has been set to maintain an oxygen level > 3,5 mg / L.

5.3.1. Livestock manure characteristics

As explain in the previous paragraph, 3 different slurries were used for the *Test 1* (named C, D, E) whose characteristics are reported in Table 31.

Table 31 Main characteristics of 3 sludges used for experiments (Test 1); A: average data, SD: standard deviation.

		TAN g/Kg	TKN g/Kg	EC mS/cm	pH	ALK $gCaCO_3/L$	ST %	SV/ST %	BOD O_2g/L	COD O_2g/L
R1AB_IN	A	3,42	4,51	26,24	7,91	17,16	3,47	52,41	14,19	31,97
(slurry E)	SD	0,43	0,72	6,25	0,16	2,56	0,80	4,55	0,82	7,63
R2AB_IN	A	2,54	3,41	21,58	7,64	11,71	1,89	55,01	9,52	25,25
(slurry D)	SD	0,22	0,37	3,72	0,27	1,32	1,25	8,24	2,84	3,57
R3AB_IN	A	0,65	1,21	5,92	7,36	4,88	1,00	69,03	4,28	9,82
(slurry C)	SD	0,16	0,19	2,15	0,16	2,80	0,35	2,87	0,42	0,83

The used slurries E and D had comparable characteristics contrary to slurry C. The differences are also related to the “origin” of the slurry. In fact, farm 2 differs from the others since farms 3 and 4 are closed-cycle pig breeding, instead the farm 2 has also a pig production section. Moreover, in this plant, the use of water is conspicuous, with the result that the collecting livestock waste used for laboratory tests has a low content of TAN, as well as of TKN, total solids, BOD and COD in comparison with the characteristics observed for the slurry C and D. Anyway, all types of slurries have typical characteristics of livestock waste (please see also paragraph. 3.4).

For all inlet sludge used, the ratio between the content of carbonaceous compound (COD) and nitrogen compound (in this case, TKN) is higher than 7 (in particular for couples 1AB: 7,1, 2AB: 7,36, 3AB is 8,18). This means that the system could be able to denitrify correctly and reach higher removal rate (Obaja et al., 2003), as also explain in chapter 3.

In Figure 39 the variation of inlet flow for each reactor that occurred during all the ***Test 1*** of the tests is reported.
Once again, is evident the importance of definition of slurry characteristics for correct slurry management.

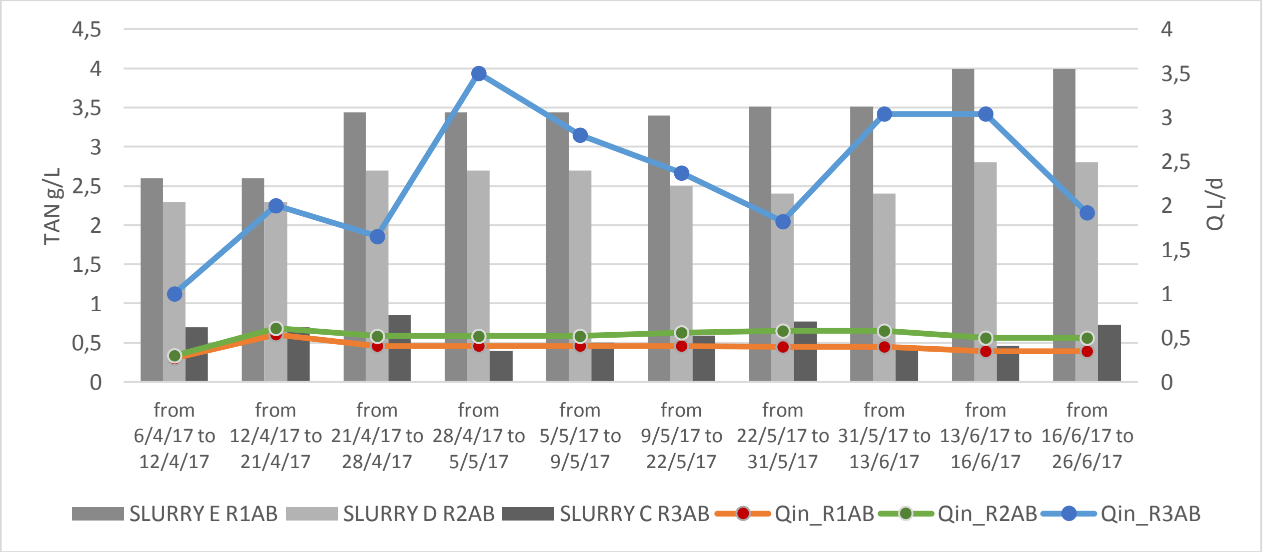

Figure 39 Variation of the slurry inlet flow, and TAN concentration attested during Test 1 for maintaining the constant nitrogen load.

5.3.2. Experiments' results: Test 1

In the following graphs are reported the results of the test 1, include the detected nitrogen compound concentration in the inlet and in the outlet samples (Figure 40).

As possible to deduce from the graphs, the output concentration of nitrogen (as TAN) for reactors 1A and 1B and reactors 2A and 2B was always lower than 1,5 g/L; instead, for reactors 3A and 3B, the maximum concentration observed was lower than 0,35 g/L (as TAN). Clearly, since the nitrogen concentration in inlet slurry of reactors R3A and R3B was lower, the outlet concentration for this reactor was much lower rather than other couples.

Analysing the graphs, it is also possible to observe how, during the first period of operation of the system, the concentration of nitrogen detected at the output of the reactors was lower than those observed for the following period. In fact, during the first days of process, the nitrogen load supplied to the reactors was lower (0,1 gN/L/d) and for this reason in the subsequent days the outgoing concentrations increase.

The content of nitrate and nitrite (defined only as sum e.g. NO_3^- + NO_2^-, for the limit imposed by the used method) were always lower than 0,12 g/L for all reactors. This evidence can be related to two different causes: the first is that all NO_3^- produced during nitrification process has been reduced to N_2 during denitrification process, and so, that the systems work correctly, or, in a negative perspective, that nitrification did not occur. However, given that the TAN and the TKN concentrations had undergone a reduction, it could be that the process has effectively reduced the nitrogen content in the slurry. In support of this statement, it is possible observed also the removal rate reached. In order to calculate the removal rate, a daily nitrogen load is been considered.

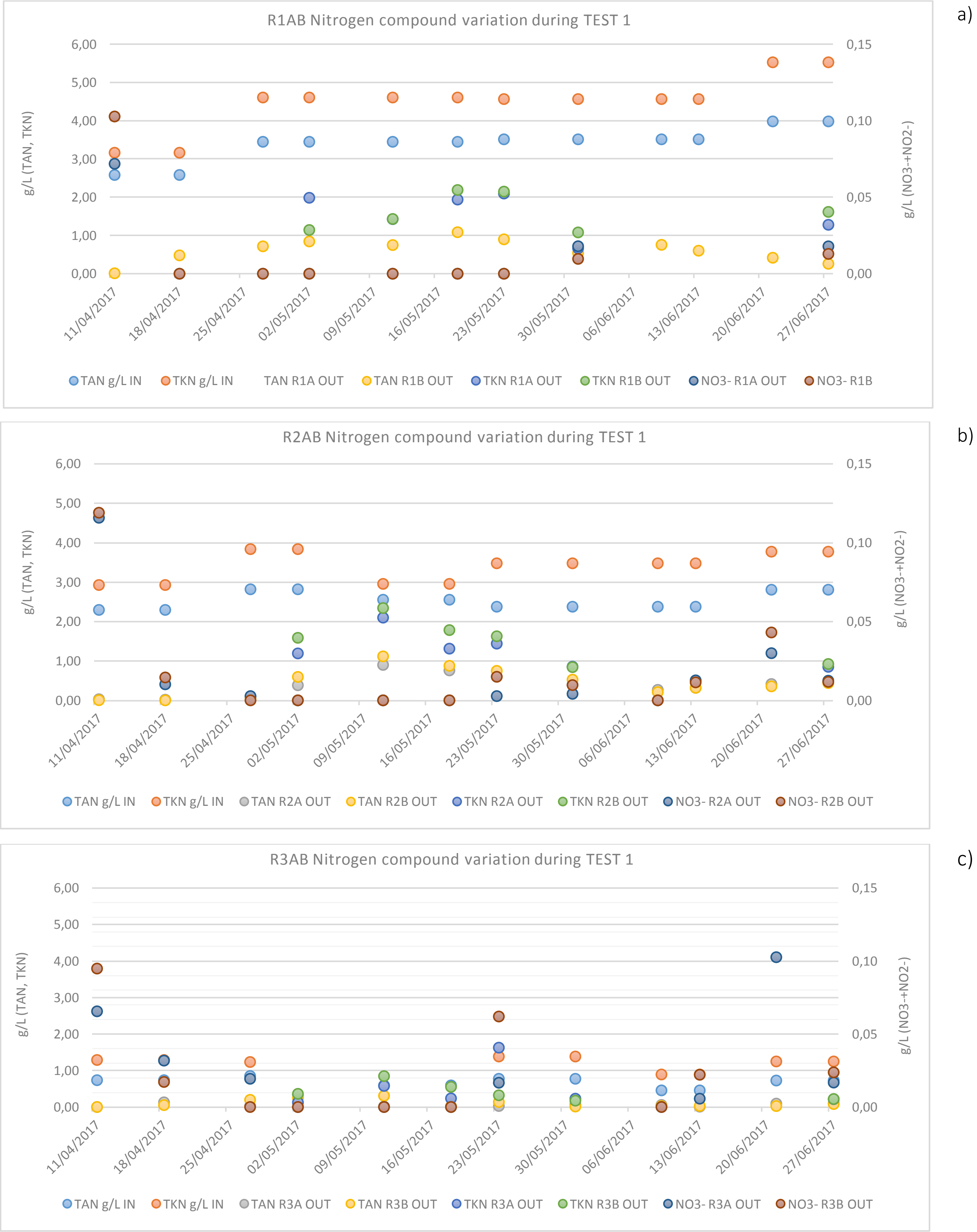

Figure 40 Nitrogen concentration detected in inlet and outlet samples for reactors R1AB (a), R2AB (b), R3AB (c) during Test 1.

For the evaluated period (experiment-*Test 1*) the minimum and maximum removal rate for TAN achieved for the reactors 1A & 1B were 70,8%-98,5% and 68,5%- 99,5% respectively, for reactors 2A & 2B 64,8%-99,3% and 56,3%- 100% respectively, and for reactors 3A & 3B 70,4%-100% and 25,7%- 100% respectively. TKN removal yields, on the other hand, were lower and on average stand at 65% for R1AB reactors, 58% for R2AB reactors and 43% for reactors R3AB. It should be noted that, despite the operation of the two reactors is identical for each pair, the results obtained are not always equal. This could be due to temporary malfunctions that affected only one of the two reactors (such as software blocks during the research) or differences in air diffusion, or due to the bacterial consortium created in each individual reactor.

For a better comprehension of obtained results, it would be very interesting and useful also to analyse microbiologically the contents of the reactors. Anyway, only referred to nitrogen, in all three systems, even each of them worked with different slurry type, guarantee high removal rate.

Nevertheless, to enhanced understand it is necessary to analyse not only removal yields but also the pH and RedOx trends.

In order to explain the variation of pH and RedOx, parameters detected during one day of functioning at the beginning of the experiments, after 1 month, after 2 months and at the end of the tests, are considered for all pairs of reactors (Figures 41, 42, 43). The reported pH and RedOx value recorded every 15 seconds during one day of operation.

Regarding the reactors R1A and R1B, as possible to see in Figure 41, the trend of ORP and pH did not always coincide for the reactors of the couple. As explain above, this can be caused by multiple factors related to both operation problems occurred and differences in the slurry inside the reactor.

Anyway, as expected, the path of ORP increased during aeration period, reaching value over the 0 mV; the highest observed peak was 200 mV and the lowest reached values was below -500 mV.

The two parameters’ evolution changed over time during the experiment: the modification of inlet load may influence the operation of the system. In the first period (Figure 41,a) the systems R1A and R1B reached very high TAN removal rate (>98%): the very low nitrogen load allowed to the system, already acclimatized and working, to better remove nitrogen.

The lowers removal value reached (77%) was attested in the day 11/05/2017 represented in the Figure 41, b. In that case, the reported graph is very “un-normal” compared with the classical output of a conventional SBR system operating with a NDN process (Figure 44). In fact, the pH remains always on basic value (pH>9,25) and did not have a real modification and during an aeration period.

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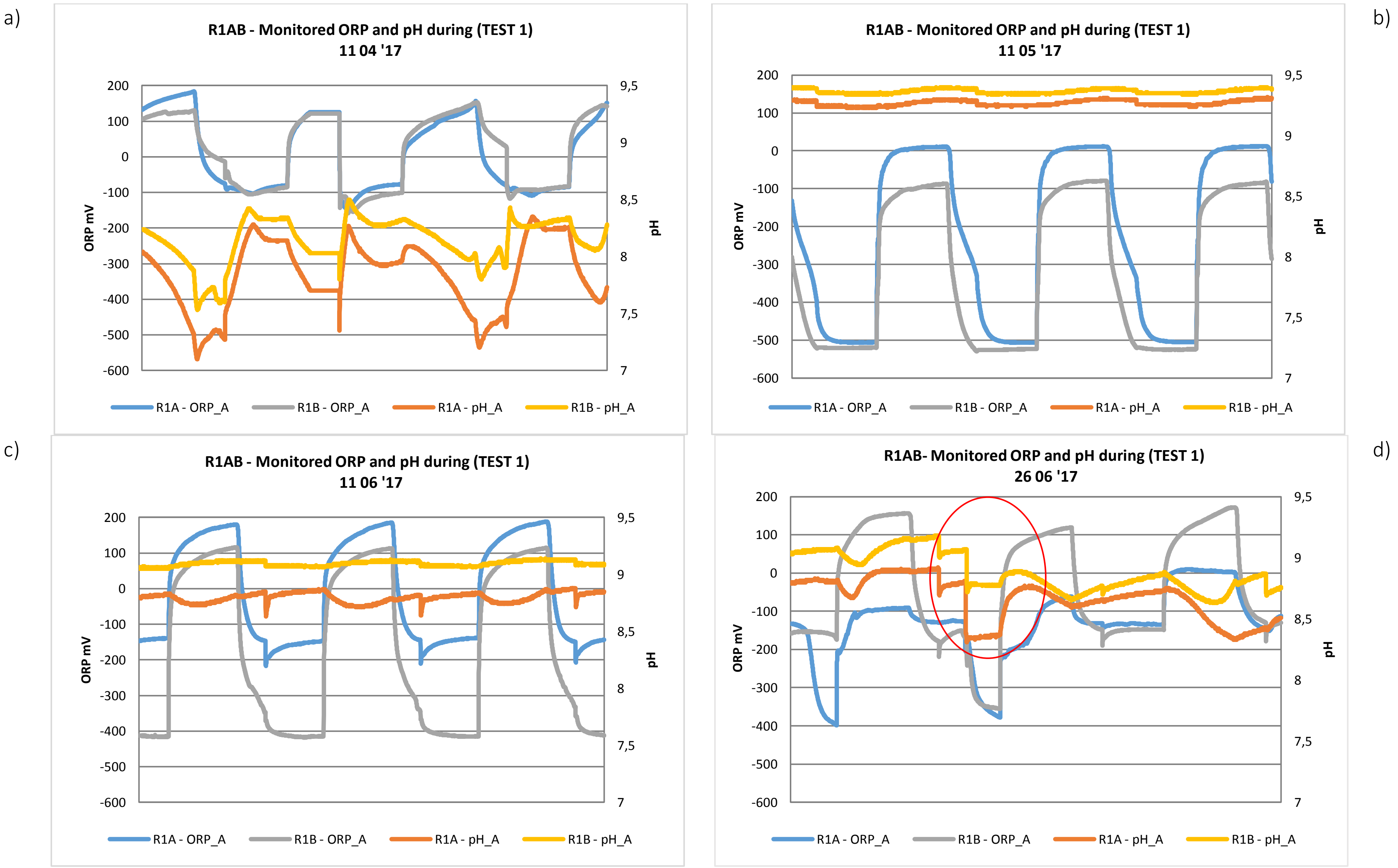


Figure 41 Trend of parameters monitored for reactors pair R1A R1B during one day (24h), in particular: at the beginning of experiment (TEST 1), after 1 month of operation, after 2 months of operation and at the end of experiment.

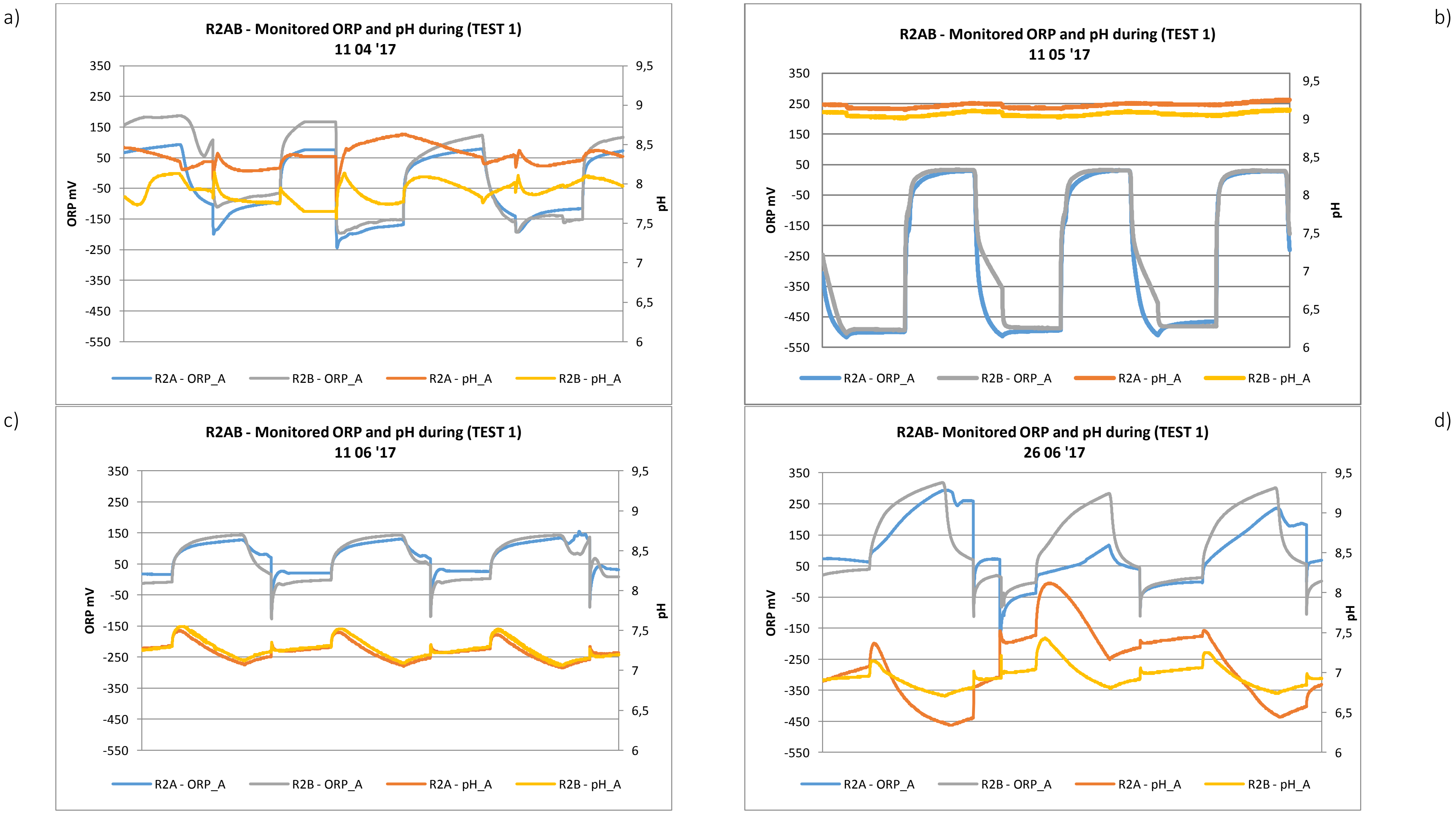


Figure 42 Trend of parameters monitored for reactors pair R2A R2B during one day (24h), in particular: at the beginning of experiment (TEST 1), after 1 month of operation, after 2 months of operation and at the end of experiment.

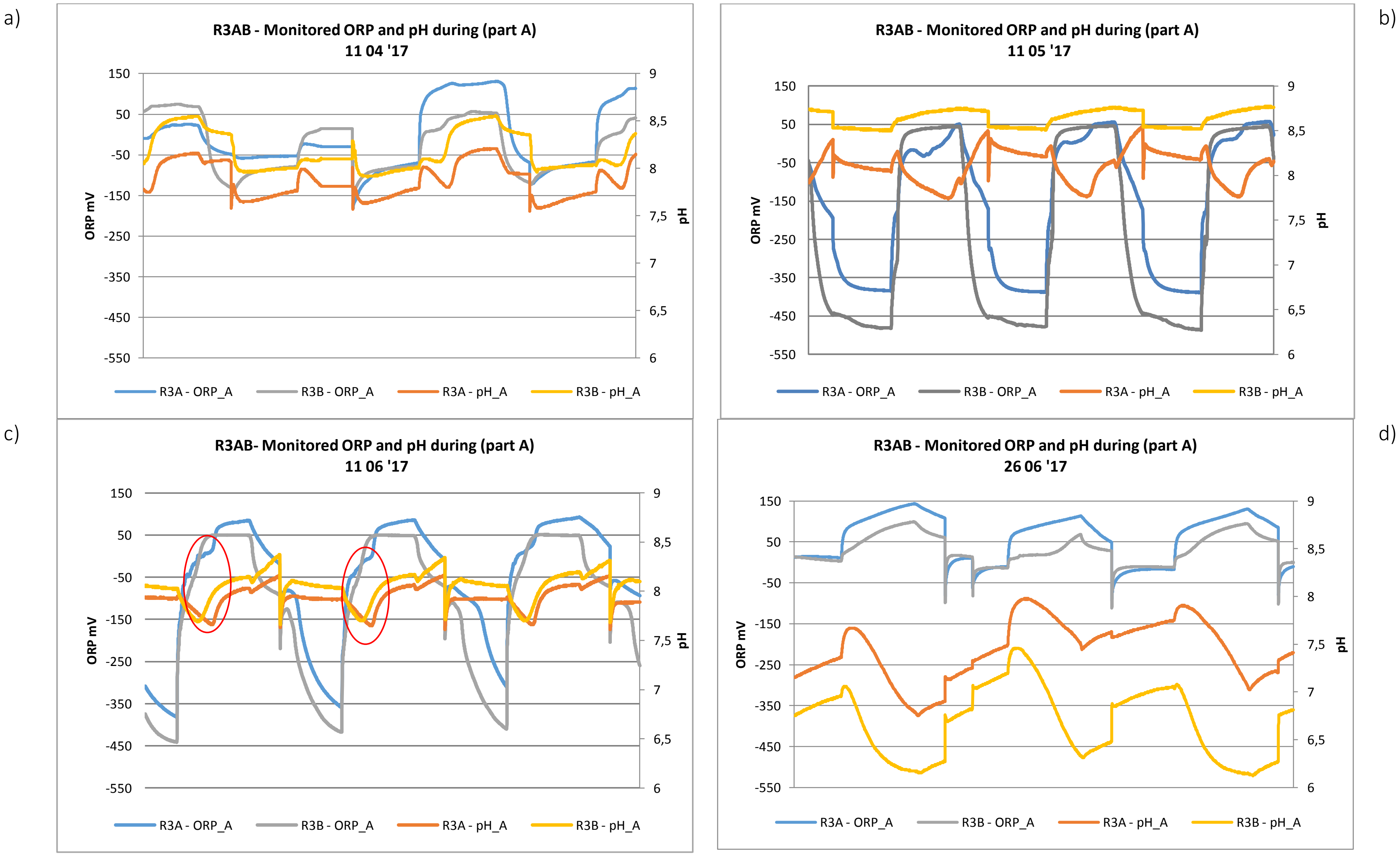
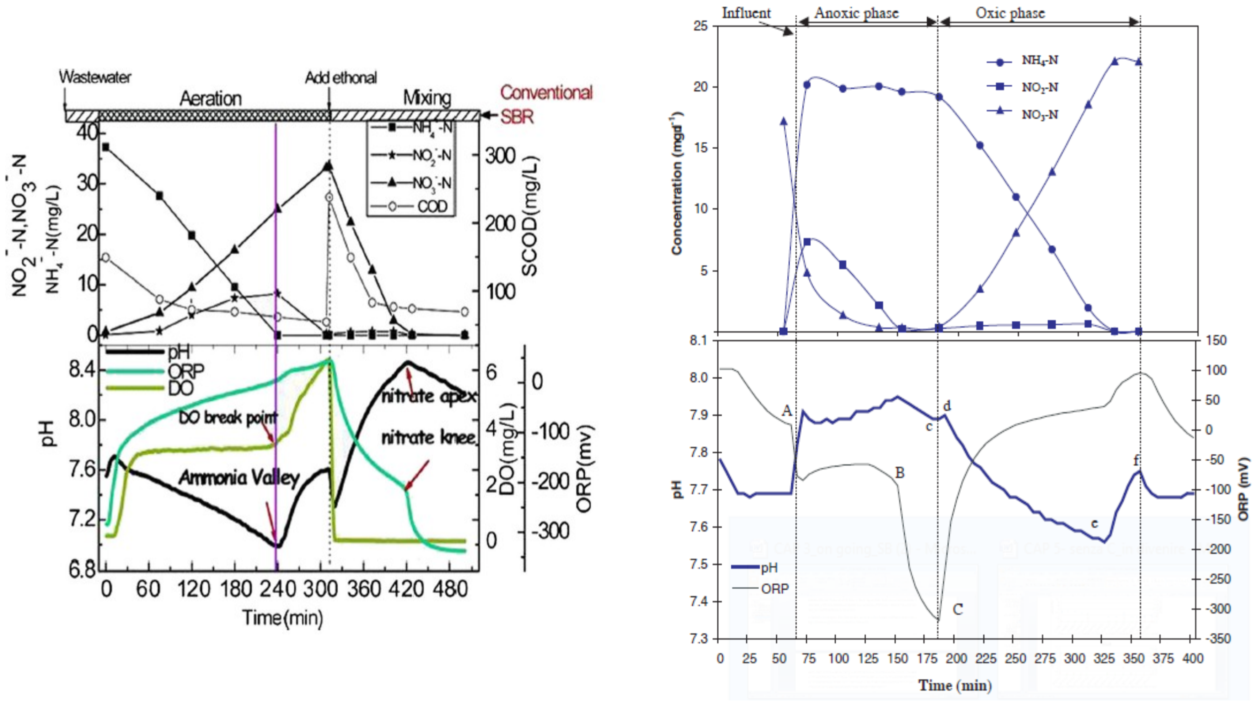


Figure 43 Trend of parameters monitored for reactors pair R3A R3B during one day (24h), in particular: at the beginning of experiment (TEST 1), after 1 month of operation, after 2 months of operation and at the end of experiment.

If the functioning of the system was correct, during the mixing phase a reduction of the RedOx potential should be observed, with a simultaneous increase in the pH due to the production of alkalinity during denitrification phase; vice versa, during the aeration phase, the pH should decrease, for the production of hydrogenions.
For reactor R1A and B, it may also happen that the ammonia present in the slurry was stripped by the airflow supplied to the system, assisted by the very high pH. Also during the day 11/06/17, the pH-trend is not as expected. In addition, in the final period of observation, R1A has an unconventional ORP's trend: in fact, ORP remained always below the 0, indicating that the oxygen furnished did not reach suitable values for nitrification to take place (please refer also to Chapter 3). Nevertheless, the removal rate reached is anyway above 90% (R1A); so is also possible that the ORP probe was dirty and did not read correctly the values. This fact can happen also at real scale: for these reasons it is very important to make maintenance of the plants, to guarantee their removal rates wanted. The strange step that is visible in Figure 41,c is related to a manual control of fed phase (please see the red circle in the figure).

Regarding the reactors R2A and R2B, the obtained trends of pH and ORP are reported in Figure 42.
Also in that case, the trend of two parameters changed over time and, considering the couple of reactors, and did not always coincide. During first examined day (Figure 42,a) the pH of reactor 2A had a strange behaviour and increased during aeration/nitrification, contrary to expectations. Nevertheless, the removal yield of TAN reached is > 99%. The lowest removal rate recorded (<60% for the reactor 2B and <66% for reactor 2A) correspond to the graph of 11st May 2017 (Figure 42, b). The very high pH value, which did not have variation during all the three cycles of the day, is a symptom that the system was inhibited and, in any case, that it was not working correctly. An anomaly is also visible during the day 11/06 where the RedOx potential of the rector R2A did not remain negative after the loading of new waste as should. In fact, the load of fresh waste enhanced a reducing environment (Zhu et al., 2006). In that case (R2A), a negative RedOx peak was observed due to the addition of fresh waste, but the value, in a short time, returns to positive levels, remaining in this window of values until the next slurry load. It is therefore possible that, in these cycles, the nitrogen-reducing bacteria did not work properly, and denitrification of the nitrate ion present in the slurry did not occur. However, the pH during the denitrification phase had a slightly increasing trend, in line with what expected; this is true also for nitrification phase, where the pH decreased. The yields achieved by the two reactors in this period are> 86%.
During the last day of operation (Figure 42,d), both reactors reached high removal rate of TAN (>70%), but lower than those one observed for reactor R1AB.

Regarding the reactors R3A and R3B, the situation seems a little be different (Figure 43) and in any case, the pH graph is not flat and devoid of variations: this could mean that for these reactors the anomaly observed for the 2AB and 1AB couples was not observed. In addition, even if is very difficult to identify the typical significant control points on the ORP and pH curve (Figure 44), during the 11/06/17 for the R3A reactor it seems possible to recognize the ammonia valley (namely the end of the nitrification in the aerobic phase) indicated with the red circle on the figure.



a)

b)

Figure 44 a) Typical variations of DO, pH, and ORP value and concentrations of NH_4-N, NO_2-N, and NO_3-N during nitrification and denitrification process in a conventional SBR (Dutta et al., 2015). b) Real-time control points in high C=N ration load cycles (TOC/TN ratio of the influent: 1.4) A: Feeding, B: Nitrate knee point, C and c: Beginning of the oxic phase, e: Ammonia valley point, f: End of the oxic phase.

Using the slurry C that has the lowest content of total solid and the highest COD (BOD)/TAN ration, the reached removal rate over time are very high, with the maximum observed for first day (lowest load) and in any case with values always higher than 85%. Only during one day, the nitrogen removal for reactor 3B was very low (48%), and this malfunction can also be found in the pH chart (Figure 43,b). During the last operating period the pH inside the R3AB reactors (Figure 43,d) dropped a lot, perhaps also due to the lower amount of alkalinity brought by the fresh effluent entered the reactor at each cycle, not sufficient to buffer the production of H^+ ions during nitrification. As already said, the understanding of what happens during a biological process is much more complex than what can be expected.

In Table 32, are reported data concerning the input and output of each reactor during *Test 1* in term of daily load. Despite all considerations done for nitrogen, is very interesting also to note what occur for solids content: also these characteristics has a great contribute to the reachable nutrient removal.

Table 32 Characteristics of the input and output in terms of daily load for 3 pairs of reactors during Test 1 (A: average; SD: standard deviation).

		TAN (g/d)		TKN (g/d)		ST (g/d)		SV (g/d)	
		A	SD	A	SD	A	SD	A	SD
R1AB	IN	1,34	0,20	1,77	0,30	14,02	3,44	7,48	2,17
R1A	OUT1	0,21	0,14	0,62	0,23	10,66	2,12	5,14	1,26
R1B	OUT2	0,25	0,12	0,63	0,20	10,76	3,82	5,51	1,43
R2AB	IN	1,35	0,21	1,81	0,33	10,53	7,61	6,28	5,18
R2A	OUT1	0,23	0,17	0,71	0,27	8,12	1,52	4,18	1,36
R2B	OUT2	0,24	0,20	0,84	0,32	10,38	2,00	6,06	1,88
R3AB	IN	1,33	0,20	1,76	1,13	20,53	5,65	14,24	4,22
R3A	OUTA	0,09	0,09	1,02	1,02	9,46	3,42	5,07	2,71
R3B	OUTB	0,23	0,29	0,98	0,66	9,05	4,40	4,86	3,59

For reactors R1AB and R2AB the solid daily loaded are comparable for input and output samples. This means that the sedimentation phase did not work correctly. In fact, if the activated sludge in the reactor had a good characteristic of sedimentation, only the clarified part is discharged. Barely for the couple of reactors 3AB the withdrawn sample has a total solids and volatile solids content of 56% and 65% lower than the input, respectively. For the other couples, this does not occur. It can be useful to evaluate if the sedimentation time applied is correct for reaching the quality effluent requested. The high content of solids can also mean that a lot of organic material (volatile solid) it is released with the effluent and is not used by the bacterial biomass.

Since *manure C* had the lowest concentration of nitrogen, the reactors 3A and B, which treated the same nitrogen load that the other couples but had a higher flow rate with only "more diluted" characteristics, achieved better removal rates and quality of the effluent discharged.

Nevertheless, as already widely repeated, slurry C does not represent very well the characteristics of typical slurry normally sampled and treated at the real scale.

5.3.3. Experiments’ results: Test 2

For the reasons provided before and for the difficulties occurred during the ***Test 1*** caused mainly by the continuous variations of loading and unloading, it was decided to modify the experiment setting.

In particular, the experiment 2 was focused on the **application of same HRT**, using 3 different sludge.

Before to start the new experiment, the “0 condition” was defined: the activated slurry inside the reactor was characterized for content of TAN, total solid and volatile solid (Table 33). Each reactor was filled with a volume of 7 L.

Table 33 Some characteristics of the activated sludge inside the reactors before to start the Test 2 .

REACTOR	TAN g/L	ST%	SV/ST%
R1A	0,55	6,10	54,47
R1B	0,42	4,99	53,09
R2A	0,14	2,12	52,53
R2B	0,20	2,20	52,62
R3A	0,43	3,33	66,06
R3B	0,12	3,51	65,24

The characterization of slurries used in the second part of the ***Test 2***, are reported in Table 34.

Table 34 Main characteristics of 3 slurrys used for experiments (Test 2). A: average, SD: standard deviation.

		TAN g/Kg	TKN g/Kg	EC mS/cm	pH	ALK gCaCO$_3$/L	ST %	SV/ST %	BOD O$_2$g/L	COD O$_2$g/L
R1AB_IN	A	2,53	3,69	22,30	8,47	12,90	1,77	33,27	3621,3	14502,4
(slurry E)	SD	0,27	0,69	8,28	0,16	2,50	0,38	4,00	-	-
R2AB_IN	A	1,30	1,43	16,54	8,44	12,08	0,96	26,74	1510,5	5402,7
(slurry D)	SD	0,34	0,14	7,88	0,00	82,41	0,10	8,77	-	-
R3AB_IN	A	2,17	3,10	24,40	8,22	11,21	2,90	49,33	5970,4	31787,8
(slurry F)	SD	0,22	0,45	0,91	0,078	5,40	0,62	7,63	-	-

Is very interesting to note how the slurries E and D have different characteristics from previous test (TAN slurry E during Test 1 is equal to 3,4 g/L and in the second part 2,5 g/L, for example). This difference is due to the high variability of the slurry: in fact, as explain before and as observed at real scale monitoring activity (see Chapter 4), the pig slurry characteristics change a lot during the time of pig breeding. In addition, it is possible to explain these differences (also pH is very different) for the storage time: the second batch of slurry, before to be used, are stored and it can be led to modification of chemical-physical characteristics (Perazzolo et al., 2017). Even if the new introduced slurry is a digestate, the pH is comparable with those one of other 2 types of slurry, as alkalinity, electrical conductivity. The main difference regards the chemical oxygen demand, and the content of solids. The ratio between BOD and COD is lower than 20%, this may mean that only a small part of organic compound is biodegradable: this can affect the ability of the denitrifying bacteria to transform NO_3^- into N_2. In addition, the percentage of volatile solids content in slurry D is very strange, and lower than those observed during Test 1. In the following graphs are reported concentration of nitrogen compound detected in the inlet and in the outlet samples withdrawn during *Test 2* (Figure 45).

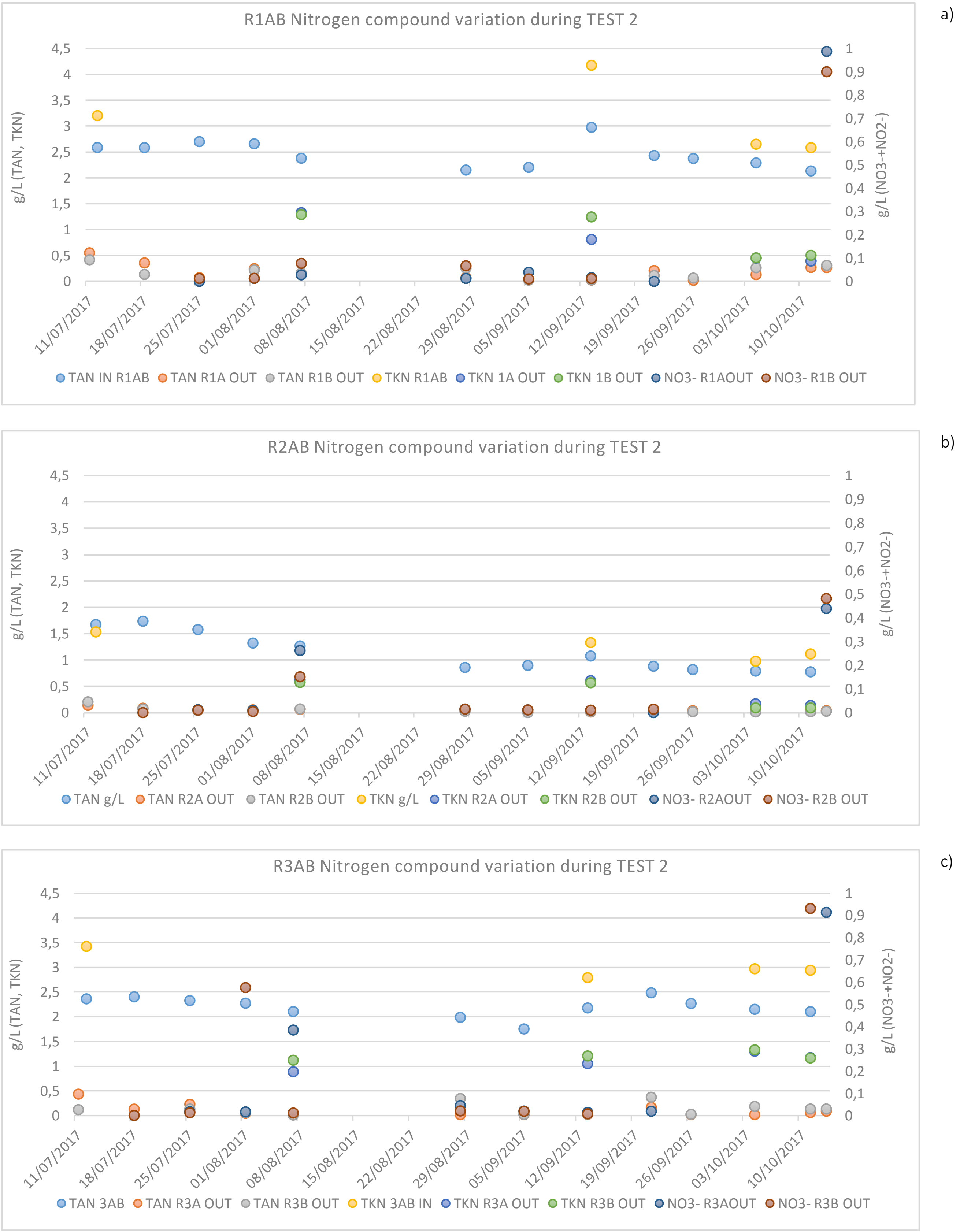


Figure 45 Characteristics of inlet and outlet samples for reactors R1AB (a), R2AB (b), R3AB (c) during *Test 2*.

Also for the considered period (*Test 2*), since the inlet flow rate was equal to the output flow rate, the minimum and maximum nitrogen yields removal achieved for the 1A & 1B reactors were 86,2%-99,1% and 81,6%- 99,0% respectively, for reactors 2A & 2B 91,35%-98,82% and 94,5%- 100% respectively, and for reactors 3A & B 90,28%-99,3% and 82,4%- 99,6% respectively.

Removal rate's range for all reactors remains above 85% during all the tests, contrary to what observed for *Test 1*. TKN removal yields, on the other hand, are lower and on average stand at 75% for R1AB reactors, 71% for R2AB reactors and 62% for R3AB reactors, but anyway higher than that observed in the first part of the tests.

In order to study the variation of pH and RedOx, the parameters detected during one day of functioning at the beginning of the experiments approximately, after 20 days, after approximately 2 months and at the end the test are considered for all pairs of reactors (Figure 46, 47, 48). The reported pH and RedOx value recorded every 15 seconds during one day of operation.

During the first period of operation, all reactors' pairs shown ORP and pH graphs with a strange path: the pH value did not change over time, and remained settled on very basic pH values. The trends of the RedOx potential during the 12th of July (Figure 46,a) for R1AB reactors were very strange and is reasonable that an operational malfunction occurred: it seems that the loading phase (negative peak of RedOx) was followed by the aeration phase and not by the mixing (denitrification one).

Although this problem did not occur and the loading phase was followed by the mixing one, even for the other reactor pairs (2AB and 3AB), the value of the RedOx potential increases without reaching values above 0, and thus guaranteeing (theoretically) the conditions to make denitrification takes place.

The removal rate reached during initial period (Figure 46,a) is equal to 86% and 92% for reactors R1A and B respectively. At the beginning of August 2017 the RedOx potential never reached value below 0 (Figure 46,b). This situation remains until September, when, as shown by the graph (Figure 46, c), the value reached, during denitrification phase, negative values. Nevertheless, the pH shown a peak after the start of aeration (red circle) and then a drop, as expected from nitrification.

The initial rise on the pH curve is caused by carbon dioxide stripping from the system and the rapid consumption of VFA that is produced during the anoxic phase (Kim et al, 2004). Still in the last phase of operation, the performance of the parameters remains almost the same, even if the pH varies in a more acidic range: this could mean the occurrence of nitrification. During time, despite the pauses imposed on the system (as explain in the paragraph *Materials Methods*) the yields increase and reach values of 99-97% for the 2 reactors. Concerning the pair of reactors R2AB, the removal rate reached during initial period is equal to 95% and 96% for reactor A and B respectively. The same considerations made for this operational phase for reactors R1AB remain valid also for R2AB. Also for that system, at the beginning of August, the value of RedOx potential never reached value below 0 (Figure 47, b).

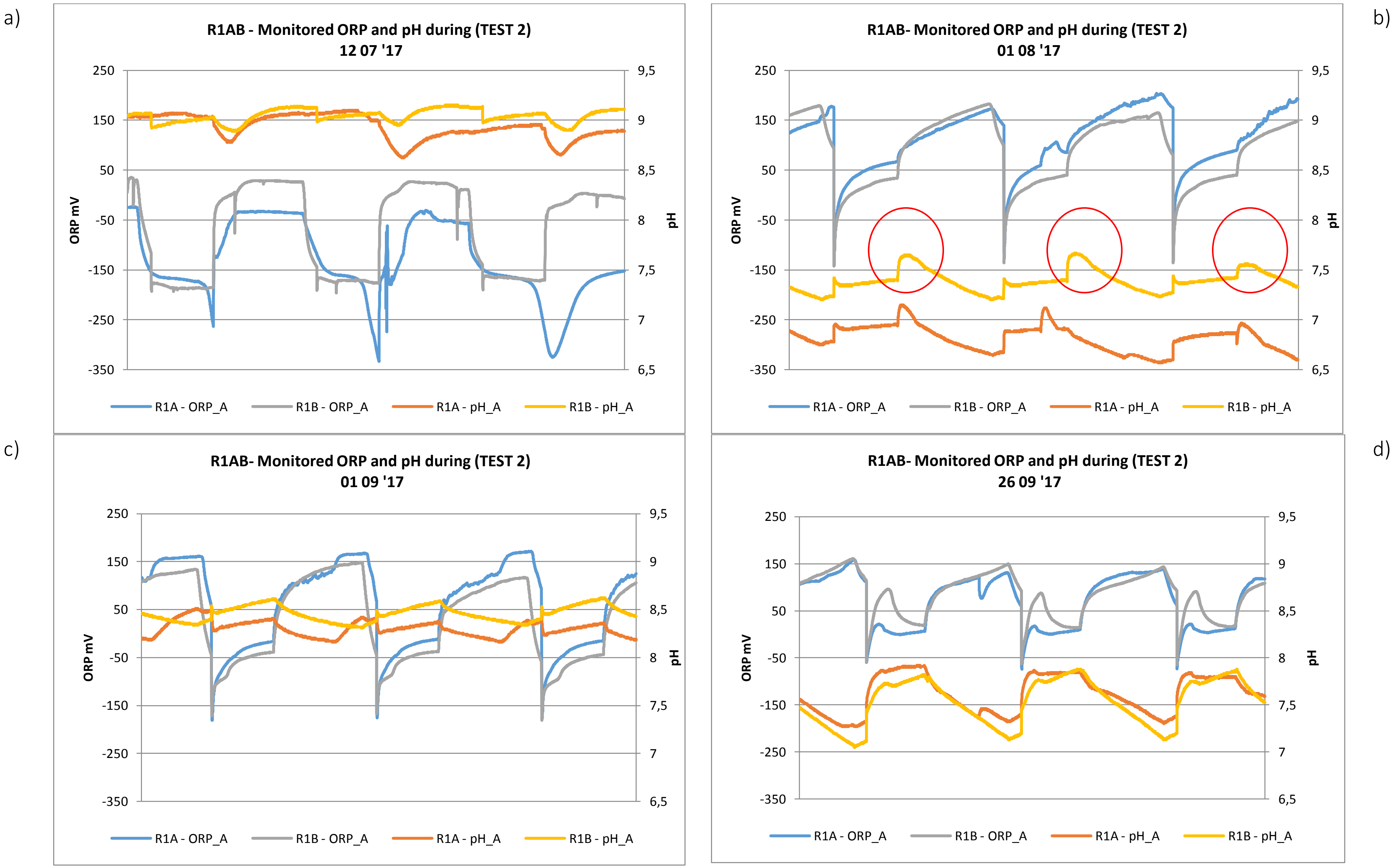


Figure 46 Trend of parameters monitored for reactors pair R1A e R1B during one day (24h), in particular: at the beginning of experiment (TEST 2), after about 1 month of operation, after about 2 and 3 months of operation.

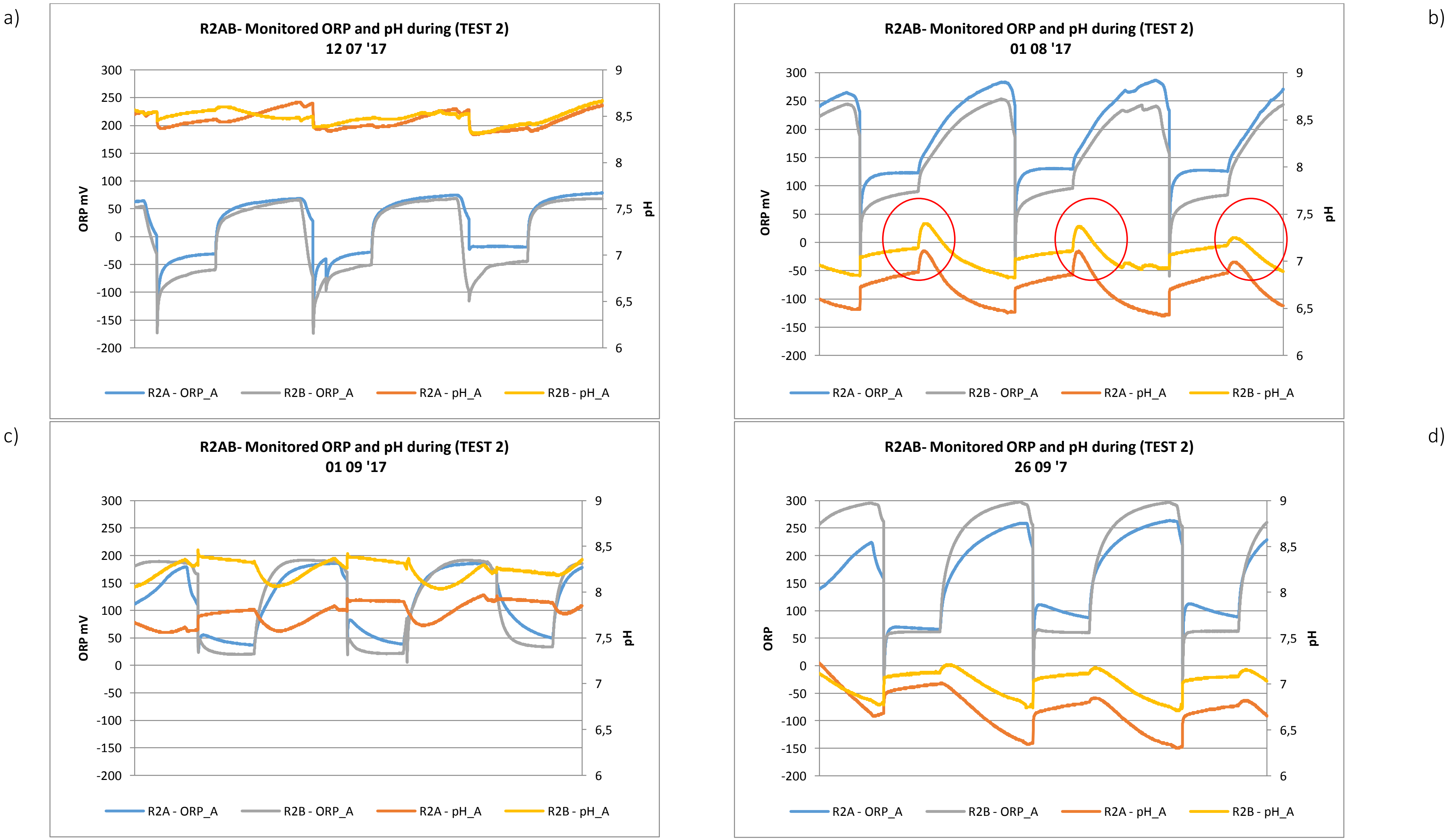
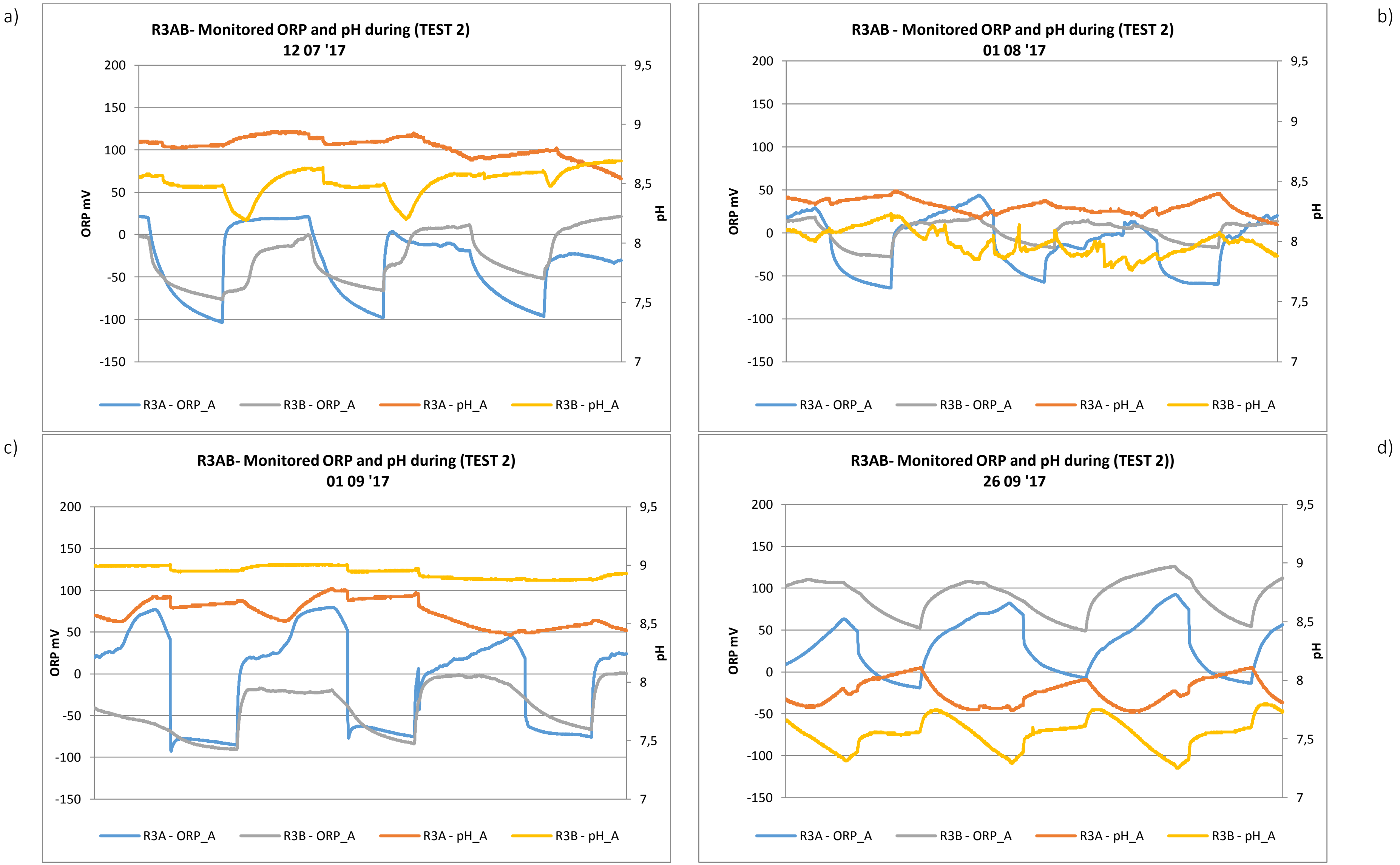


Figure 47 Trend of parameters monitored for reactors pair R2A R2B during one day (24h), in particular: at the beginning of experiment (TEST 2), after about 1 month of operation, after about 2 and 3 months of operation.


a) R3AB- Monitored ORP and pH during (TEST 2) 12 07 '17

b) R3AB - Monitored ORP and pH during (TEST 2) 01 08 '17

c) R3AB- Monitored ORP and pH during (TEST 2) 01 09 '17

d) R3AB- Monitored ORP and pH during (TEST 2)) 26 09 '17

Figure 48 Parameter trends monitored for reactor pairs R3A R3B during one day (24h), in particular: at the beginning of experiment (TEST 2), after about 1 month of operation, after about 2 and 3 months of operation.

This situation continued also in September, when, as shown by the graph (Figure 47, c), the value lingered in a positive span. Nevertheless, the pH, shown a peak after the start of aeration (red circle) and then a drop, as expected from nitrification. Also in this case, the greatest variation is that observed for the pH which went from basic values (pH> 8) to more neutral pH values (7-7,5).
However, the graphs obtained are not always comparable with the output obtained in the standard operation of SBR reactors. Analysing what happens for the reactors 3A and B, the yields obtained during the whole course of the test are attested on values ranging from 95-100% for reactor A and B respectively, and then decrease (slightly) during the time, reaching around 99% of removal rates in the last period (Figure 48,d). Considering the graphs obtained, it is generally possible to deduce that the quantity of air supplied was in excess of that required by the nitrification process; this statement is made by observing what happens during the mixing phase: it would be expected that the RedOx potential decreases. This could be caused by the oxygen present in the sewage, which, was released during mixing, raising the ORP value. As observed by Han et al., 2008, applying for example step-fed SBR process with the excess of aeration, the DO concentration at the end of aeration before feeding could increase and could lead to an aerobic waste of organic matters contained in the influent. In Table 35, are reported data concerning the input and output of each reactor during *Test 2* of tests in terms of daily load.

Table 35 Characteristics of the input and output in terms of daily load for 3 pairs of reactors during Test 2 of experiment (A: average; SD: standard deviation).

		TAN (g/d)		TKN (g/d)		ST (g/d)		SV (g/d)	
		A	SD	A	SD	A	SD	A	SD
R1AB	IN	1,15	0,12	1,47	0,34	8,78	3,26	3,26	2,01
R1A	OUT	0,07	0,05	0,35	0,20	11,22	3,36	4,84	2,39
R1B	OUT	0,07	0,06	0,41	0,21	10,26	3,29	4,03	2,29
R2AB	IN	0,53	0,17	0,58	0,11	4,43	0,49	1,32	0,51
R2A	OUT	0,02	0,01	0,18	0,12	5,92	1,48	2,16	1,06
R2B	OUT	0,01	0,01	0,15	0,13	5,87	1,44	2,09	1,07
R3AB	IN	1,03	0,09	1,41	0,13	13,20	2,56	6,76	1,88
R3A	OUT	0,03	0,03	0,51	0,08	12,42	1,26	7,04	1,08
R3B	OUT	0,06	0,06	0,56	0,04	13,31	0,70	7,65	0,59

During *Test 2* of the experiments, having imposed a constant hydraulic residence time, the inlet flow resulted equal to 0,47 L/d for each reactor. Despite the differences in characteristics of the slurry introduced into the system, the nitrogen load is comparable for all the reactors (0,16 gTAN/L/d for the pair R1AB, 0,08 gTAN/L/d and 0,15 gTAN/L/d for rectors R2AB and R3AB respectively).

The N-load resulting from the settings is lower than that applied in *Test 1* of the experiments. As said before, is very interesting also to note what occur for solids content: also, this characteristic has a great contribute to the reachable nutrient removal.

For all reactors the solid daily loaded are comparable for input and output samples. This fact means that the sedimentation phase did not work correctly. In fact, if the activated sludge in the reactor had a good sedimentation, only the clarified part is discharged. The high content of solids can also mean that a lot of organic material (volatile solid) it is released with the effluent and is not used by the bacterial biomass.

5.4. Test 3 and Test 4 (influence of operational parameters on removal efficiency)

After attested and examined the results obtained in the previous part of experiment (paragraph 5.3.), we decide to vary the operational set of SBR in order to study how different phases' length influences the nitrogen removal rate (*Test 3*). For doing that, all reactors were fed with the same slurry: in particular, we decided to continue only with the new **waste G**: a liquid fraction of pig slurry taken from a fattening pig breeding. That choice was done for focused the attention not on the characteristics of influent but on the duration of phases. In addition, the selection of using that type of slurry is a consequence of a consideration made on the chemical-physical characteristics of this waste: in fact, given that it was an out stream of at a closed-cycle farm, from the point of view of the composition, this guarantees a greater consistency along time. For all reactors an HRT equal to 8 days (corresponding to 0,875 L/d feed and draw) was imposed and SRT (sludge retention time was set equal to 35 d (corresponding to a sludge withdraw equal to 0,2 L/d).

Phase duration for reactor pairs R1AB is the same applied during *Test 2*. Instead, the reaction phase of reactor R2AB was modified: the aeration phase lengthening to check if nitrification is enough to convert all nitrogen in NO_3^-. About the adjustment done for reactor R3A and B, the number of cycles is decreased and the influent was fractionated. The discharge for all reactors' pairs must be carried out in two different phases: 7 seconds just after the aeration phase and the remaining 25 s after the sedimentation. The first phase of discharging is necessary in order to respect the SRT imposed: during this phase, in fact, with the effluent also the activated sludge was drained.

The second part of that slot of experiments (*Test 4*) is aimed to reduce the reactor load and to leave them in a long aeration phase in order to see if the nitrification phase takes place correctly. Besides all analysis done for check the reactors operation, during Test 3 and 4, the evaluation of sedimentation time for activated sludge was done.

5.4.1. Livestock manure and activated sludge characteristics

In the following paragraphs, the characteristics of used activated sludge and the inlet slurry used during experiment (Test 3 and 4) are shown in Table 36 and 37 respectively. The activated slurry had the same origin as that used in *Test 1 and 2*, but in order to re-start the different tests without any interference on the possible obtainable results, the new experiment was performed with a new batch of activated sludge. In comparison with the activated sludge used during the Test 1 and 2 (paragraph 5.3.3), the content of TAN and ST is much lower.

Table 36 Characteristics of activated sludge used during *Test 3* and *4*.

N-NH_4^+ (g/L)	N-($N0_2^-$+ NO_3^-) (g/L)	TKN (g/L)	pH	EC (mS/cm)	RedOx (mV)	ST%	SV/ST	COD mg/L
0,02	0,01	0,32	7,63	3,11	-169,0	0,56	59,74	4800

The chemical-physical composition of pig slurry used as inlet (Table 37) is comparable with the literature data. Linked with the slurry used in the previous tests, the content of TAN is lower, as the TKN and total solids.

Table 37 Characteristics of inlet slurry used during experiment (Test 3). (A: average; SD: standard deviation).

	TAN (g/L)	N-$N0_X$ (g/L)	TKN (g/L)	pH	EC (mS/cm)	RedOx (mV)	ST%	SV/ST	SS (%tq)	SS (%ST)	COD g/L
A	1,74	0,06	2,75	7,34	16,21	- 417,27	2,24	69,15	1,40	0,66	45,59
SD	0,12	0,02	0,26	0,26	3,44	20,96	0,66	2,26	0,50	0,06	13,55

The ratio between the COD and TAN and between COD and TKN is equal to 26 and 16,6 respectively: these values attest to the presence of carbonaceous substances useful for biological processes. Any consideration cannot be made on the biodegradability of wastewater, as BOD values are not available. In this part of tests, in fact, BOD analysis did not carry out. The process outputs obtained during the preliminary operation period did not give valuable results, and for these reasons are not reported. The malfunctioning of the systems attested in the first month was linked with the higher solid content accumulated in the reactor. In addition, during that period, the level of dissolved oxygen detected in each tank and value of RedOx potential were incongruous: the high values read by the DO probe were higher than 2 mg/L but the RedOx potential are lower than -200 mV. For these reasons, some system's check was done and the experiment was restarted. *Test 3* ended in February 2018 and *Test 4*, started immediately after Test 3, only lasted a month.

5.4.2. Experiments' results: Test 3

During *Test 3* not only the inlet and outlet sample are withdrawn but also the mixed liquor during the mixing phase (defined with _M). The results obtained are reported in Table 38. During the sampling of activated sludge, the reactor was in denitrification phase.

Table 38 Characteristics of activated mixed sludge detected during experiment (*Test 3*) for each reactor's pair. A: average, SD: standard deviation.

		$N\text{-}NH_4^+$ (g/L)	$N\text{-}N0_X$ (g/L)	TKN (g/L)	ST%	SV/ST	SS (%tq)
R1A_M	A	0,71	0,07		0,91	63,04	0,59
	SD	0,24	0,05		0,13	1,30	0,13
R1B_M	A	0,83	0,08	1,19	0,98	62,89	0,63
	SD	0,10	0,03	-	0,16	1,06	0,08
R2A_M	A	0,47	0,06	0,94	0,87	62,79	0,53
	SD	0,02	0,01	-	0,16	1,22	0,12
R2B_M	A	0,79	0,05	1,02	0,91	63,21	0,57
	SD	0,18	0,00	-	0,20	0,63	0,14
R3A_M	A	0,42	0,04	0,85	0,79	57,63	0,36
	SD	0,09	0,01	-	0,20	12,94	0,15
R3B_M	A	0,59	0,04	1,05	0,75	64,73	0,43
	SD	0,028	0,003	-	0,16	5,79	0,14

The most interested value that need an explanation is the content of suspended solids in the reactors. In fact, considering that the common measure detected in activated sludge system is 3-5 gSST/L, the values here reported are higher, except for what observed for the pair of reactors 3A and B. This fat can affect the quality of sedimentation of the sludge.

In Table 39 instead, are reported the effluents' characteristics for each reactor. First, considering each pair, the characteristics of the outputs of the two reactors are comparable. TKN and TAN parameters were lower than 1 g/L for all reactors and the concentration of solids content was lower than 5,4 g/L. In addition, the concentration of suspended solids was very low (< 1 g/L) and the percentage of the volatile solids compared to that one detected in the inlet flow was reduced, indicating that a degradation process has occurred. Since the time of sedimentation is longer for the 3A and B reactors, a RedOx was expected to be more negative than those found in the other reactors: however, the measure in that case was higher. This can also be explained considering that, for the last couple of reactors, the aeration period length was almost double than others, and therefore the "residual" oxygen that remained in the activated sludge at the end of the anoxic period can explain the value of the RedOx.

The pH of outlet samples was in a neutral range, and electrical conductivity is about 7 mS/cm for all samples, much lower than those one detected in the inlet flow. In order to calculate the removal rate (Table 40) reached during *Test 3* of these experiments, the daily load of each components have been defined.

Table 39 Characteristics of outlet of each reactor detected during experiment (*Test 3*) for each reactor's pair. A: average, SD: standard deviation.

		TAN (g/L)	NO_X (g/L)	TKN (g/L)	pH	EC (mS/cm)	RedOx (mV)	ST (%tq)	SV/ST (%)	SS (%tq)	COD (g/L)	Alk T (g/L)
R1A	A	0,57	0,05	0,80	7,33	7,21	-202,2	0,35	41,60	0,08	1,15	2,11
	SD	0,08	0,02	0,14	2,61	1,91	35,1	0,04	2,79	0,10	0,02	0,59
R1B	A	0,76	0,05	0,91	7,64	7,79	-137,7	0,54	49,01	0,08	1,13	2,52
	SD	0,27	0,02	0,30	2,13	1,84	38,7	0,27	9,47	0,03	0,01	0,52
R2A	A	0,71	0,03	0,75	7,30	7,47	-124,4	0,48	40,59	0,12	1,25	2,35
	SD	0,27	0,01	0,30	2,74	0,62	9,8	0,12	12,01	0,12	0,146	0,96
R2B	A	0,76	0,04	0,91	7,96	7,94	-83,8	0,40	43,38	0,07	1,24	2,81
	SD	0,18	0,01	0,07	1,59	1,80	83,1	0,08	3,47	0,04	0,116	0,40
R3A	A	0,68	0,04	0,97	7,62	7,63	-89,3	0,42	46,20	0,12	1,28	2,70
	SD	0,15	0,01	0,11	2,29	1,57	14,4	0,06	4,36	0,09	0,158	0,68
R3B	A	0,69	0,05	0,84	7,73	7,01	-92,9	0,39	44,65	0,09	1,17	2,45
	SD	0,20	0,03	0,35	2,01	1,40	4,0	0,06	2,22	0,06	0,014	0,46

Table 40 Removal rate reached during Test 3.

	TAN	$N\text{-}(NO_2^- + NO_3^-)$	TKN	ST	SS	COD
R1A_S	67,2%	16,7%	70,9%	84,4%	94,3%	95,4%
R1B_S	56,3%	16,7%	66,9%	75,9%	94,3%	94,5%
R2A_S	59,2%	50,0%	72,7%	78,6%	91,4%	94,8%
R2B_S	56,3%	33,3%	66,9%	82,1%	95,0%	93,8%
R3A_S	60,9%	33,3%	64,7%	81,3%	91,4%	94,1%
R3B_S	60,3%	16,7%	69,5%	82,6%	93,6%	94,6%

Generally, the removal rate achieved is lower than outcomes observed during the Test 1 and 2. Analysing the behaviour of each pairs of reactors the removal rate obtained is not always comparable.

In these experiments, the inlet slurry used was the same for all the reactors, and the only thing that varying was the number of cycle (R3AB) and the duration of aeration/mixing phase. So, different removal yields would expect when these settings changed. However, considering for example TAN, the results do not allow to establish whether a setting yields better results than other.

Considering the average yield obtained from the reactor pair, the highest removal rate for total solids is 81,95% for reactor 3A and B: this can also be explained by the longer sedimentation time applied for these reactors compared to the other pairs.

Regarding the removal yields obtained for carbonaceous substance, the maximum removal yields was 94,95% for couple R1AB. This can be signified that the aeration phase length for couple 1 was not sufficient for the establishment of the nitrification process: heterotrophic bacteria degrade the carbonaceous substances, but the autotrophic ones are unable to carry out their activity. The increase in the aeration time imposed for R2AB reactors (1 h) compared to R1AB reactors does not allow considering the quality of the effluent obtained significantly different. The settling time of 0,2 h applied for the R2AB pair was enough for obtaining a clarified waste with a solids content equal to 4-5 g/L. The fewest number of cycles imposed for the 3AB pair (2 cycle/day), does not seem to be the best solution.

Considering the times allocated for the different phases (please also see Chapter 3), the significant points that allow the eventual application of real time control of the process do not appear in the graphs.

The load fed at 0,214 gTAN/L/d (or 5,7 gCOD /L /d) was too high to allow removal of the nitrogen load > 60%. In the Table 41 are reported nitrogen (as TAN) and COD load for each reactor.

Table 41 Nitrogen (as TAN) and COD loading rate for each reactor during Test 3.

	gTAN/gSST/d	gCOD/gSST/d
R1A_M	0,04	0,97
R1B_M	0,03	0,90
R2A_M	0,04	1,08
R2B_M	0,04	1,00
R3A_M	0,06	1,58
R3B_M	0,05	1,33

Also for Test 3, in order to study the variation of pH and RedOx, parameters detected during one day of functioning at the beginning of the experiments and during its operations are considered for all pairs of reactors (Figures 49, 50, 51). The reported pH and RedOx value recorded every 15 seconds during one day of operation.

For reactors R1A and B is possible to recognize the 3 cycles per day that occurred, even if, as shown during the first day of operation (Figure 49,a) a malfunctioning occurred during last cycle of the day.

The system R1A and R1B shown always a pH trend very flat, without any noticeable change.

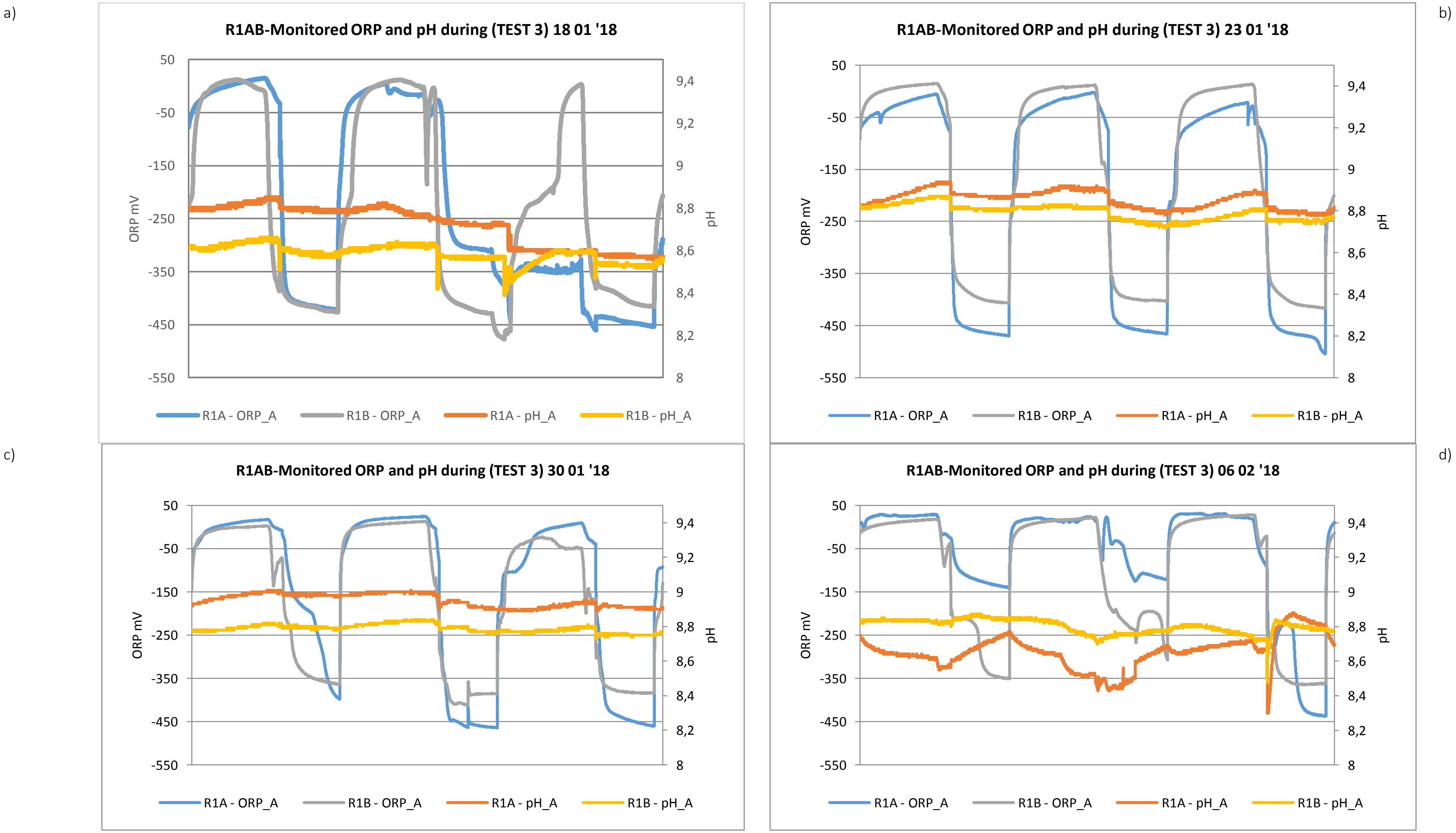


Figure 49 Trend of parameters monitored for reactors pair R1A R1B during one day (24h), in particular: a) at the beginning of experiment (TEST 3); b) during 23/01/2018; c) during 30/01/2018 and d) at the end of experiment 06/02/2018.

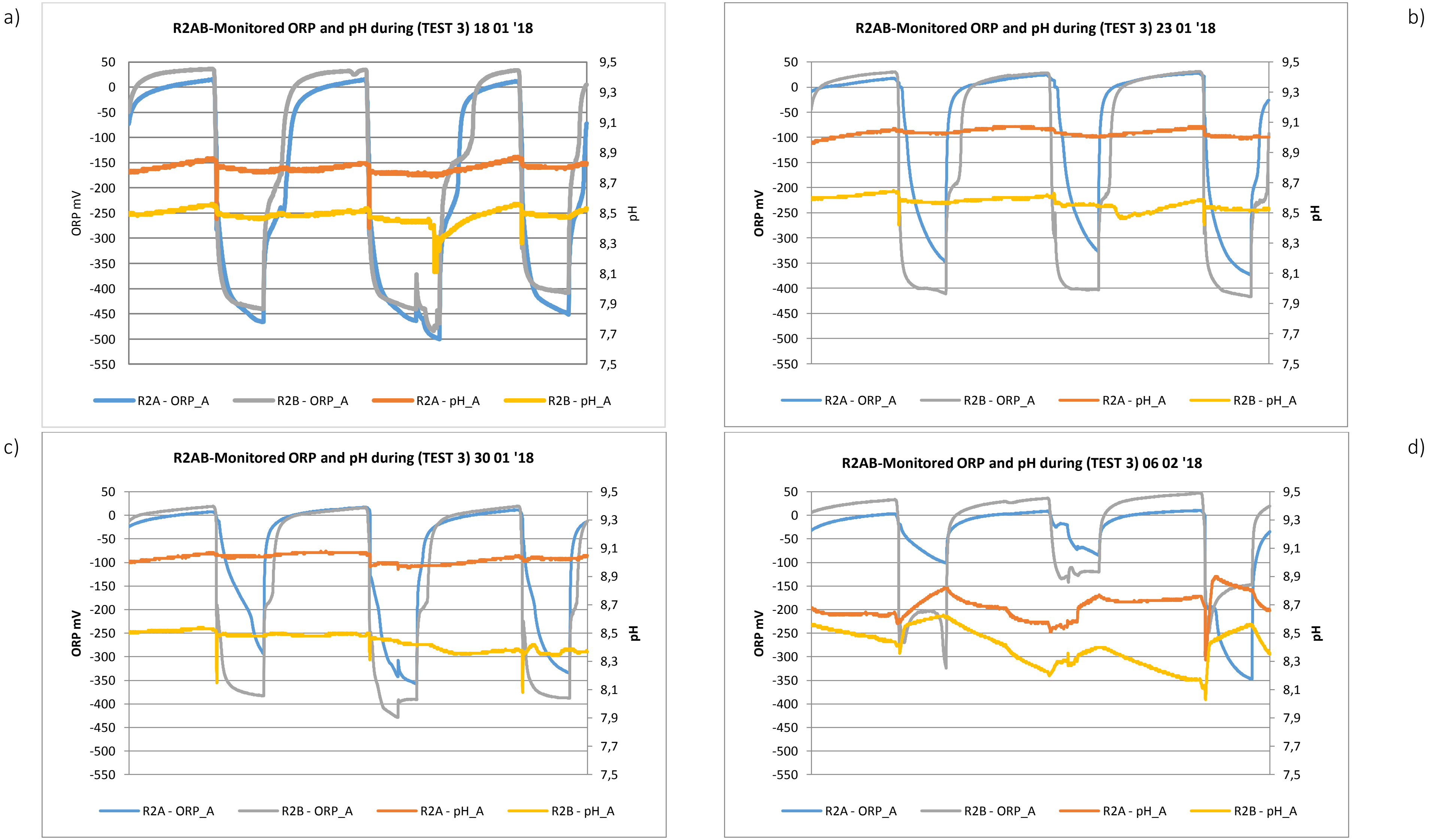


Figure 50 Trend of parameters monitored for reactors pair R2A R2B during one day (24h), in particular: a) at the beginning of experiment (TEST 3); b) during 23/01/2018; c) during 30/01/2018 and d) at the end of experiment 06/02/2018.

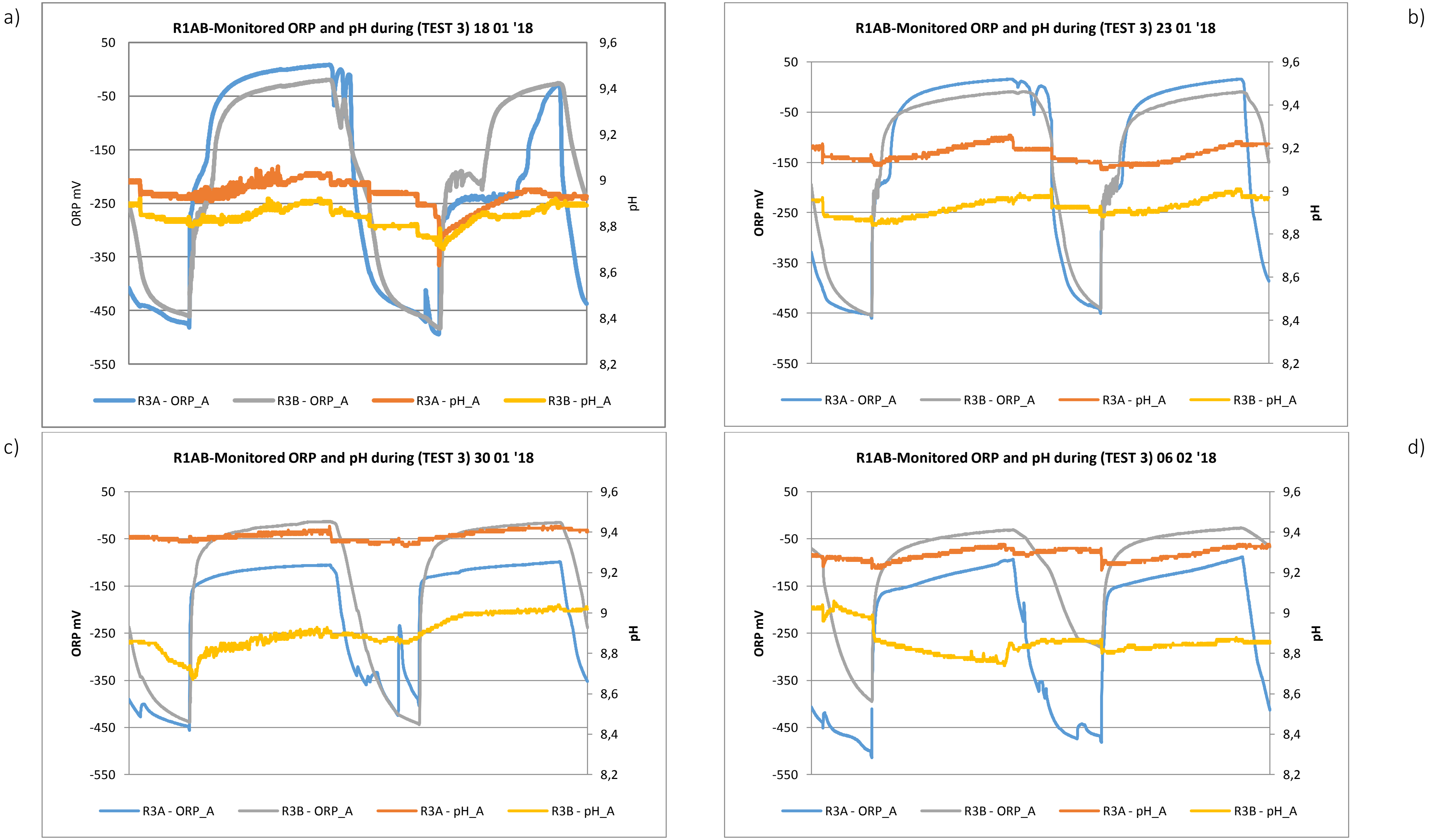


Figure 51 Trend of parameters monitored for reactors pair R3A R3B during one day (24h), in particular : a) at the beginning of experiment (TEST 3); b) during 23/01/2018; c) during 26/01/2018 and d) at the end of experiment 06/02/2018.

Only in the final part of experiment (Figure 49,d) it is possible to distinguee the decrease in pH in conjunction with the aeration period. During other days, the path was not as expected: during denitrification phase, the pH increase, maybe for the stripping process that involved NH_3.
The ORP was always negative during denitrification phase, until reach values lower than -450 mV. Contrary, during the aeration phase, positive values were attested.

Also concerning R2A and B reactors (Figure 50), the situation seems similar, even if the air phase duration is longer. The pH does not show the expected typical trend during any day of observation.
The "ammonia valley" does not appear and the ORP, despite having positive values, for a short time remains above 0 mV: the aeration period may not be adequately long. This can also be deduced from the high values of removal of the carbonaceous substance: the nitrification phase didn't start and only the deterioration of the COD occurs. Also in this case the process seems to have undergone a change during the last period of operation (Figure 50,d).
The same considerations can be made for the R3AB pair (Figure 51), even if the differences between the setting operations is completely different: also in this case, the longer duration of aeration phase does not allow observing the occurrence of typical hot points. For reactors R3A and B, during last period of operation is detected a difference in the pH and ORP path: the two reactors had different behaviour. Nevertheless, as seen above, the removal values of the nitrogen compounds are higher than 56%.

In order to understand better different situations, as said before, during Test 3 also the activated sludge characteristics was monitored.
At the beginning of this series of experiments, although the initial settings were the same, the activated sludge showed a different sedimentation capacity (Figure 52). In addition, the two reactors of the pair, shown different behaviours. The reactors 1A and 2A in 1 hour can reach Mohlman index equal to 37 mL/SS and 41 mL/g that indicated a good quality of the sludge.
The same cannot be said for reactors 1B and 2B, which have a worse quality of sludge sedimentation.
Furthermore, the reactors 3 exhibited the worst quality of the sludge.
Nevertheless, acting on the regulation of the air entering the reactors in order to ensure a suitable content of dissolved oxygen (an operation that was done daily), the sludge presents a gradually better quality during the course of the experiment, reaching levels almost comparable in all and 6 reactors.
At the end of experiment, the situation was completely different in comparison with the beginning of experiment: the reactors 3AB had the best SVI index.
Also concerning that pairs, during the 30/01/2018 something has happened in the reactor A, and the and the quality of the activated sludge was very bad.

Figure 52 The performance of the activated sludge-water interface during the time for each pair of reactors on different days: a) 18/12/2018, b) 23/01/2018, c) 30/01/2018, d) 06/02/2012.

However, by correcting the amount of air supplied, in a week, the sedimentation of the activated sludge returned to improve. These malfunctions could be caused by the presence of filamentous bacteria that interfere with the good sedimentation of the sludge. These bacteria form colonies with a high surface/volume ratio and therefore with a reduced tendency to sedimentation. This phenomenon is favoured by situations of substrate limitations or as more plausible in this case, of oxygen lacks.

5.4.3. Experiments' results: Test 4

During ***Test 4***, a long aeration phase was applied. The results of the characteristics detected for the inlet flow and for the outlet flow of each reactors were reported in Tables 42.

Table 42 Characteristics of the input and output in terms of concentration for 3 pairs of reactors during Test 4 (A: average; SD: standard deviation).

		TAN (g/L)	NO$_X$ (g/L)	TKN (g/L)	pH	EC (mS/cm)	ST (%tq)	SV/ST (%)	SS (%tq)	COD (g/L)	Alk T (g/L)	Alk VFA (g/L)
IN	A	1,00	0,01	1,36	7,59	4,85	0,93	66,03	0,63	11,92	4,39	2,15
	SD	-	-	-	-	-	0,33	6,40	0,42	-	-	-
R1A	A	0,07	0,05	0,36	7,25	2,43	0,81	61,06	0,56	2,58	0,21	0,07
	SD	0,03	0,03	-	0,83	-	0,13	1,87	0,19	3,63	0,30	0,10
R1B	A	0,06	0,02	0,39	7,51	2,26	0,93	61,93	0,65	2,70	0,52	0,17
	SD	0,04	0,02	-	0,64	-	0,07	1,85	0,14	3,81	0,74	0,24
R2A	A	0,08	0,01	0,42	7,46	2,35	0,85	61,27	0,60	2,80	0,28	0,10
	SD	0,03	0,01	-	0,88	-	0,10	2,07	0,16	3,95	0,40	0,14
R2B	A	0,06	0,01	0,44	7,67	2,15	0,88	60,15	0,76	3,52	0,45	0,15
	SD	0,03	0,01	-	0,37	-	0,25	7,49	0,11	4,96	0,64	0,21
R3A	A	0,10	0,01	0,45	7,50	2,57	0,97	62,56	0,73	3,30	0,32	0,10
	SD	0,06	0,00	-	0,51	-	0,21	2,32	0,25	4,66	0,45	0,14
R3B	A	0,08	0,01	0,40	7,55	2,32	1,03	64,97	0,51	3,00	0,30	0,09
	SD	0,04	0,00	-	0,68	-	0,76	11,84	0,07	4,23	0,43	0,12

In the following are reported concentration of nitrogen compound detected in the inlet and in the outlet samples withdrawn during *Test 4* (Figure 53).

In order to calculate the removal yields reached during that part of experiment, the inlet and outlet load are considered.

The removal rate for TAN are higher for couples of reactors, and always greater than 90%. The long period of aeration was sufficient to allow nitrification to be established and, therefore, to considerably reduce the amount of ammonia nitrogen. Also the nitrogen removal was very high and always greater than the 66%. Concerning the removal of carbonaceous compounds was attested above 70%.

Figure 53 Nitrogen concentration detected in inlet and outlet samples for reactor R1AB (a), R2AB (b), R3AB (c) during Test 4.

The lowering of the imposed nitrogen load and the prolongation of the aeration phase make possible to achieve higher removal yields than those observed during Test 3. Nevertheless, the effluent from the Test 4 had a high solids content: sedimentation does not occur optimally.

5.5. General final observation

5.5.1. Test 1 and Test 2

The yields obtained during both Test 1 and Test 2 are very high, and higher than observed at full scale (Chapter 4). Although a detailed analysis during the complete cycle of the reactor (for example analysis at the end of nitrification/denitrification, etc.) has not been carried out, with the collected data available, it is possible to state that the obtained treated effluent has a reduced content of nitrogen, and, therefore, that the biological process of denitrification/nitrification took place during experiments.

Working at constant nitrogen load and variable flow (as done in the *Test 1* of experiment), it is not an easy management choice, nevertheless when a very low nitrogen load (0,1 gN/L/d) was applied, the removal rates obtained were very high.

Also considering the *Test 2*, where the fixed value was HRT, the achieved nitrogen removal yields were higher when the nitrogen in the inlet flow is lower.

The obtained elevated removal yields, however, could be also connected to stripping phenomena that can occur during the aeration phase. In fact, the content of ammonia present in the slurry (always upper than 2 g/L as TAN, except for slurry C), associated with the high pH could promote the removal of NH_3 through the air flow applied for the oxygen supply. To attest this possibility, which would explain that part of the nitrogen content in the slurry was removed by stripping and not by the biological process, other types of measures can be made.

At first it would be useful to deepen with respirometry if the nitrifying activity is present in reactors; moreover, by capturing the gas leaving the reactors and analysing them, a mass balance should be carried out to obtain the quantity of NH_3 actually nitrified.

Anyhow, from the results obtained, it is not possible to deduce any clear result on the correlation of types of slurry and removal of ammonia nitrogen: for all the cases considered, the yields of removal of the TAN are very high.

However, if the removal rates of TKN are analysed, during *Test 1*, at the same imposed TAN load, the slurry for which was observed the lower removal efficiency was slurry fed to the 3AB reactors (<50%) (pig slurry after a solid/liquid process). Nevertheless, for these reactors, the variability of the collected data was very high (as attested also by the high standard deviation). Also for reactor 3AB, during *Test 1*, the highest

efficiency of total solids removal is attested, which, as said, is evidence of a good sedimentation of the activated sludge.
With the change of systems' operation, and of type of slurry (digestate instead liquid fraction of pig slurry) the obtained result of TKN removal rate was higher for all pairs of reactors, even if the removal rate of solids content remains intangible. During ***Test 2***, the operation was more constant, without too much variation, and as underline, the TAN removal was higher. The best results occurred for slurry D.

In addition, as underlined before, the system in any set operation (except for reactor 3AB during Test 1), cannot remove the solids content. This is surly related to both the very high solid contents of the slurries (3-4 time higher than the classical concentration of typical activated sludge system e.g. 3 gSSV/L), and to sedimentation time that can be too short for the proper activated sludge sedimentation. In order to obtain a good quality of outlet slurry in terms of solids content, the definition of this phase of SBR operation is crucial.

Ammonia valley and nitrate knee (which appeared during the NDN process in the SBR reactor is widely explained and reported in Chapter 3) are not easily identifiable in our experiments.

In conclusion, despite the good results attested during the different experiments, is necessary to analyse and characterize not only the input and output flows enter and exit the reactors, but also what happen during the cycle. For this reason, it could be useful to examine the content of nitrogen as TAN, NO_3^- during the operation of single phase: they would be indicators of the actual progress of the process (nitrification and denitrification).

5.5.2. Test 3 and Test 4

The removal rate reached is lower than those observed using other type of slurry (see *Test 1* and *2*).
Nevertheless, although a detailed analysis of what happens during the complete cycle of the reactor has not been carried out, with the collected data available for each reactor, the effluents have a reduced content of nitrogen.

The different operating settings of the 3 reactors pairs during test C did not allow to define which one of the three operational settings was the best applicable configuration.
Nevertheless, the solution applied to the reactors 3A and 3B, which provides 2 cycles per day with the very long aeration phase, is the least indicated solution. Operating shorter cycles, splitting the total flow fed to the plant daily, as they also attest to scientific literature, is the most frequent choice.

Increasing settling times to the detriment of the denitrification phase (as applied for R2A and B reactors), does not make significant changes to the quality of the output flow, compared to lower settling times (R1AB) and greater denitrification times.

The HRT applied in that experiments (Test 3 and 4) was lower than that applied for other experiments. However, since during the experimentation A the variability of the HRT was very high (1 d- 20d, depending on the type of slurry) and the yields obtained were still greater than those observed for Test 3, the lower nitrogen removal rate was not immediately attributed to this parameter.

As also attested during Test 1 and Test 2, the highest nitrogen-loading rate applied in Test 3 caused a lowest removal rate achievable. This observation is comparable with those one done by Poo et al., 2005: they found that when the loading became very high (about more than 0,5 kgNH_4^+-N/m^3/d), the ORP was depicted abnormally. The explanation of this phenomenon is might be the high suspended solids concentration of 13 g/l accompanied with high NH_4^+-N loading (Figure 54).

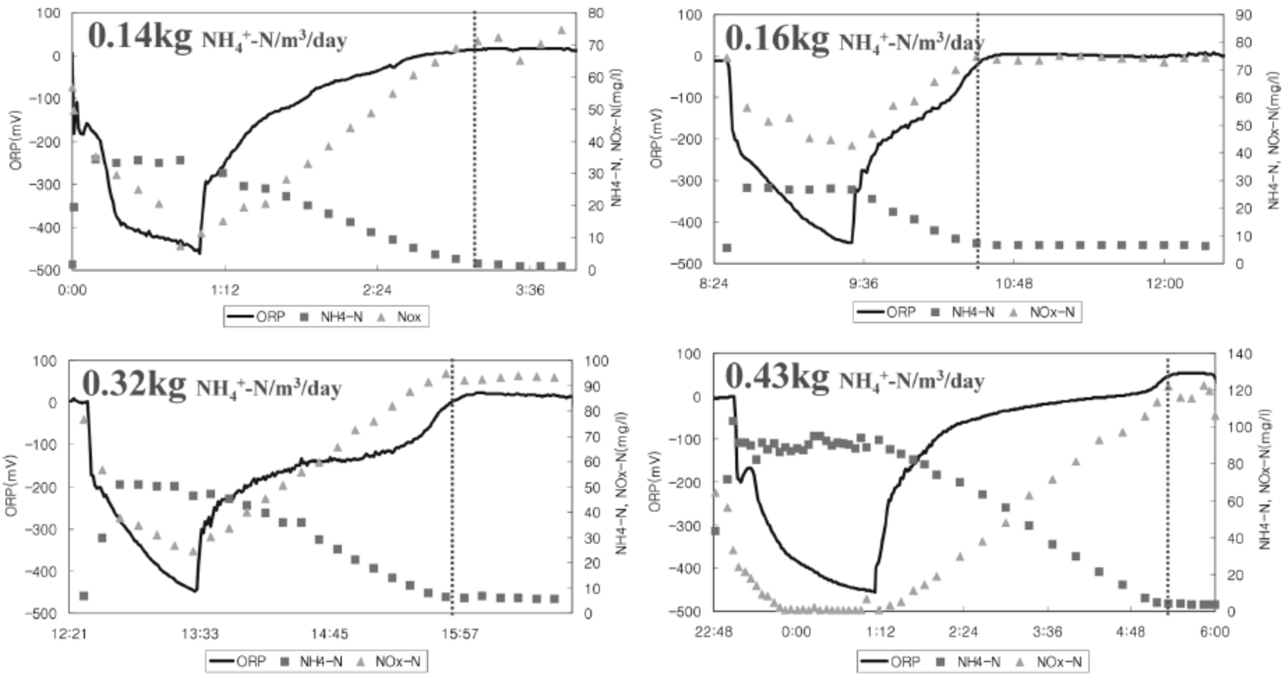


Figure 54 ORP and nitrogen profiles under various loading rates during one sub-cycle during SBR operation (From Poo et al., 2005).

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SECTION 3 - LIVESTOCK MANURE TREATMENT: THE PROBLEM OF AMMONIA AND GREENHOUSE GASES EMISSIONS AND THEIR EVALUATION

6. Assessing NH_3 and GHG emissions from livestock manure storage: comparison of measurements with dynamic and static chambers

The content of this chapter is used for preparing an article that will be submitted in "Atmospheric Environment".

6.1. Introduction

Although animal manure represents precious nutrients for plants, it can be considered cause of soil, water and air pollution if incorrectly handled (Sommer et al., 2003).

Regarding air pollution, agriculture represents the mainly source of anthropogenic ammonia (NH_3) emission (90%) (Seidel et al., 2017), malodors gases, such as sulphur compound organic acids, phenolic compounds and indoles (Sommer et al., 2013).

In 2014, agriculture was the great emitter of NH_3 (94% of total emission in EU-28) and represents the sector providing the minor emission reduction (only 7% from 2000 to 2014) (EEA, 2016). Gaseous ammonia in the atmosphere leads also to formation of secondary inorganic aerosol, the main constituents of anthropogenic PM2.5 emissions. Fine particulate matter lead to onset of lung cancer and cardiopulmonary disease (Bauer et al., 2016). Indirectly, the NH_3 emissions also contribute to N_2O emissions (Sommer et al, 2013).

Emissions from livestock manure and in general livestock production regard greenhouse gases (GHGs) which include methane (CH_4) and nitrous oxide (N_2O).

The quantification of emissions from the agricultural sector and especially from livestock manures is relevant for inventory purpose and to assess the reduction that can be obtained adopting mitigation techniques (Provolo et al., 2017; Misselbrok et al., 2016). Therefore, suitable methods for the determination of emission from manures are required.

Several devices can be used to obtain volumetric concentration of ammonia. However, in order to define ammonia emissions (emission rate) the ammonia mass concentration and airflow rates are also required.

There are different methods to estimate the emissions rate among which it is possible to count (Parker et al. 2013):

1. the *mass balance approach*, based on the difference of the ammonia nitrogen manure content and composition of manure before and after the emission period considered;
2. *indirect methods*, that use dispersion models and measured ambient concentrations for the calculation of source emission rates. Among indirect methods, there are also micrometeorological techniques that foresees gas emission calculation from a large footprint using tower-based instrumentation (e.g., gradient methods, eddy covariance, backwards Lagrange dispersion model);
3. IPCC approaches;
4. *direct methods,* where the fluxes measure and calculation are made with portable wind tunnels or flux chambers.

As underlined by other authors, the different approaches do not give comparable emission rates and the variances among methods can be greater than one order of magnitude (Parker et al., 2013).

Usually in order to select the appropriate method to measure emissions, besides the final purpose, is necessary to evaluate different factors like the type of gas compounds considered, the range of measurements, and the homogeneity of the source.

The direct methods are commonly applied for earth-atmosphere and water-atmosphere fluxes of various compounds including ammonia, biogenic sulphur, nitrogen oxides, and methane and generally greenhouse gases (Pumpanen et al., 2004; Pihlatie et al., 2013; Dennehy et al., 2017).

The direct used techniques applied are mainly two: the closed static chamber (no steady- state chamber) and the open dynamic chamber (steady-state chamber).

The first one is a physical enclosure that create a limited headspace over the surface from which is necessary to estimate emission. Inside the chamber the only flux presents is generated by gradient concentration between the source and the headspace and will be quantified during time in order to obtain the undisturbed flux into or out of the emissions source (Pihlatie et al., 2013). The concentration gradient decreases during time until reaches a saturation condition. In this case, the emission is calculated considering the curve before it reaches saturation.

Many studies use this direct method for emission's measure (Miola et al. ,2015; Zhang et al., 2011; Pumpanen et al., 2004; Rochette et al., 1992).

Differently, in the open dynamic chambers an air flux passing through the hood, flushing out the headspace over the chamber. Airflow can be natural (environmental wind) but in most cases is generated artificially with a fan. Also in this case is possible to find in literature many studies that apply this methods for measuring emission flux (Perazzolo et al., 2015; Dinuccio et al., 2008; Shah et al., 2006; Parker et al., 2009; Yang 2017; Zhang et al., 2011).

Direct method technique (both static and dynamic) presents some limitation linked with a possible disturbance inducing for example by spatial variability of emission source, chamber size, air velocity (di Perta et al. 2016; Saha et al. 2006), alteration of natural gradient between atmosphere and emissions source. Moreover, the use of chambers cannot allow obtaining representative emission rates of absolute value of gas emissions from a given source, being the measurements connected with the site, time, specific condition, sampling campaign etc. Nevertheless, the errors introduced by using of chambers can be diminished with correct chamber designs, the proper data analyses, and spatial and temporal sampling regimes (Davidson et al., 2002). In addition, the advantage in using chambers is to associate a particular emission site to its measurable array of physical, chemical, and microbiological properties with emissions of particular compounds or their reaction products (Aneja et al., 2006). As well, the investment cost of flux chambers is low and their use are generally easy (Ogink et al., 2013).

Nevertheless, for testing ammonia volatilization models, wind tunnels are more suitable than static chambers because better mimic natural airflow, even if cannot be mimic wind condition. Wind tunnels technique can be applied at small-scale range. However, the investment cost for wind tunnels are high (Yang et al., 2017) and the obtained results are related to a fixed and immobile time scale.

Compared with an indirect method, for example with IPCC guidebook, direct methods allow to consider specific situations for which the guides do not provide specific emission factors (as emissions derive from by-product storage from a processing plant) (Torrellas et al., 2018).

The numerous and variety existing techniques for measuring NH_3 and greenhouse gases concentrations under agricultural field conditions show as a uniquely known technique is not yet defined and used. However, the choice of proper ammonia emission evaluation method is crucial and the need to rely on the data obtained from the gases measurements is fundamental both for the research aims and mostly for the proper definition of mitigation techniques and interventions aimed at reducing emissions.

The overall goal of this study was the evaluation and comparison of the use of different technologies applying direct methods (two steady-state chambers and one static chamber) for the monitoring of ammonia and greenhouse gas emissions. The comparison of the results obtained applying direct evaluation methods was done for an ammonia solution and for stored slurry in three different full-scale treatment plants.

6.2. Materials and methods

6.2.1. Experiments set-up

In order to evaluate and compare the use of different hoods for measuring ammonia and other emission gases, two different types of experiment were carried out.

The first group of tests were performed using a synthetic source of ammonia (synthetic ammonia solution) in order to assess and study ammonia emission in a controlled condition without any disturbance induced by the complex characteristic of animal slurry.

The second group of experiments, which goals was also the comparison of the obtained results using synthetic ammonia solution, were carried out at filed condition, applying the use of hoods for measuring ammonia and GHGs emissions from animal slurries and agro-industrial waste.

The experiments in controlled conditions were carried out at IRTA (Institut de Recerca i Tecnologia Agroalimentàries), located in Caldes de Montbui, Barcelona (41,612140, 2,169617). For these experiments, an ammonia solution was prepared adding ammonium hydroxide extra pure, 25% solution in water (≥25% as NH_3) (Acros Organics) to different amounts of water in order to obtain the desired ammonia concentration. Trials were performed in a period of 6 months.

The ammonia solution is placed inside a heavy-duty PVC and polyester square containers with triple-layer side walls enclosed in a metal frame (1,22 m x 1,22 m; A: 1,49 m^2). Specifically, the plastic vessel is filled with 30 L of tap water mixed with a defined amount of NH_4OH, based on the required ammonia concentration. In addition, a group of tests was performed with ammonia solution added with an amount of sulphuric acid 95-97% in order to drop pH from 12 to 7,8 and sodium bicarbonate for simulating the buffer capacity of the slurry ($NaHCO_3$) (concentration: 0,1 mol TIC/L, where TIC is total inorganic carbon defined as : TIC= $CO_{2\ aq}$ +$HCO_3^-{}_{aq}$ + H_2CO_3).

During all experiments, the content of ammonia was determined also analytically.

In every sampling campaign carried out at field conditions, the chamber and hoods were placed in the same point.

A summary of experiments carried out both with ammonia solution and using animal slurries and agro-industrial waste is reported in Table 43.

Table 43 Summary of all experiments. DH1: dynamic hood1, DH2: dynamic hood 2; SH1: static hood 1. C: compost, LFD: liquid fraction of digestate; SFPS: solid fraction of pig slurry.

Hood	**Request concentration mgN-NH^3/L**	**Buffer (y/n)**	**Exp. Replicates**
	AMMONIA SOLUTION		
DH1	0,5	n	3
	1	n	3
	2	n	3
		y	3
	3	n	3
DH2	0,5	n	6
	1	n	6
	2	n	6
		y	3
	3	n	3
SH1	0,5	n	1
	1	n	1
	2	n	4
		y	3
	3	n	3

Hood	**Type of slurry**	**Buffer (y/n)**	**Exp. Replicates**
ANIMAL SLURRIES AND AGRO-INDUSTRIAL WASTES			
DH1	C	-	3
	LFD	-	3
	SFPS	-	3
DH2	C	-	3
	LFD	-	3
	SFPS	-	3
SH1	C	-	3
	LFD	-	3
	SFPS	-	3

Trials at full scale, using different hoods for estimating emission from livestock manure were performed:

- (a) in a biogas production plant located in north of Catalunya, Spain, where cow slurry is digested together with other co-substrates, agri-food wastes, sludge from wastewater treatment plant etc.; here the emissions of ammonia, CO_2, CH_4 and N_2O were analysed from a pile where the thick phase of the digestate is composted (***Compost, C***);
- b) using liquid fraction of digested produced in the same plant of case a). For this experiment, a volume of 45 L of digestate is put in a plastic basin for performing analysis (***Liquid Fraction of Digestate, LFD***).

- c) in a biological treatment plant located in Vic, Catalunya, Spain, where pig slurry were biological treats for removing nitrogen. In that case, NH_3, CO_2, CH_4 and N_2O are measured from a stored pile of solid fraction after Solid/Liquid separation (***Solid fraction of pig slurry, SFPS***).

6.2.2. Equipment used

Three different hoods have been used in the experiments. Two of them were dynamic hoods and the third was a static hood. The dimensions of each equipment are reported in the Table 44.

Table 44 Characteristics of different hoods and static chamber used in the tests.

	L1 length (m)	*L2 width (m)*	*D Diameter connection (m)*	*H height (m)*	*Emitting surface (m^2)*	*Volume (m^3)*
DH1	0,96	0,96	0,1 m	0,15	0,92	0,14
DH2	1,03	0,53	-	0,25	0,55	0,14
SH	0,40	0,41	-	0,75	0,16	0,12

The first dynamic hood (**DH1**) is a **Lindvall Hood** (Figure 55).
The use of DH1 foreseen the application of float system when it was used in a real tank for avoiding the drawing of the instrumentation. The air sampling was done through a fitting pipe inserted in the middle of the outlet hole of the DH1.

The flow velocity required inside the dynamic hood should be between 0,2 - 0,3 m/s (EPA 2001). To obtain wanted air speed inside the hood, a blower was connected to the inlet port of the Lindvall Hood with nalophane pipe. The air speed velocity was evaluated by means of thermal anemometer flow rate device at the outlet port of the hood. In order to define the air flow that must be maintained at the outlet of the hood for ensuring 0,2 – 0,3 m/s inside the DH1, a mass balance was applied (equation 15, and equation 16).

$$S_1 * v_1 = S_2 * v_2$$ Equation 15

where:

S1= hood's surface, S1=L1*L2 [m^2];
v1= air speed inside the hood [m/s];
S2= outlet surface, S2= Π(D/2)^2 [m^2];

D= outlet diameter [m];

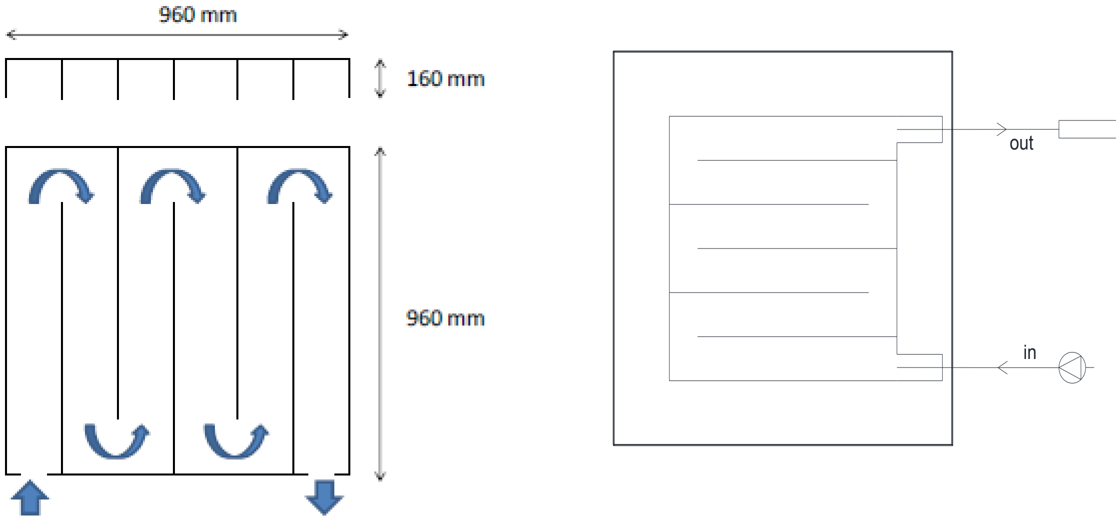
In this way v2 can be calculated as:

$$v_2 = 0{,}25 * {}^{S_1}/_{S_2}$$ Equation 16

Therefore, the measured velocity at the outlet of the hood must be around 0,8 m/s.

To estimate the emission rate for DH1, it is also necessary to define the hood factor w, which can be calculated according to equation 17 and hood dimensions.

$$w = \text{flow path section } (m^2)/\text{covered area } (m^2)$$ Equation 17



a) b)

Figure 55 Scheme of Lindvall Hood dimensions (a) and its scheme (b)- IRTA documents.

In DH1 hood, gas samples were collected via a cross-sectional sampling tube at the outlet point of the hood.

The second **dynamic hood (DH2)** (*Odournet ventilating hood*), which dimensions are reported in Table 44, consists of two radial fans that ensuring with a constant volumetric flow. In that case, the air velocity cannot be checked inside the hood and the flux is set by the equipment producer (30 m^3/h, Odurnet Technical brochure), that corresponding to an airflow equal to 0,005 m^3/s. Also for DH2 application, the its use at full scale required a floating system to allow the sailing of the hood when it is positioned on a liquid surface. A pressure sealing connection on the structure allowed plugging the PTFE pipe to the hood for air catching and sampling. In DH2 hood, gas samples were collected using the special connection (DH2) foresees in this model of hood.

The **static hood** (**SH**) (Figure 56) is a plastic enclosure, with a self-regulating pressure system that allows to avoid a vacuum effect. That hood is provided with two holes, one to insert the temperature probe and other one used as air connection. When was used on solid manure, that chamber was introduced 2 cm into the heap for guaranteeing hood's sealing.

Figure 56 Two holes in the static hood (SH) used for withdrawing air sample and for measuring temperature inside the hood.

6.2.3. Emissions sampling and calculation

Sampling procedure

The measured emissions were NH_3 and GHGs (N_2O, CH_4 and CO_2).

For the quantification of ammonia-N emitted for both dynamic hoods and static chamber, a wet direct method was applied.

Ammonia nitrogen (NH_3-N) content in the sample were trapped bubbling with a low flow rate pump (Universal PCXR8, 1000 ml/min, time accuracy ± 0,05%, SKT, Pennsylvania, USA) a known volume (5 L and 1 L for dynamic hoods and static hood respectively) into two serial glass impingers containing 10 mL of sulphuric acid 0,1 N each.

Each measure was repeated in triplicate.

Air flushing time was equal to 5 minutes for dynamic hood, as defined by preliminary tests.

Concerning static hood SH, sampling volume is equal to 1 L (< of 1% of the volume exchange rate inside the chamber) and so, sampling time is equal to 1 minute.

Nitrous oxide, CO_2 and CH_4 sampling were made with a syringe (200 mL) taking a gas sample (10 mL) from the pipe connected with the hood outlet; then was inject into an vacutainer tube with butyl rubber stopper.

For static chamber, during sampling period of lab-trial experiment each air sample was collected from the chamber headspace at 0, 10, 20, 30 and 40 min. The concentration of gases obtained during the experiment were used to construct the curve needed to calculate the emission factor (please also see paragraph "*Emissions rate calculation*"). Two more sampling time was added for full-scale tests (50 min and 60 min) in order to check if the concentration had reached saturation condition or not.
The concentration of NH_3 (Cg, mg/L) was calculated according to equation 18:

$$Cg = (C/V) \qquad \text{Equation 18}$$

Where:

C (mg) is ammonia concentration in air sampled;
V is the volume (L) of air sampled.

All NH_3 and GHGs measurements were done also for the inlet flow for background concentration.

Emission rate calculation

Emission rate (E) calculation for each of the hood was performed as follows.
Regarding **DH1**, the specific emission rate *E1* can be calculated according to Equation 19.

$$E1 = (Cout,j - Cin,i) \ * w * v \qquad \text{Equation 19}$$

Where:

E1 is the flux expressed in terms of $mg/m^2/s$;
Cout,j is the NH_3, N_2O, CH_4 or CO_2 concentration of air outlet from each chamber in mg/m^3;
Cin,i is the NH_3, N_2O, CH_4 or CO_2 concentration of air inlet into each chamber in mg/m^3;
v is the air speed expressed in (m/s);
w is hood factor (-).

The ammonia and GHGs fluxes E2 in the dynamic chamber **DH2** were calculated as follow.

$$E2 = {}^{Q \ * \ (Cout,j - Cin,i)}/_{A} \qquad \text{Equation 20}$$

Where:

E2 is the flux expressed in terms of $mg/m^2/s$;

Cout,j is the NH_3, N_2O, CH_4 or CO_2 concentration of air outlet from each chamber in mg/m^3;

Cin,i is the NH_3, N_2O, CH_4 or CO_2 concentration of air inlet into each chamber in mg/m^3;

Q is the airflow expressed in m^3/ s;

A is emitting surface (m^2).

Using static chamber **SH**, in order to calculate the ammonia emission and GHG emissions flux is necessary to build a saturation curve and defined the regression coefficient γ.

γ was calculated for each experiment using linear regression based on the concentration change as a function of collection time for the 5 or 7 samples (Hernandez-Ramirez Get al., 2010; Duchemin et al., 1999).

In order to define potential emission flow rate, the applied calculation was as follow:

$$\mathrm{E3} = {}^{\Delta gas}/_{\Delta t} * {}^{V}/_{A} \qquad \text{Equation 21}$$

Where:

E3 is the flux expressed in terms of mg/m^2/s;

γ= Δ gas/ Δ t is angular coefficient of the curve (mg measured gas/m^3/s);

V is volume of chamber's headspace (m^3);

A is emitting surface (m^2).

6.2.4. Analytical methods

Compost, digestate and pig slurry solid fraction using as emission source in the trials were characterized for total Kjeldahl nitrogen (TKN) (4500-Norg C), ammonium nitrogen (NH_4^+-N) (4500-NH_3 B C), pH (4500- H + B), electrical conductivity (EC), total solids (TS) (2540 B), volatile solids (VS) (2540 E), (APHA, 2014), and the chemical oxygen demand (COD) following de method described in Noguerol-Arias et al., (2012). NO_2^-, NO_3^-, SO_4^{3-}, PO_4^{3-} were analysed by ionic chromatography following Standard Method 4110. Total phosphorus (P) was analysed by colorimetric Standard Method 4500-P C.

During all experiments, liquid temperature and air temperature (inside and outside of the chamber for **SH**) were monitored.

Regarding ammonia concentration in the air samples, a colorimetric analysis method was applied (Spectroquant ® kit), analogues to phenate method, APHA 4500-F. Then, the samples were read in a spectrophotometer (Hach Lange, USA) using wavelength equal to 640 nm.

GHGs concentration was performed using a gas chromatograph coupled with a flame ionization detector and an electron capture detector. CH_4 was determined using a THERMO TRACE 2000 (Thermo Fisher Scientific, USA) gas chromatograph equipped with a flame ionization detector (FID).

N_2O and CO_2 emissions were determined with an Agilent 7820A (Agilent, USA) GC system using an electron capture detector (ECD).

6.3. Results and discussion

Table 45 summarizes the main characteristics of the 3 different types of manure used as emissions source during full scale trails. The characteristics of different slurry were typical of that kind of sludge (Torrellas et al., 2018). The solid fraction of pig slurry had the same characteristics that those ones reported in the study of Dinuccio et al., 2008 even if nitrogen content (TAN and TKN) are 25% lower. This can be due to the dilution of the sample caused by the rain fell in the days before the sampling. In fact, in the monitored plant the storage tank was not covered.

Table 45 Characteristics of analysed effluents and sludge. LFD: liquid fraction of digestate; C: compost; SFPS: solid fraction of pig slurry.

Parameter	*U.M.*	*LFD*	*C*	*SFPS*
pH	-	7,86	8,62	8,80
EC	mS/cm	20,60	4,10	2,73
ST	%	5,43	28,45	27,75
SV	%	3,68	24,91	23,96
$N\text{-}NH_4^+$	gN/L	3,05	2,89	1,96
TKN	gN/kg	4,33	8,36	3,68
NO_2-	mgN/kg	0,00	0,00	763,00
NO_3-	mgN/kg	0,00	0,00	578,00
COD	mg O_2/kg	58,55	459,85	412,20
SO_4^{2-}	mg/kg	-	313,00	-
PO_4^{3-}	mg/kg	62,00	73,00	-
Ptot	g/kg	0,43	0,85	1,15

6.3.1. Ammonia emissions from synthetic ammonia solution

Table 46 shows results concerning experiments using ammonia solution at different concentrations. Regarding physico-chemical parameters, data average is referred to different measurements carried out during the same experiment for each hood. All the samples have been analysed at least in duplicate.

The ammonia emissions obtained by using the water and ammonia solution as an emission source grew with the increase of ammonia concentration (as nitrogen) in the water. Therefore, due to the proportionality of emission flow to the size of the chamber and the regression coefficient γ, it is expected that emissions will have the same behaviour. In fact, as shown in Table 46, the ammonia emission rates increase with the increase of ammonium concentration in emission source.

However, emissions data obtained using an addition of buffer compound to synthetic ammonia solution were much lower. Indeed, NH_3 emissions from the slurry surface is strongly dependent on pH (Misselbrook et al., 2016), on buffer capacity (Sommer et al., 2013) and the ionic strength that has an inverse proportionality to ammonia emission (Sommer et al., 2003).

The main conclusion was that using SH, achieved emissions were lower than one order of magnitude than those detected using dynamic hoods, in any given situation, with or without buffer. Those results are supported by the ones observed in other studies where using steady state chambers, an underestimation until 50% was observed (Norman et al., 1997; Pumpanen et al., 2004). Also comparing use of static chamber with other methods as the periodic IHF technique (Shah S.B.et al.2006, Sommer et al., 2004) or boundary layer equation (Duchemin et al., 1999) the emissions of gases measured were much lower.

The possible causes of disparity in the results obtained can be in the (1) the mode of transport of the dissolved gases, (2) air temperature and water temperature, and (3) *wind* speed (Duchemin et al., 1999). These conditions can be resulted by the higher resistance to atmospheric vertical transfer in absence or under low headspace air movement (Miola E.C.C et al., 2016). The underestimation may also derive from a linear model applied for calculate ammonia flux (and other gas flux) that cannot accurately represent fundamental physics of the diffused gas transport in static chambers (Livingstone et al., 2006).

Emissions rate obtained with the two dynamic hoods DH1 and DH2, were comparable in all executed experiments. The greatest difference between the emission rates detected by the dynamic chambers was observed with higher nitrogen concentrations in the emissive source (33%).

Table 46 Characteristics of experiments with ammonia solution and emissions value measured during different experiments by the different hoods. DH1: dynamic hood1; DH2: dynamic hood 2; SH: static chamber. n: without adding any buffer compound, y: using of buffer compound.

Buffer	N. of repetition DH1	N. of repetition DH2	N. of repetition SH	Environmental Temperature °C	Slurry Temperature °C	TAN $gN\text{-}NH_4^+/L$	pH solution	Emission DH1 $mg/m^2/s$	Emission DH2 $mg/m^2/s$	Emission SH $mg/m^2/s$
n	3	3	3	10,96 ± 1,99	11,33 ± 1,39	2,26 ± 0,3	11,32 ± 0,08	1,85± 0,12	2,14± 0,18	0,35
n	3	3	1	12,37 ± 0,2	10,52 ± 0,08	1,88 ± 0,14	10,72 ± 0,12	1,6± 0,02	1,1± 0,02	0,1
n	-	-	3	13,02 ± 3,06	10,8 ± 1,43	1,26 ± 0,27	11,67 ± 0,31	-	-	0,12
n	-	3	-	13,25 ± 0,07	13,3 ± 0,14	1,1 ± 0,01	10,92 ± 0,04	-	1,2 ± 0,06	-
n	3	3	1	13,72 ± 0,37	11,72 ± 0,47	0,77 ± 0,03	10,33 ± 0,08	0,5± 0,03	0,5± 0,01	0,1
n	3	3	1	14,17 ± 0,10	12,05 ± 0,14	0,29± 0,06	11,1 ± 0,06	0,3± 0,03	0,3± 0,04	0
n	-	3	-	14,2 ± 1,13	13,5 ± 0,42	0,5 ± 0,01	10,56 ± 0,03	-	0,5 ± 0,004	-
n	-	3	-	17,35 ± 0,92	13,5 ± 0,28	0,31 ± 0,01	10,38 ± 0,05	-	0,3± 0,03	-
y	3	3	-	9,03 ± 0,76	13,77 ± 0,83	2,36 ± 0,05	8,3 ± 0,13	0,35± 0,07	0,37± 0,09	-
y	-	-	1	18,94 ± 3	15,96 ± 2,05	1,76	8,7	-	-	0,01
y	-	-	3	13,38 ± 1,49	12,72 ± 0,94	1,44 ± 0,04	8,53 ± 0,06	-	-	0

To compare the three hoods, a linear regression of ammonia emission rate in front of concentration of the emissions source was done (Figure 57). As can be seen in Figure 57, R^2 of regression is greater than 0,8 for all hoods. The regression lines for DH1 and DH2 were similar but seems that the obtains value is more different when ammonia source's concentration is higher. That situation could be due to the analytical determination and not to substantial differences between the use of two hoods.

In this case, the difference between the emission rate of the static chamber and the dynamic hoods increased with the raise of the ammonia concentration in the emission source.

At low concentration, the results of measures using two dynamic hoods DH1, DH2 and the static chamber SH are less dissimilar. From these results, it may be concluded that at low concentration of emission source, all the chambers are suitable to estimate emissions rates.

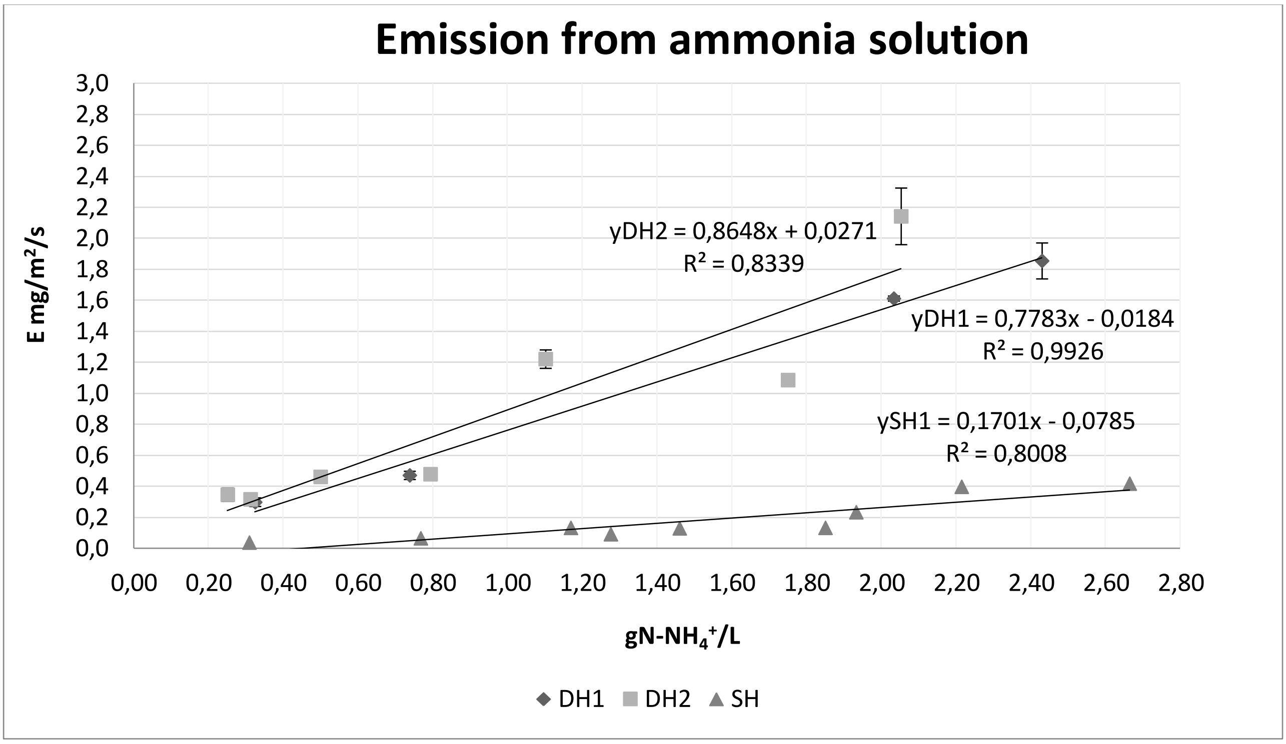


Figure 57 Linear regression of results obtained for each hood (bars error represents standard deviation).

6.3.2. Ammonia and GHGs emissions from livestock manure slurries

Ammonia emissions

Table 47 shows results of ammonia emissions measured from stored slurries as emissions sources.

Similarly to the emissions measured from ammonia solution in controlled conditions, emissions rates calculated with dynamic hoods DH1 and DH2, gave similar emission rates. In addition, ammonia emission rate resulting from livestock slurries and agro-industrial waste measurements with different hoods and chamber were similar to those obtain using the ammonia solution with buffer (Table 47 and Table 48). Anyway, similar behaviour for the hoods were observed for both emissions resulted from ammonia solution at different concentrations and from different slurries and waste.

Instead, at real scale, considering the amount of emission rates observed, with a given concentration of ammonia in the emission source (both in the ammonia solution and slurries and waste), the emission rate was different compared with those observed in the experiment executed in a control condition. It is reasonable to consider that, even if the ammonium content in the livestock slurry or agro-industrial waste exceeded 2 g/L, the emissive flows of ammonia is lower than those observed for ammonia solution for which the volatilization of ammonia was higher. The explanation is connected also with the slurry buffer capacity for the reasons explains in Paragraphs 6.1 and 6.3.1.

The ammonia emissions resulted from digestate storage obtained using dynamic hoods were lower than those one observed in the study of Sommer et al., 2000 (up to 30 $gNH_3/m^2/d$), even if in that study the technique used was micrometeorological mass balance methods. Contrary, always considering digestate, the results obtained with DH1 and DH2 were similar to those observed in the study of Perazzolo et al., 2015 (cattle digestate liquid fraction 300 $mg/Nm^2/h$ at the beginning of storage period).

Emissions from solid fraction of pig slurry in this research were more than twice times higher than those observed in the study Dinuccio et al., 2008 (60-180 $mg/m^2/h$ (min-max) @5°C and 20-280 $mg/m^2/h$ (min-max) @ 25°C). These differences can be attributable to the composition of slurry (as the content of nitrogen, 4,05 g/Kg as TAN and 12,2 g/Kg as TKN, and pH- 8,82).

The highest ammonia emissions were detected for compost, and the lowest for liquid fraction of digestate (C > SFPS > LFD).

Besides ammonia content, ammonia emissions resulted were influenced by the temperature of considered slurry/waste that has a direct influence on emission; moreover, as attested by the study of Pagans et al., (2006), which regards the determination of ammonia emissions coming from the composting process of three typical wastes, the total amount of ammonia emitted was directly related to the C/N ratio of the waste. In addition, there is a strong influence of pH on ammonia emission (Perazzolo et al., 2015) and also in this research, the trend of flux measured attested that the lower NH_3 emission correspond with the lowest pH. As well, the texture of solid fraction and compost facilitate ammonia volatilization allowing the passage of air through porous.

<u>Greenhouse gases emissions</u>

The measures of GHGs emission were performed only for slurries and for agro-industrial waste.

The use of hoods for defining of GHGs emission gave different outcomes than those observed for ammonia emission.

Relating to **N_2O**, in all tests the emission flux observed was very low and the use of static chamber, similar to what observed for ammonia emission, gave results two orders of magnitude lower. As said before, the emission of N_2O is related to incomplete ammonium oxidation or to incomplete denitrification process. So, in the considered slurries, neither of the two process was involved.

In the study of Petersen et al., (2013) a dynamic method was used for measuring ammonia and GHGs emissions. They found that stored of pig slurry resulted in total GHG emissions of up to 1,3 kg CO_2 eq/m^2/d during summer. No **N_2O** emissions were observed from pig slurry

during storage in any period of observations except in the presence of a crust, during which N_2O emissions was c. 80 mg/m^2/h.

In this study the maximum emission value detected for liquid fraction of digestate was 11,6 mgN_2O/m^2/h, measured with DH2. Nevertheless, this value is comparable with the ones obtained using DH1. Also in the study of Perazzolo et al., (2015) no N_2O emissions were observed before 32 days of storage period. In literature are reported cases of N_20 emission also from stored slurries, but in that cases the emissions were imputable to a crust presence (Sommer et al., 2000). In the study of Fukumoto et al., (2003), that analyzed emission produced from composting of pig slurry, the N_2O emissions started only when the NH_3 emissions and temperature of the compost material began to decline.

Despite CO_2 is a greenhouse gas, in the context of manure management, its emissions are considered as biogenetic and are not computed as GHGs. In that study, CO_2 emissions measured from compost and liquid fraction of digestate using different dynamic hoods gave comparable results; instead, also in this case, the use of SH1 measured emission one order of magnitude lower. Considering the solid fraction of pig slurry, CO_2 emissions detected with different hoods were similar despite the different type of the hood considered.

As reported in the study of Pumpanen et al., (2004), the use of static chamber can be appropriate for detecting CO_2 flux until 0,015 mg CO_2/m^2/s: so, as also observed when static hood is applied to synthetic ammonia solution, this technique can be applied when low emissions have to be detected.

For all chambers, **CH_4** emissions were comparable for all dynamic hoods and static chamber (same order of magnitude). As observed in the study of Perazzolo et al., (2015) the C lost via CH_4 was lower than the lost via CO_2. The smallest emission was observed using DH2 for solid fraction of pig slurry, the highest one was detected by DH1 for compost. Digestate (liquid fraction) produced lower CH_4 emission fluxes than compost.

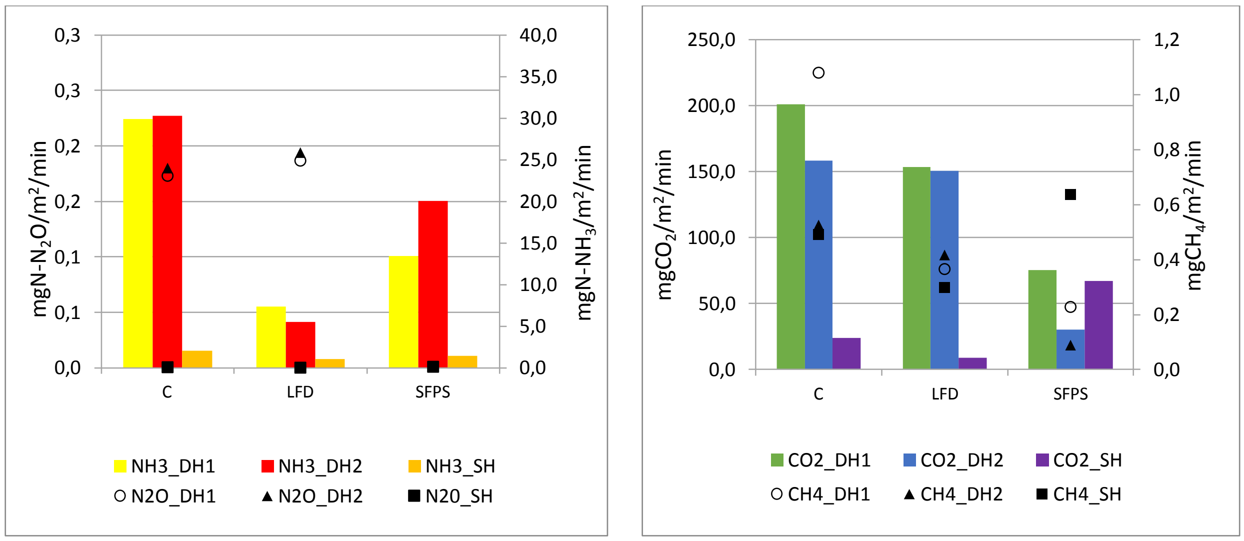
Table 47 Slurry characteristics during experiments and emissions obtained for different hoods DH1: dynamic hood1;DH2: dynamic hood2; SH: static chamber. * means that that tests were carried out with real waste but from an artificial basin and not in the real treatment plant.

Emission Source	N. of repetition DH1	N. of repetition DH2	N. of repetition SH	Environmental Temperature °C	Slurry Temperature °C	Ammonia Slurry $gN\text{-}NH_4^+/L$	pH solution	Emission DH1 $mg/m^2/s$	Emission DH2 $mg/m^2/s$	Emission SH1 $mg/m^2/s$
Stored Compost	3	3	2	16,04 ± 2,27	25,56 ± 3,32	3,05	8,62	0,5±0,05	0,51±0,23	0,03
Liquid Fraction of Digestate*	3	3	2	17,21 ± 2,93	14,39 ± 3,06	2,95 ± 0,04	7,86	0,12±0,02	0,09±0,02	0,02
Solid fraction of pig slurries	3	3	2	6,43 ± 1,59	13,70	1,96	8,8	0,22±0,03	0,33±0,03	0,02

Table 48 Emission flux of GHGs detected for slurries and agro-industrial waste. DH1: dynamic hood1; DH2: dynamic hood2; SH: static chamber. * means that that tests were carried out with real waste but from an artificial basin and not in the real treatment plant.

Emission source	E $mgN\text{-}N_2O/m^2/min$			E $mgCO_2/m^2/min$			E $mgCH_4/m^2/min$		
	DH1	*DH2*	*SH*	*DH1*	*DH2*	*SH*	*DH1*	*DH2*	*SH*
Compost	0,2	0,2	0,0	201,2	158,7	24,1	1,1	0,5	0,5
Liquid Fraction of Digestate*	0,2	0,2	0,0	152,8	150,3	9,2	0,4	0,4	0,3
Solid fraction of pig slurries	0,0	0,0	0,0	75,4	30,3	67,4	0,2	0,1	0,6

As explained in Paragraph 6.2.1, the digestate collected at the plant was then transferred into heavy-duty PVC and polyester square containers at experimental site, where the emission analyses were carried out. Certainly, if the analyses were carried out on "fresh" separated slurry, the emissions obtained would have been higher. In fact, the high emission of CH_4 are observed at the beginning of storage and decreasing over time. In Figure 58 is shown the emission of nitrogen compounds and carbon compounds from different type of source.



a) b)

Figure 58 NH_3, N_2O (a) and CO_2, CH_4 (b) emission rates for all considered slurries. C: compost, LFD: liquid fraction of digestate, SFPS: solid fraction of pig slurry.

The comparison of NH_3 and GHG emissions from stored slurries and agro-industrial waste was very difficult not only for the differences between the characteristics of the effluents analysed, but also for the unique features of environmental conditions and site-specific characteristics. Actually, the use of dynamic hoods imposed a constant value of “*wind speed*” far from reality condition; as well, this method does not consider some contribution as rainfall, creating, as defined by Miola E.C.C et al., (2015), an “oasis effect”. Also, for these reasons, evaluation of full-scale ammonia or GHG emissions can be realized through other approaches, as IPCC approaches, that can be consider more appropriate for its completeness in considering a general environmental aspect, bigger scale and that is not site-specific. In spite of this, the results obtained with direct method technique were closer to those reported in literature, obtained with other indirect methods. In addition, the emissions measured applying direct methods allow having emission factors closer to punctual reality. For this reason, this data, could be used as a “correction factor” for the output of general model. For example, equation used for evaluating emission that are applied in indirect methods can be incorporated with a punctual emission obtained using direct emission measurements, in order to provide a less generalize output.

6.4. Conclusions

In this study a comparison of ammonia and GHG emissions measurements were carried out using two different technologies both applying direct methods: dynamic hoods and static chamber.

The use of two dynamic hoods, despite the different size, operation, applied air flux, present comparable emissions rate results when used for estimating emissions from ammonia solution and slurry; the same consideration can be done also for GHG emissions. The obtained results shown how the use of dynamic hoods and static chamber can be consider valuable techniques for comparing ammonia emission rate at full scale.

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SECTION 4 - PHYSICO-CHEMICAL TREATMENT FOR PHOSPHORUS RECOVERING AND REMOVAL FROM LIVESTOCK MANURE

The content of this chapter will be used for preparing an article that will be submitted in "Science of Total Environment".

7. Effect of additives on phosphorus, copper and zinc separation from raw and treated animal slurries

Animal excreta is not just waste material that need to be through away but represents raw material that can be used for plant production essential to feed and nourish a growing world population (Sommer et al., 2003).

Currently, there is an increasing interest in finding the most suitable treatments necessary not only to reduce the environmental problems related to the management of animal manure, but also to allow the recovery of those elements of which the manure is rich.
The phosphorus separation using chemicals represents an example of this kind of approach.
This treatment can be applied stand-alone for removing phosphorus (for example in a wastewater treatment plant where a chemical phosphorus precipitation is often performed for respecting the law threshold limits) or can be applied as a pre- or post-treatment for removing not only P but also a suspended solid.

In this chapter, a possible application of phosphorus separation using chemicals is analysed. In particular, a possible use of calcium hydroxide and aluminium salt were applied for removing and recovering phosphorus compound from different slurries. In addition, the possible coupling of the separation treatment with a stripping process was considered.

7.1. Introduction

As much as 40 million tons of phosphorus (P) as P_2O_5 is consumed by human population each year; this has impact on P demand that will increase by 1,5% annually (Harris et al. 2008). The artificial phosphorus fertilizers production from phosphate rock is also energy intensive and lead to significant emissions of carbon, radioactive by-products and heavy metals (Cordell et al., 2009; Tervahauta et al. 2014). Furthermore, the rock phosphate is not a renewable resource: it is predicted that could be exhausted in 100–250 years (Grigatti et al. 2015; Tervahauta et al. 2014). It is therefore necessary to limit unnecessary use and find alternative to mining phosphate rock improving the recycling and recovery of P from wastes and other waste products. As an alternative, P can be obtained for example from civil wastewater, livestock wastewater, livestock slurry (Tervahauta et al. 2014).

Recovered P in an easily dewatered form could be stored and applied as a slow-release fertilizer in accordance with crop needs (Taddeo et al. 2016).

Reducing nutrient emissions from agro-industrial sources and the need to recycle nitrogen (N) and phosphorus (P) are important for improving agro-industrial management and environmental protection. In fact, surplus nutrients (N and P) can be transported from watersheds by surface runoff or base flow of contaminated groundwater and cause water eutrophication (Taddeo, Kolppo, and Lepistö 2016). Because of the imbalance in the amounts of N and P in animal manure relative to crop needs, P is often applied in excess of crop requirements, increasing the P concentration in soils and consequently the potential for P movement to water resources. Thus, removing P from manure can reduce environmental pollution and represent a source of renewable P to be used as fertilizer in agriculture.

Besides the need to recover P from livestock manure to avoid its over application especially in intensive livestock areas, some concerns in correct disposal of livestock manure are also related to heavy metals, especially copper (Cu) and zinc (Zn).

Heavy metals are present in low-concentration in livestock diets and can be added to certain foods for health and wellbeing or as growth promoters (Nicholson et al. 2003; Sommer et al. 2013). Nevertheless, only a small percentage of ingested heavy metals are absorbed by animals, the remaining is excreted in the faces or urine, as the 15-40% of the phosphorus (Suzuki et al., 2010). In this way the excess of elements not used by the plants can cause soil and water pollution. Approximately 40% of total annual input of Zn and Cu to agricultural land is derived from livestock manures (Marcato et al., 2003; Suzuki et al., 2010) and the continuous application of manure can lead to an increase of the concentration of Cu and Zn in soils and in the food chain (Provolo et al. 2018).

One possibility for recovering P and removing heavy metals from animal manure is solid-liquid separation. This physical treatment technique produces a solid fraction having a high dry matter (DM) content that is rich in nutrients, as well as a dilute liquid fraction; the fractions can be managed separately. The solid fraction, which has a relatively small volume, contains high concentrations of carbon (C) and nutrients, particularly N and P. The liquid fraction has a relatively lower nutrient content, but can be easily transported and spread to fields (Burton 2007).

Physical and mechanical separation technology is used also for P removal from animal manure and digestate and represents a critical step in the sustainable and correct use of nutrient. Even if solid/liquid separation can be considered a suitable method to remove these elements (Møller et al., 2007), higher removal efficiencies can be reached only with the use of chemical additives, flocculants and/or coagulants (Fernandes et al., 2011; Vanotti et al. 2017).

Among the possible separation treatments, there are some, as coagulation process, that removes not only phosphorus in the form of phosphate, but also dissolved organic matter, colloids and particulates which can be coagulated (Duan and Gregory 2003).

Some successful studies at laboratory scale attempted to recycle P in the form of struvite [$MgNH_4(PO_4)\cdot 6H_2O$] from the liquid phase of manure (Cerrillo et al. 2015). However, struvite formation requires a special chemical condition that can be difficult to achieve (Fernandes et al. 2011). P recovery also can be achieved by thermo-chemical processes, such as incineration, sintering or smelting sewage sludge or animal wastes (Cabeza et al. 2011).

In hydroxide precipitation process, the addition of coagulants such as alum, iron salts, and organic polymers can enhance the removal of heavy metals from wastewater (Fu and Wang 2011). Concerning chemical precipitation, several types of additives have been studied to coagulate manure, to flocculate it or to remove orthophosphate (PO_4^{3-}). Experimental and actual applications have used iron (Fe), aluminium (Al) and calcium (Ca) salts, several types of polyacrylamides and clay as coagulants/flocculants (Christensen, Hjorth, & Keiding, 2009; Hjorth & Christensen, 2008; Møller et al. 2007; Vanotti et al. 2002).

Aluminium sulfate ($Al_2(SO_4)_3$) is one of the most use chemical compound for coagulation process. According to the $Al_2(SO_4)_3$ stoichiometric reaction, 0,87 g of chemical is required for 1 g of P present in a mixture. However, many side reactions occur and increase the required amount of reagents compared to the stoichiometric values. In fact, Al^{3+} as Fe^{3+} dose in liquid manure can reach 2 to 3 for the molar ration of metal to P in order to achieve over 95% removal (Lin et al., 2013).

The best pH condition for P-removal using aluminium is 5,5-6,5; this condition can achieve by exploiting the acid hydrolysis of $Al_2(SO_4)_3$. Concerning the use of aluminium salt, in the study of Szabo et al., 2008 it is reported that for a concentration ranges between 0,5 to 6 mg/L of soluble phosphorus (PO_4-P) is necessary doses above 1,5 to 2,0 Me_{dose}/P_{ini} (where Me/P is molar ratio between the metal and soluble phosphorus) for reaching 80-98% of removal rate. The application of Ca compounds for P removal may initially precipitate a number of Ca phosphate minerals (brushite, monetite, octacalcium phosphate, hydroxyl apatite, or amorphous calcium phosphate) (Szogi, et al., 2015). By increasing pH with $Ca(OH)_2$ soluble P can be precipitated as amorphous calcium phosphate (ACP, $Ca_3(PO_4)_2{\cdot}xH_2O$) but also calcite ($CaCO_3$). In addition, when calcium hydroxide is added to liquid manure, the hydroxide reacts with the existing bicarbonate to form carbonate, with NH_4^+ to form NH_3, and with phosphate to form phosphate precipitate compounds (Szogi et al., 2015). Also for these reasons the dosage of Ca compounds be define independently from the P-content.

Concerning use of Ca components, results presented in the study of Harris et al., (2008) showed an interactive effect of CO_3^{-2} and Mg^{2+} on Ca phosphate precipitation when they co-exist; i.e., addition of $MgSO_4$ can suppress carbonate precipitation and enhance $Ca_3(PO_4)_2$ precipitation at elevated pH (>9). Vanotti et al., (2003), attested that P removal efficiency for a lagoon swine wastewater after nitrification pre-treatment reached 98% with a molar ration Ca:P of 2.5.

While there are several experiences reporting the P removal efficiencies from livestock manure using mechanical separation with or without additives, the effect of these treatments on Cu and Zn removal is not adequately addressed, especially with regard to digestate from anaerobically digested animal manure. Moreover, there is little information about P, Cu and Zn removal efficiencies from animal slurries pre-treated using a N removal process based on ammonia stripping process.

The objective of this study was to evaluate how different doses of $Al_2(SO_4)_3$ and $Ca(OH)_2$ added to a solid-liquid separation treatment can affect P, Cu and Zn separation efficiencies from raw pig slurry and co-digested slurry before and after ammonia stripping treatment. The overall goals of the research are (1) the evaluation of the separation efficiency for possible phosphorus-recovery and for the removal of heavy metals and (2) if that treatment can be coupled with a novel stripping process aimed to N-recovery. In particular, we assessed how the type of slurry, the used pre-treatment (stripping process and physico-chemical treatment) and the dose of additives influence the separation efficiencies of these elements.

7.2. Materials and methods

7.2.1. Slurries and ammonia stripping

Laboratory scale solid-liquid separation experiments were conducted on manure slurries using two additives $Ca(OH)_2$, hereafter shortened to CaH, and $Al_2(SO_4)_3$, hereafter shortened to AlS, to study the effect of different doses of the additives on removals of total solids (TS), P, Cu and Zn.

Three types of animal slurries were used, two of which were tested both before and after being subjected to an ammonia stripping process. The three types of slurries were:

- liquid fraction of pig slurry extracted from raw slurry using a screw press mechanical separator (designated “pig slurry, PS”), collected from a fattening pig farm;
- liquid fraction (from a screw press separator) of anaerobically co-digested pig slurry and corn silage before being subjected to a stripping process for ammonia removal (“pig digestate, PD”) and after stripping (“pig digestate stripped, PDS”); and
- liquid fraction (from a screw press separator) of anaerobically digested slurry from 25000 pigs and 1800 dairy cattle before being subjected to a stripping process for ammonia removal (“cattle and pig digestate, (C+P)D”) and after (“cattle and pig digestate stripped, (C+P)DS”).

All slurries were collected from farms located in the province of Bergamo (Italy).

The laboratory scale stripping process was carried out in 50 L reactors, each having a hydraulic residence time of 12 days. During the process the reactor temperature was maintained at 40°C and slurries were mixed at 50 rpm. The airflow was set at 20 L/min. In the first 2 days, 33% of the airflow was provided from the bottom of the reactor through air stones and the rest was introduced into the headspace. Subsequently, 100% of the aeration was introduced into the headspace. The stripping process achieved an N removal efficiency of approximately 80% (Provolo et al. 2017).

7.2.2. Determination of optimal chemical doses

In order to define the correct dose of CaH and for AlS for precipitation and separation treatment, preliminary tests were executed. An appropriate dose (D) of two chemicals were individuated with studies of pH and EC patterns with the addition of increasing doses of each additive in a 500 mL of each slurry analysed.

As noted in paragraph 7.1, there is no direct relation between P concentration in animal manure slurries and the CaH dose necessary for its precipitation. In addition, P precipitation using CaH occurs only after the precipitation of carbonate. With the addition of CaH the carbonate ions precipitate massively as confirmed by the decreasing of conductivity. Then, the conductivity decreases until the carbonate concentration becomes limiting and prevents the solubility limit of this salt from being exceed. The finest dose of CaH for carbonate precipitation corresponds to a maximum reduction of conductivity. Above this dose all inorganic carbon being consumed and the excess of the optimum dose introduced in the solution results in the increase in Ca^{2+} content and thus in conductivity (Renou et al., 2008). For this reason, it was decided that the reference dose D is the one to which corresponds EC minimum value.

Regarding AlS, in addition to observing pH and EC, evaluation of produced foam level and the flock dimension were carried out. For this last evaluation, constant amounts of chemical were added in 500 ml of slurries. After the injection of the additive, a mixing at 200 rpm for 20 second than 30 rpm per 2 minutes occurred. Then flock dimension was evaluated (scale from 1: liquid to 5: flakes of ~ 5 mm) and the foam produced measured. The selected dose of AlS corresponded to the Al^{3+} concentration for which the flock dimension did not change, and the foam level remained relatively constant.

7.2.3. Solid liquid separation test

Jar-test with six-place gang stirrer was used for experiments. For each slurry, separation was carried out using both selected additives at 3 different doses: (1) null dosage, (2) the optimal dose determined with preliminary test for each slurry and (3) 25% lower dosage than optimal.

CaH was introduced in the sample as “lime milk” at concentration of 200 g/L.

Solution 1 M of AlS was prepared for each experiment and dosed at desired concentration using a micropipette.

In each test 500 mL of slurries was transferred in a 1 L glass beaker where each dose was added simultaneously before starting the mixing phase that occurred in two steps. The first rapid mixing step (5 minutes at 250 rpm and 30 sec at 250 rpm for CaH and AlS respectively) followed by a slow mixing phase at 30 rpm for 20 minutes for CaH and 20 rpm for 20 min for AlS. Then, suspensions were allowed to settle for 30 minutes (Renou et al. 2008; Szabó et al. 2008). At the end of settling phase, each solution was separated using a static filter with a 0,25-mm mesh and allowing 30 minutes of drainage time. The two different fractions obtained, filtrated and retained solids, were weighed. All tests were conducted in duplicate.

7.2.4. Analytical methods

Each slurry was characterized for Total Kjeldahl Nitrogen (TKN), electrical conductivity (EC), total ammonia nitrogen (TAN), pH, dry matter content (TS) and volatile solid (VS), which were determined using Standard procedures (APHA/AWWA/WEF 2012). The total contents of P, Cu and Zn in the slurries were evaluated using inductively coupled plasma mass spectrometry after acid mineralization (EPA 1998; APHA 2012).

All analyses were conducted in duplicate.

7.2.5. Calculation and statistical analyses

To evaluate the separation results, two indexes have been calculated (Sommer et al. 2013).

The separation efficiency (Et(x)) that expresses the distribution of a given compound (dry matter, nitrogen, phosphorus, etc.) between the solid (fraction retained by the filters) and liquid fractions (clarified fraction) was calculated as the ratio between the mass of a specific compound in the solid fraction and the mass of the same compound in the input slurry, as reported in the equation 22 (Cocolo et al. 2016).

$$E_t(x) = \frac{m(x)_{solid}}{m(x)_{solid} + m(x)_{liquid}} \qquad \text{Equation 22}$$

The simple separation index gives no indication of an increase in concentration of compound x in the solid fraction; therefore, the reduced separation index, E't(x), also was calculated (Cocolo et al. 2016):

$$E'_t(x) = \frac{E_t(x) - \frac{m_{solid}}{m_{slurry}}}{1 - \frac{m_{solid}}{m_{slurry}}} \qquad \text{Equation 23}$$

where m_{solid} and m_{slurry} are the total masses (g) of solid fraction and separated slurry, respectively.

Equation 23 has all requirements for an efficiency definition: assumes value 0 when no separation takes place and 1 when separation is complete (Moller et al., 2000). In particular, $E'_t(x)$ ranges from -1 to +1: positive values indicate an increase in concentration of x in the solid fraction compared with the raw slurry and negative values indicate an increase in concentration of x in the liquid fraction.

Statistical analyses were completed using SPSS v. 25 software (IBM Corp., Armonk, NY, USA). A descriptive statistical analysis was performed on all the data, and the results were expressed as means and standard deviations. The data were also subjected to analysis of variance (ANOVA) using a full factorial model (fixed factors: dose, additive and slurry type). The homogeneity of the variances has been checked with the Levene's test. The significant treatments were compared by utilizing Tukey's HSD multiple range test ($p < 0.05$).

7.3. Results and discussion

7.3.1. Slurry characteristics

The physico-chemical characteristics of the five slurries used are summarized in Table 49.

The P concentrations in raw pig slurry and digestates were comparable to those reported by Finzi et al., (2015) and Møller et al. (2007); the Cu and Zn contents are similar to those reported in the study of Nicholson et al., (2003), even if found values for Cu and Zn concentration are higher than those results from other study (Suzuki et al., 2010).

The increase in concentrations of heavy metals and other elements after the stripping process can be explained as consequence of water evaporation that occurs during the treatment (at 40°C). The same explanation applies for solids content: without considering evaporation, the stripping process increased the TS concentration by 16% and 19% for pig and cattle digestate and pig digestate, respectively.

Table 49 Main characteristics of five slurries used in the tests.

	Pig slurry	Pig digestate	Pig digestate stripped	Mix digestate	Cattle and pig digestate stripped
pH	6,75 (±0,18)	8,1(±0,08)	7,6 (±0,03)	8,2 (±0,24)	8,4 (±0,04)
EC	25,84 (±0,32)	29,7 (±1,15)	15,8(±0,13)	20,9 (±1,2)	12,0 (±0,23)
TKN (g/kg_{TQ})	3,93	4,6	2,2	2,4	1,2
$N\text{-}NH_4^+$ (g/kg_{TQ})	2,26	3,4	0,6	1,9	0,2
TS (%)	2,5 (±0,18)	4,2 (±0,44)	5,0 (±0,23)	2,5	3,1 (±0,49)
VS (as % TS)	63,0(±2,28)	58,2 (±0,89)	55,7 (±2,41)	59,5	51,1 (±8,78)
Alkalinity (mg $CaCO_3$/L)	17102,0 (±1734,1)	18542,6 (±3464,1)	9175,7 (±629,7)	18463,6 (±9494,5)	8554,8 (±2645)
P (mg/kg_{TQ})	370,6 (±43,0)	1137,4 (±354,6)	1508,9 (±30,8)	451,8 (±50,6)	612,7 (±98,7)
Cu (mg/kg_{TQ})	12,9 (±1,6)	6,8 (±2,4)	10 (±0,9)	5,1 (±0,5)	6,6 (±0,5)
Zn (mg/kg_{TQ})	20,4 (±2,8)	41,5 (±12,1)	61,7 (±3,9)	25,1 (±3,2)	31,6 (±4,8)
Al (mg/kg_{TQ})	10,6 (±1,5)	54,5 (±7,3)	71,2 (±4,2)	85,2 (±12,6)	136,0 (±18,9)
Ca (mg/kg_{TQ})	761,3 (±81,8)	2845,9 (±698,6)	3433,7 (±195,4)	1014,1 (±69,0)	1298,4 (±159,7)
Mg (mg/kg_{TQ})	344,6 (±34,8)	563,3 (±272,8)	671,5 (±11,1)	275,6 (±30,9)	422,0 (±78,0)
Fe (mg/kg_{TQ})	42,9 (±6,4)	495,2 (±123,4)	699,4 (±36,9)	91,1 (±15,4)	135,4 (±16,9)
K (mg/kg_{TQ})	2964,6 (±251,7)	2777,1 (±1069,2)	5058,0 (±425,3)	2779,3 (±178,0)	3628,2 (±321,5)
Cr (mg/kg_{TQ})	0,1 (±0,1)	1,0 (±0,2)	1,5 (±0,1)	0,3 (±0,0)	0,4 (±0,0)

Notably, VS content (expressed as percentage of TS) decreased: probably, in the digested slurry there were still biodegradable components that were degraded during the process. In addition, alkalinity decreased after ammonia stripping: in fact, that kind of process affected the content of carbon dioxide, ammonia nitrogen, which are strictly connected with alkalinity concentration. As expected, the main effect of the stripping process was a reduction of TAN concentration, and TKN concentration.

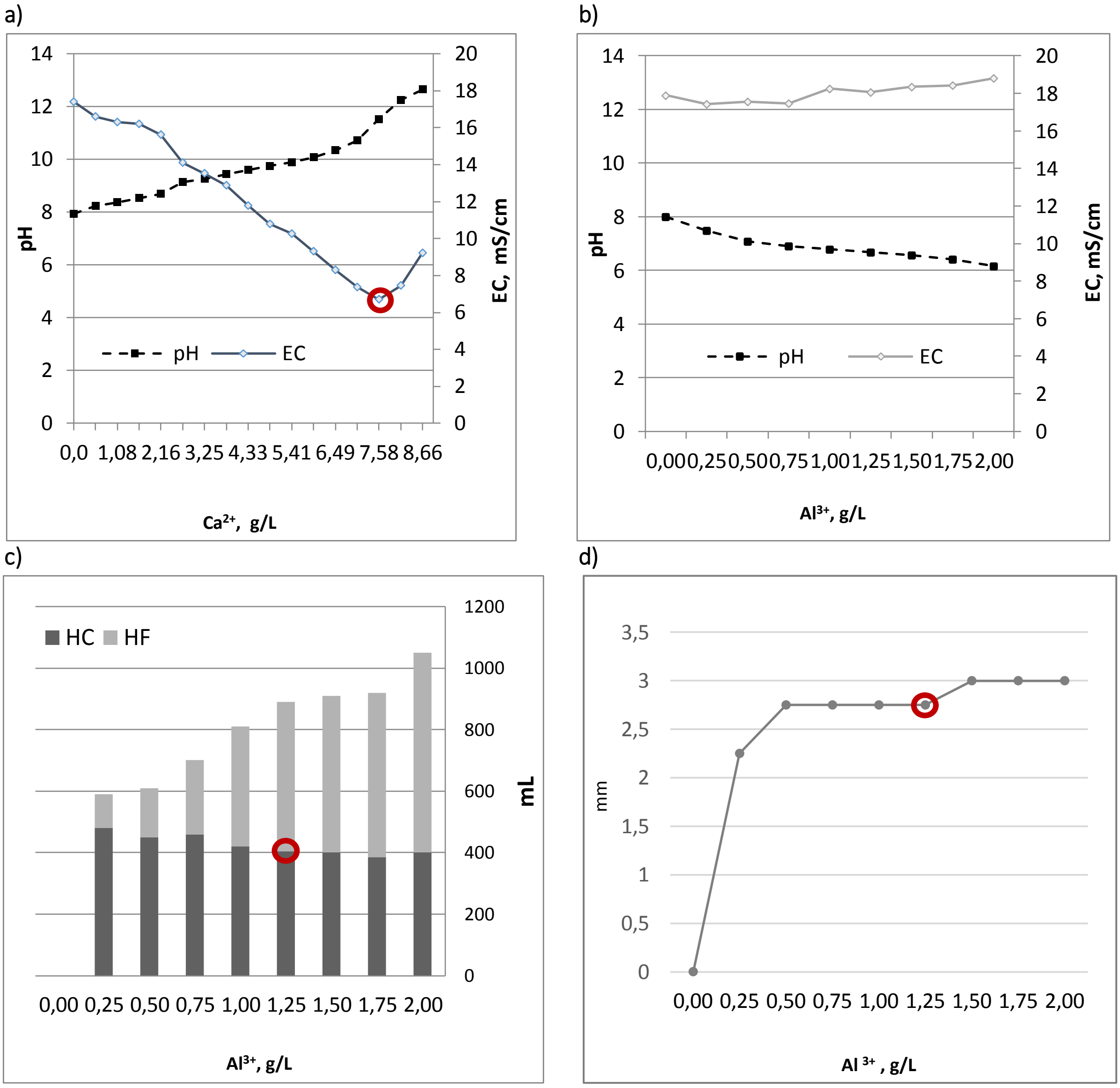
7.3.2. Determination of chemical dose

In the Figure 59 are reported some examples of results obtained during preliminary tests for determining chemical doses.

In particular, for liquid fraction of pig and cattle digested slurry, as optimal dose equals to 7,58 g/L of Ca^+ was identified, corresponding to an EC value of 7,4 mS/cm and a pH value of 11,5 (Figure 59a).

Concerning Al^{3+}, the optimum dose of 1,25 g Al^{3+}/L corresponded to pH 6,68 and, EC 18,06 mS/cm, a foam level of 400 mL and floc dimension slightly larger than 2,5 mm (Figure 59b, 59c and 59d).

Figure 59 Example of results from jar tests used to determine optimal doses (D) of $Ca(OH)_2$ (CaH) and $Al_2(SO_4)_3$. (AlS) for “cattle and pig digestate”: a) pH and EC responses to CaH; b) pH and EC responses to AlS; c) Level of solid/liquid interface (HC) and foam fraction (HF) in response to AlS; d) Flock dimension in response to AlS. The lowest value of EC (denoted by the red circle) was used to identify D for CaH. The reference dose of ALS selected corresponded to the Al^{3+} concentration beyond which the flock dimension did not change and the foam level remained relatively constant.



The optimal identified doses for each slurry are reported in Table 50.

It can be observed that for CaH the optimal dose is lower for both types of stripped digestate than for raw digestate. This was due to the decreased alkalinity concentration resulting from the stripping process, which in turn decreased the amount of CaH required to precipitate carbonate.

Table 50. Optimal dose (D) of $Ca(OH)_2$ (in terms of Ca^{2+}) and $Al_2(SO_4)_3$ (in terms of Al^{3+}) for different types of animal manure slurries. The numbers in brackets identify specific repetitions.

Slurry types	DOSE D	
	Al^{3+} g/L	Ca^{2+} g/L
PIG DIGESTED SLURRY (1)	1,5	11,91
PIG DIGESTED SLURRY (2,3)	1,5	10,82
PIG SLURRY (1,2)	1,5	6,49
MIXED DIGESTED SLURRY (1)	1,25	7,58
MIXED DIGESTED SLURRY (2,3)	1,25	8,12
PIG DIGESTED STRIPPED (1,2)	1,5	3,25
MIXED DIGESTED STRIPPED (1,2)	1,5	3,79

7.3.3. Influence of additives on physical-chemical characteristics and separation efficiency of solid, phosphorus, copper and zinc from slurries

The separation efficiencies obtained for total solids, P, Cu and Zn are reported in Figure 60. The addition of AlS generally improved the separation efficiencies for all parameters for all the slurries tested.

The separation efficiencies, however, varied consistently for different elements and different slurries.

The separation efficiencies for TS varied according to slurry type. Regarding cattle and pig digestate, observed removal rate ranged between 58-60% without application of chemicals. For this slurry, the maximum solid removal efficiency is reached with AlS (79%). After the stripping process, the TS separation efficiency for cattle and pig digestate without chemicals was very low (29–40%). These differences could be due to the physical-chemical changes in the slurries caused by the stripping treatment.

Interestingly, in relation to CaH for cattle and pig digestate (both before and after stripping), the TS separation efficiency seemed to increase as the dosage of CaH increased. However, this difference was not statistically significant (Table 51).

The separation of TS from pig digestate was quite dissimilar to that from pig and cattle digestate. In fact, using AlS highest TS separation efficiency is attested for slurry after ammonia stripping process (92%). In contrast, the opposite response was observed when using CaH, and the maximum separation efficiency (63%) was achieved at the sub-optimal dose applied to untreated (not stripped) pig digestate. Using CaH, the lowest TS separation efficiency among all slurry types occurred with pig slurry, both at null dose (< 10% TS separation) and when additives were used. The P separation efficiencies (Figure 60b) from all slurry types were higher when AlS was added than when CaH was used.

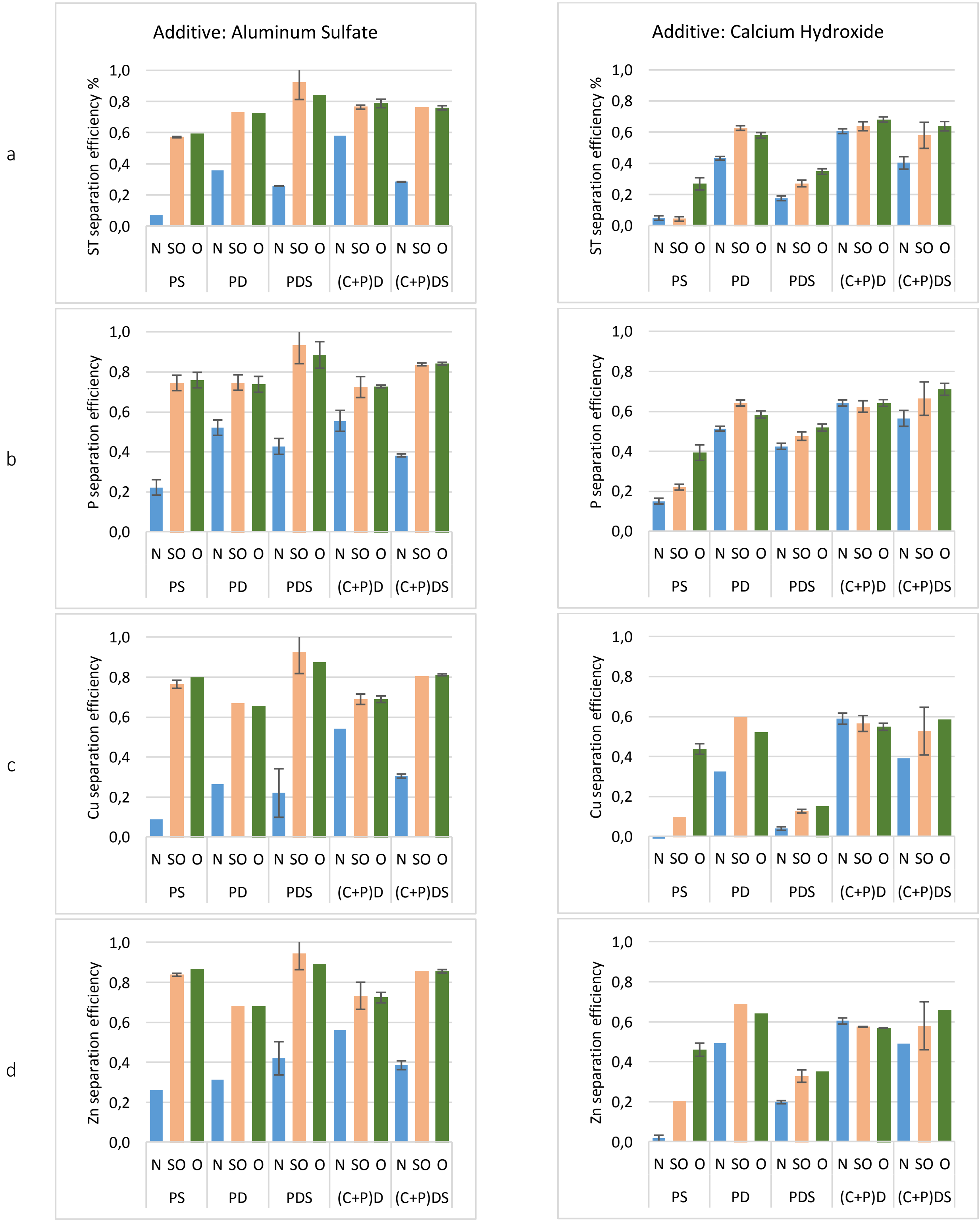


Figure 60 Separation efficiencies achieved using and $Al_2(SO_4)_3$ (left) and $Ca(OH)_2$ (right) as additives to different slurry types: a) total solids (TS); b) phosphorus (P); c) copper (Cu); d) zinc (Zn). N: null dose; SO: sub-optimal dose; O: optimal dose. PS: pig slurry; PD: pig digestate; PDS: pig digestate stripped; (C+P)D: cattle and pig digestate; (C+P)DS: cattle and pig digestate stripped.

Furthermore, both chemicals achieved higher P separation efficiencies from most slurry types (excepting pig plus cattle digestate) than were obtained without the addition of chemicals, even if the optimal dose did not always significantly improve P separation compared with the sub-optimal dose.

P-separation rate obtained for ammonia-stripped slurries (both pig and cattle digestate and pig digestate) using AlS were comparable to those obtained for the same non-treated slurry (except with the sub-optimal dose for pig digestate), despite the applied doses being lower for the stripped digestates. The maximum P-separation efficiency obtained with AlS was 93% for pig digestate stripped, with application of a sub-optimal dose(0,75D).

For two digestates (pig digested slurry and pig and cattle digested slurry) P-separation efficiency seemed to be unaffected by the use of CaH. Nevertheless, the maximum P separation efficiency (71%) was achieved from the pig plus cattle digestate at the optimal CaH dose after ammonia stripping.

For the pig digestate (before and after stripping), the maximum P separation efficiencies were 64% and 52% respectively, with the application of sub-optimal dose, and optimal dose of CaH. However, the two efficiencies were not significantly different (Table 51). This difference may also be caused by the content of TS, which are present in higher concentrations in pig digested after stripping.

The P-separation efficiency achieved from pig digestate without the use of chemicals were significantly lower (45-51%).

The P-separation efficiency in pig slurry was not particularly high: 72% using AlS and 39% using CaH; however, the results using AlS were comparable with those obtained by Ndegwa et al., (2001). In their research, the addition of aluminium sulfate to swine manure prior to the quiescent natural sedimentation were approximately 78% and 65% at dosages of 1,5 and 2,0 g/L, respectively. In contrast, the P separation efficiency obtained using CaH in present study, was lower than that observed by Szogi et al., (2009). In fact, in that research, the $Ca(OH)_2$ concentration range (0,17–0,89 g/L) used to treat swine wastewater raw or pre-treated in a nitrification process was lower than in this study; yet the doses achieved P removal efficiencies ranging from 20% to 100%.

Both AlS and CaH produced greater differences in P separation efficiencies for pig slurry than for other slurry types.

In addition, this observation is also true for Cu and Zn. For this slurry, the use of chemicals was effective: copper separation increased from 1% without chemical to 44% and 79% for CaH and AlS, respectively.

Similarly, without the application of chemicals the Cu separation efficiency for cattle and pig digestate before and after the stripping process was 54–59% and 31–39%, respectively. The Cu maximum separation efficiency was observed for stripped slurries, with the application of optimal dose AlS, but it is not significantly different from which observed sub-optimal one.

Table 51 Significance of the factors considered in the statistical analysis of separation efficiency.

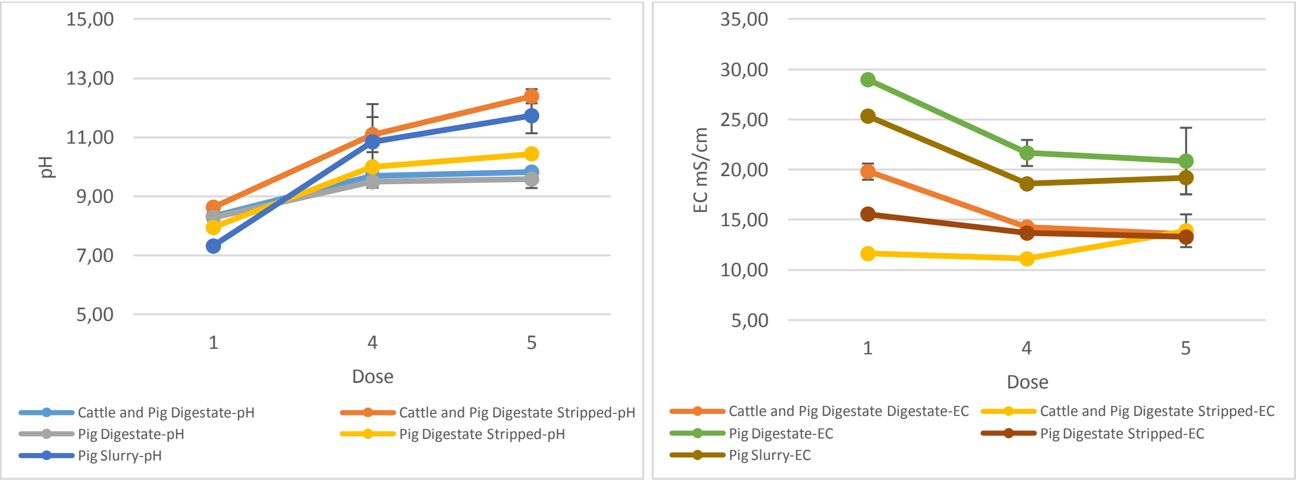
			ST	P	Cu	Zn
Slurry type	Cattle and pig digestate stripped	Mean	0,56	0,65	0,56	0,63
		Statistical significance	*b*	*a*	*ab*	*a*
	Cattle and pig digestate	Mean	0,69	0,66	0,61	0,63
		Statistical significance	*a*	*a*	*a*	*a*
	Pig digestate	Mean	0,58	0,62	0,51	0,58
		Statistical significance	*b*	*ab*	*b*	*a*
	Pig digestate stripped	Mean	0,44	0,59	0,36	0,50
		Statistical significance	*c*	*b*	*c*	*b*
	Pig slurry	Mean	0,28	0,33	0,35	0,44
		Statistical significance	*d*	*c*	*c*	*c*
Dose	null	Mean	0,31	0,39	0,25	0,36
		Statistical significance	*b*	*b*	*b*	*b*
	Sub-optimal dose	Mean	0,60	0,66	0,59	0,65
		Statistical significance	*a*	*a*	*a*	*a*
	Optimal dose	Mean	0,62	0,67	0,61	0,67
		Statistical significance	*a*	*a*	*a*	*a*
Chemical	CaH	Mean	0,42	0,48	0,35	0,44
		Statistical significance	*b*	*b*	*b*	*b*
	AlS	Mean	0,61	0,67	0,61	0,68
		Statistical significance	*a*	*a*	*a*	*a*

Using CaH, separation efficiency in cattle and pig digestate was not influenced by increasing chemical dose; contrarily for the same slurry after stripping process, separation depend on the dose applied.

Regarding achievable Cu-separation efficiency, the use of AlS led to higher separation efficiency for digested pig slurry stripped and cattle and pig digestate stripped, and, for these slurries the difference between removal rate with and without application of chemical was higher (22%-93% and 31%-81%, respectively). Using CaH, pig digested slurry treated shown the lowest separation efficiency. Notably, in cattle and pig-digestate before stripping, Cu separation efficiency decreased with the increasing of the dose.

Analysing the results obtained for Zinc separation efficiency, the general behaviour was comparable with that observed for Cu but the Zn separation efficiency obtained for all slurry types using both chemicals were higher than those observed for Cu. As reported by Marcato et al., (2009), the Zn is much more sensitive to pH than Cu, so it is possible that the chemical characteristics of different slurries influenced differently the Zn separation efficiency. It should also be noted that in all slurry types, Zn content was higher than copper (Table 49). Olatuyi et al., (2014), studied the heavy metals removal efficiencies obtainable with different separation methods and its speciation, found that the use of centrifugation (with flocculants) achieved separation efficiency of 65% and 45% for Cu and Zn respectively. Because Cu, Zn and also P are mostly present in the solid fraction, an increase in the separation efficiencies of P, Cu and Zn is expected as the TS removal increases. Moreover, as deeply stud by Szogi at al., (2009), which developed multi stage process in order to reduce inorganic buffering in slurry before the addition of CaH, similarly, the ammonia-stripping process in this study decreased NH_3, alkalinity in all slurry types, thus lowering the required chemical dosage. As shown in Figure 61, in all type of slurries, chemical additive used at increasing dose influenced the pH and EC values.

CaH



AlS

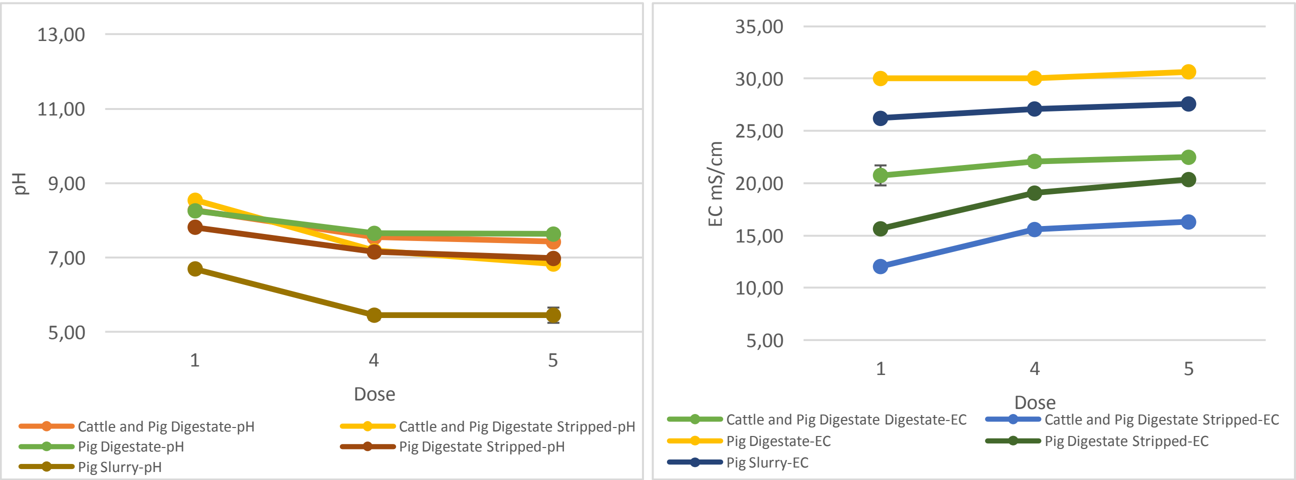


Figure 61. Variation of pH-patterns (left) and EC-patterns (right) in different slurry types with an increase dose of CaH (above) and AlS (below) for different slurries. 1 correspond to null dose 0 mg/L, 5 to optimal dose and 4 to dose Sub-optimal dose. Data reported are means and the error bars represent standard deviation of data.

In general, for all slurry types, solid-liquid separation without the use of chemicals produced a liquid fraction having a pH slightly higher than that observed for the raw slurry. With the use of AlS, the EC increased as the dosage increased and (as expected) pH decreased. At the maximum dose of Al^{3+}, pH decreased to less than 5,5. Contrary, using CaH, the pH of liquid fraction increased due to of OH^- ions (Renou et al., 2008), and pH reached values higher than 12 with application of the maximal dose of chemicals. Surprisingly, the optimal dose of CaH determined during preliminary tests did not give the minimum EC in cattle and pig digestate during separation tests.

Values of the reduced separation index (E_t'(x)) at the optimal dose (Figure 62) attest that the solid fraction increased in concentration more when AlS was used than when CaH was used.

The results concerning P-reduced separation index show that a marked increase (17–67%) occurred when AlS was used on raw or stripped slurries. A lower reduced index was observed only when AlS was added to cattle and pig digestate before the digestate was subjected to stripping.

The use of AlS a greater increase in P-reduced separation index (up to 67%), that were higher than those observed with CaH with the exception of cattle and pig digested.

The reduced separation index for Cu and Zn, showed that the concentrations of these elements increased in the solid fraction of pig slurry and all digestates after the stripping process (Figure 62). This variation is mainly observed using AlS than CaH, for which the E_t' (x) was lower (8-28%).

After the application of ammonia-stripping process, the use of CaH produced a decrease in reduced separation index for pig digestate for both Cur and Zn.

The only case in which the reduced index had a negative value was that of Cu when CaH was applied for digestate after ammonia stripping.

In contrast, compared to the reduced separation indexes for both Cu and Zn when CaH was used, the indexes were significantly larger when AlS was added to raw pig slurry (E_t'(x) 60-70%) and to the ammonia-stripped digestates (E_t'(x) of 70%-80%). The high values for raw slurry might be partly explained by the relatively low pH (6,7) compared to digestate (pH>8). However, the same consideration cannot be applied to stripped digestate for which the pH values were similar to that observed in the un-stripped digestate. Rather, the stripping process might modify the concentrations of the soluble fraction of these heavy metals.

Although the stripping process almost always increased the reduced separation efficiencies for TS, P, Cu and Zn, the differences between un-stripped and stripped digestates were not statistically significant.

Nevertheless, the stripping process produced allowed reaching higher concentration of elements (P, Cu, Zn) in the solid fraction (Table 52). For this reason, stripping process can be considered as an appropriate treatment to apply before solid/liquid separation with CaH and AlS as chemicals.

In addition, the stripping process allows using lower dose of chemical to rich given removal rate compared to what is needed for un-stripped slurry types.

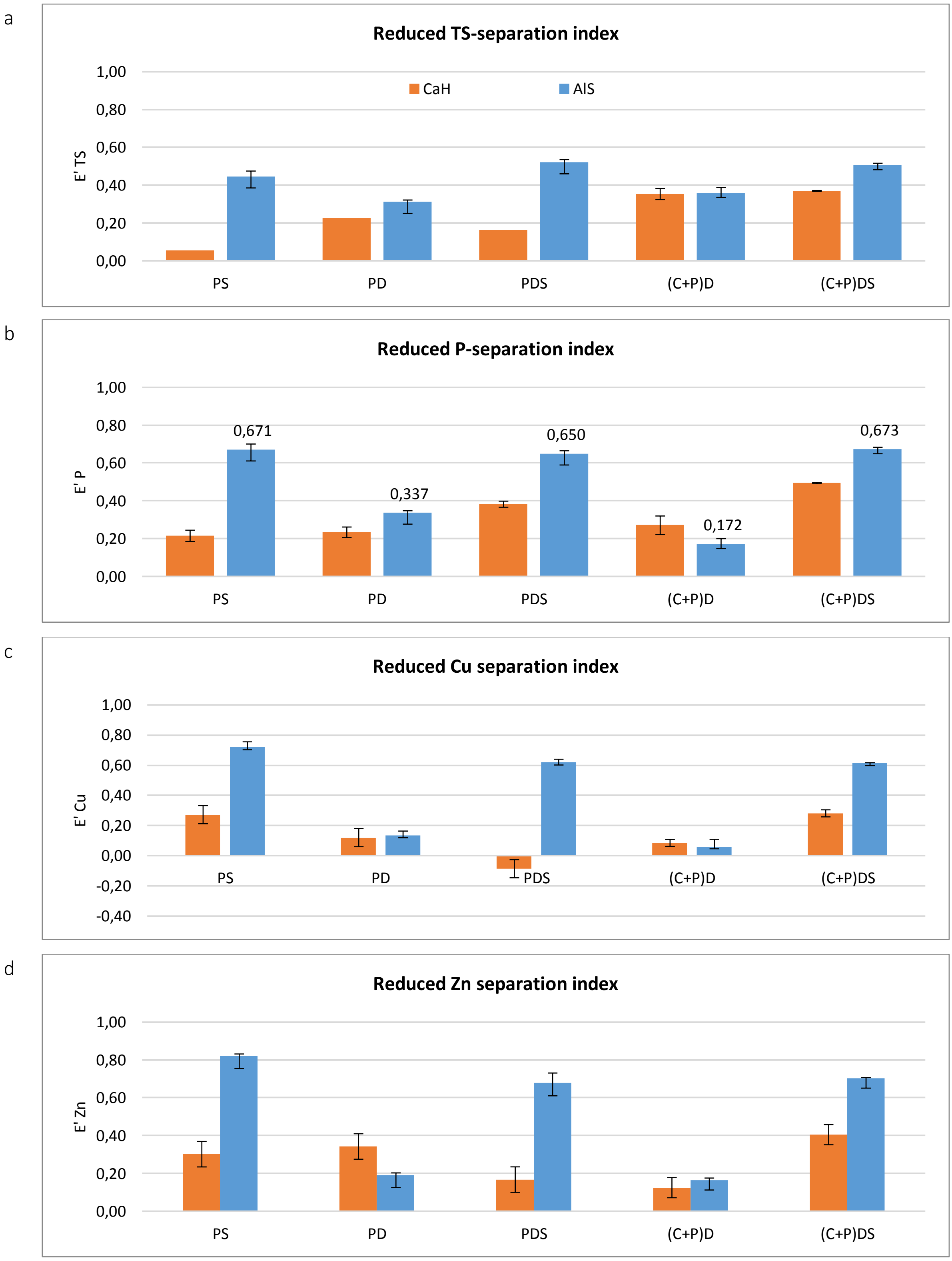


Figure 62 Reduced separation indexes resulting from $Ca(OH)_2$ (CaH) and $Al_2(SO_4)_3$ (AlS) for a): total solids (TS); b) phosphorus (P); c) copper (Cu); and d) zinc (Zn). PS: pig slurry; PD: pig digestate; PDS: pig digestate stripped; (C+P)D: cattle and pig digestate; (C+P)DS: cattle and pig digestate stripped.

In any case, the high doses of chemicals required to achieve high separation efficiencies should be carefully evaluated due to their high cost and potential environmental impact. Nevertheless, it must be considered that the objective of this kind of process is not the complete removal of P or other elements such as Cu and Zn, but only reduce the contents of these elements to match the crop requirements. For this reason, it is possible that, to obtain wanted results, the necessary dose is lower than the optimal one.

Table 52 Significance of the factors considered in the statistical analysis of reduced separation efficiency.

			ST'	P'	Cu'	Zn'
Slurry type	Cattle and pig digestate stripped	Mean	0,27	0,43	0,28	0,39
		Statistical significance	a	a	a	a
	Cattle and pig digestate	Mean	0,25	0,18	0,06	0,12
		Statistical significance	b	c	b	d
	Pig digestate	Mean	0,22	0,29	0,08	0,22
		Statistical significance	c	b	b	c
	Pig digestate Stripped	Mean	0,19	0,40	0,09	0,29
		Statistical significance	d	a	b	bc
	Pig Slurry	Mean	0,15	0,23	0,25	0,36
		Statistical significance	e	c	a	ab
Dose	null	Mean	0,06	0,18	-0,01	0,12
		Statistical significance	c	c	c	b
	Sub-optimal dose	Mean	0,27	0,35	0,21	0,33
		Statistical significance	b	b	b	a
	Optimal dose	Mean	0,33	0,39	0,27	0,37
		Statistical significance	a	a	a	a
Chemical	CaH	Mean	0,16	0,24	0,06	0,18
		Statistical significance	b	b	b	b
	AlS	Mean	0,28	0,37	0,26	0,37
		Statistical significance	a	a	a	a

7.4. Conclusions

The livestock manure represents an important source of some nutrients, which can be recovered and reused.

The study attests suitability of the application of solid/liquid separation for recovering a solid fraction rich in phosphorus from different type of slurries.

The results obtained confirm the possibility to increase the removal efficiency of P, Cu and Zn with the use of additives. In particular, the use of AlS gives removal rates that are higher than those one obtained with CaH.

The maximum efficiencies achievable using chemicals to remove P, Cu and Zn from animal manure slurries depend on the type of slurry and the additive used. In general, of the types evaluated in this study, raw (untreated) pig slurry yielded the lowest separation efficiencies.

Pig digestate and the cattle and pig digestate allow reaching higher than raw pig slurry, but ammonia stripping treatment did not produce significantly better results. Nevertheless, solid/liquid separation complemented with the addition of chemicals and coupled with stripping process can be used for producing a liquid fraction from livestock manure slurries that can be directly use in the field, as well as a solid fraction that is rich in P.

The application of suboptimal and optimal doses of CaH and AlS do not give significantly different results in the separation efficiency of P, Cu and Zn, even though the higher dose does increase the concentration of these elements in the solid fraction. Therefore, the utilization of the suboptimal doses can achieve acceptable results at lower cost compared to those achieved using the optimal dose.

The solid liquid separation of livestock manure using chemicals might increase the concentration of heavy metals in the solid fraction. Therefore, the agriculture use of this should be carefully evaluated in order to avoid over application of this element and their possible negative effects on environment quality and human health.

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SECTION 5 - A POSSIBLE TREATMENT LINE FOR NITROGEN REMOVAL FROM LIVESTOCK MANURE

In this chapter a possible treatment line suitable for the biologically nitrogen removal (denitrification/nitrification process applied in SBR) with attention to ammonia and greenhouse gas emissions control is described.

The proposed treatment line applies a biological nitrification/denitrification process which foresees a conspicuous energy expenditure, as well as a "waste" of nitrogen as N_2 .For this reason, if it is decided to build a new plant, and not to modernize an existing plant, the applicable technologies to be preferred would be those aimed at recovering nutrients and not just their removal and that favouring solutions without too much effort of energy or high investment’s cost.

In general, to decide which treatment or line represents the best option to design, the first consideration to make is the declaration of what should be the removal yield to be achieved, what type of waste must be treated and, also, if the application context is such as to guarantee the correct functioning of the line.

The proposed treatment line has as its final objective the application of the SBR system and appropriate pre-treatments applied to assist the functioning of the biological process. This line, in addition to ensuring maximum environmental protection, aims to achieve acceptable removal of nutrients with adequate investment and operating costs.

8. Livestock manure treatment line for biological removal of nutrients and GHGs emissions control

The treatment line proposed can be a solution for a pig farm that does not have enough land to spread the nutrients produced respecting the limits imposed by the Nitrates Directive.

Main objectives of this treatment line are: 1) obtain two products to use for agronomic purposes (L: liquid fraction, S: solid fraction to be send to an anaerobic process for energy recovery); 2) reducing the nitrogen content to respect the limits; 3) limiting the greenhouse gases and ammonia emissions; 4) proposed different kind of machines, instruments and control devices to make management of the plant easier; 5) optionally, provide phosphorus removal treatment as possible pre-treatment instead of “classical” solid/liquid separation techniques. The considered fattening pig farm is located in Lombardy and has about 3000 pig places.

Using data contained in Ministerial Decree, the quantities of slurry produced, and its characteristics are estimated. In particular, the average weight of each animal is equal to 90 kg and every kg of live weight of the animal corresponds to 37 tons of slurry produced: consequently, in one year the production of livestock slurry in analysed farm is equal to 9990 m^3.

In addition, considering the type of floor installed in the pig-housing structure, it is possible to calculate the annual nitrogen production, which is equal to 35,9 ton.

As pre-treatment of final biological process, it was decided to use two-separation equipment in series in order to obtain a liquid fraction with limited content in solids and organic matter.

The treatment line consists in (Figure 63):

- A reception pit, dimensioned for a capacity of 7 days of production and covered to reduce ammonia emissions;
- A screw press solid-liquid separator with an installed power of 4-12 kW, a flow rate of 40-70 m^3/h and 8 hours/day of working time;

  A flow meter is installed in the pump line feeding the separator in order to control the amount of slurry processed;
- A reception pit of the liquid fraction from the screw press separator with a capacity of 2 days of production;
- The solid fraction (around 10% of the initial volume) is collected on a concrete platform and then moved to the main solid storage;
- A centrifuge to further remove solids with the addition of polymers. Among the various options available, the most commonly used are counter-current drum centrifuges.

  The centrifuge, the feeding pump and the station for the preparation and dosing of the polymers (Figure 64) are controlled by a PLC (Control Local Panel) and the relevant information are recorded in order to control and monitor the process; The solid fraction obtained during this separation phase (20% of the entering volume on average) is temporally collected and then moved to the main solid storage.

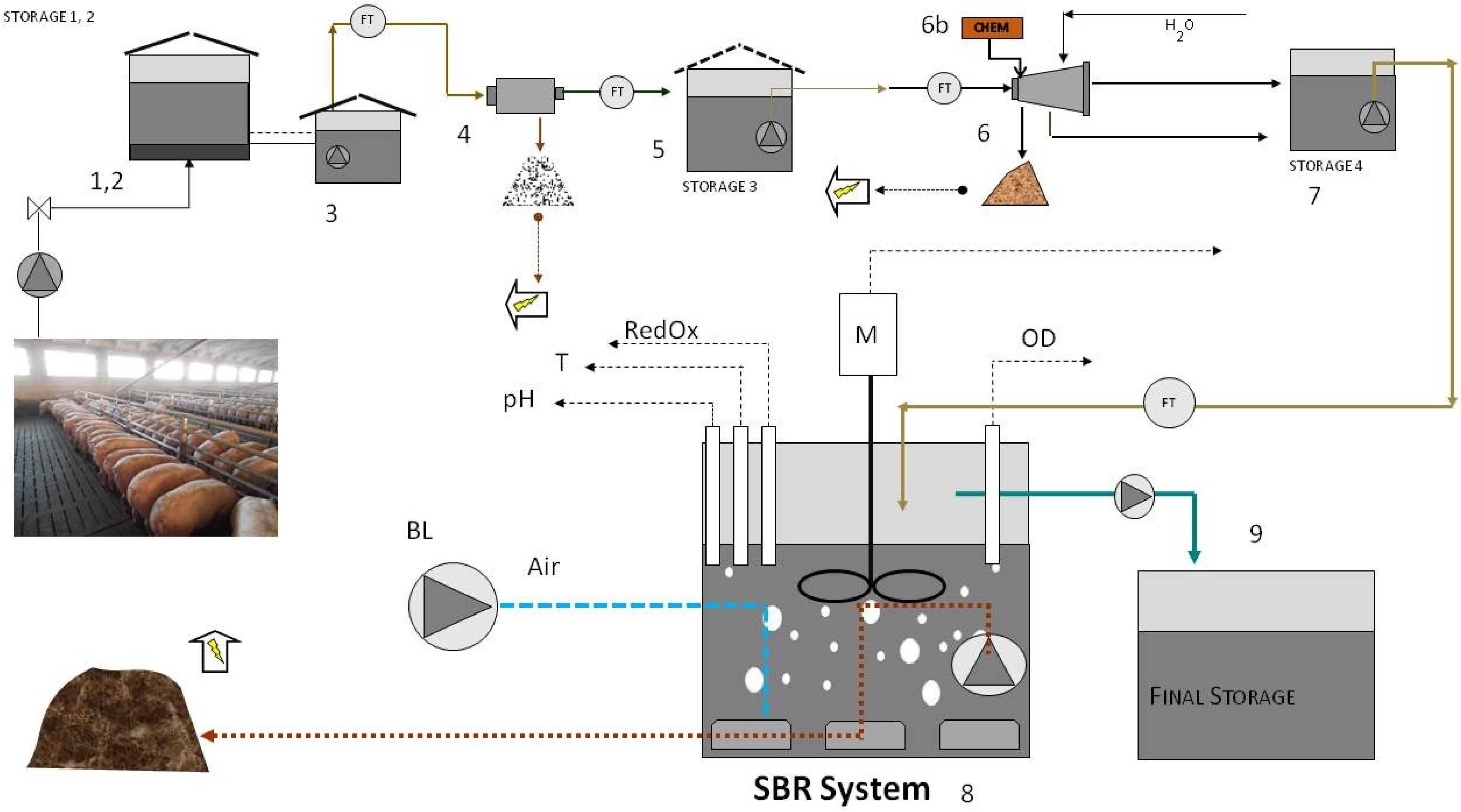

Figure 63 A possible treatment line that includes biological process applied in a sequencing batch reactor and ammonia emissions and GHGs emissions control.

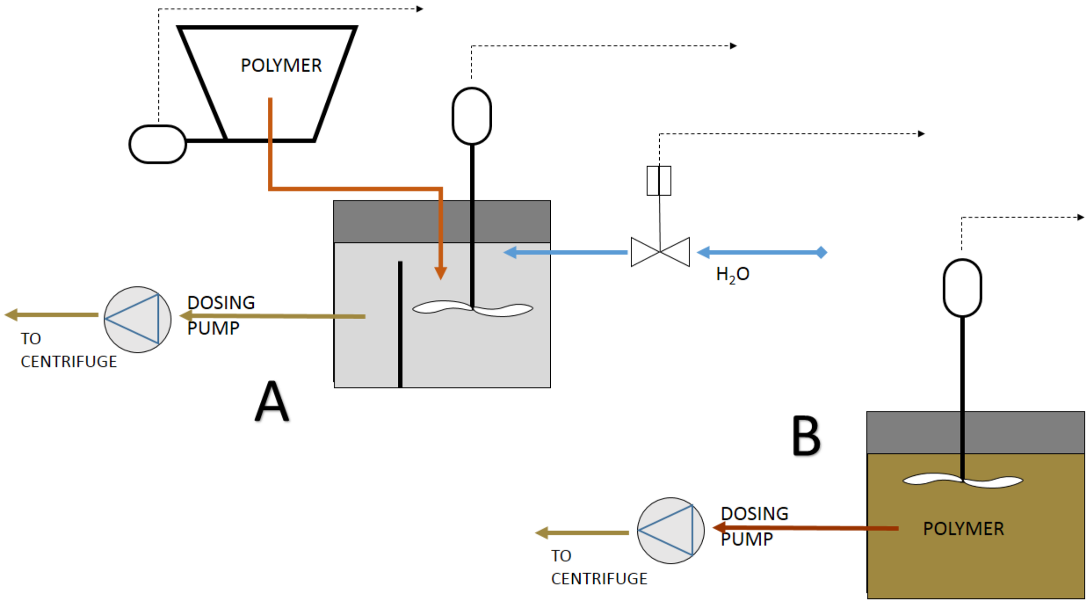

Figure 64 Two different possibilities that can be adopted for the dosage of polymer. The dots line represents the control connection.

- An equalization/buffer tank to receive the liquid fraction from the centrifuge, with a capacity of 2 days. From this tank, the liquid is pumped through a pipeline equipped of a flow meter to feed the biological stage.
- Nitrification/denitrification process occurs in a SBR system, that will be provided with:
    - aeration system, which includes diffuser in an appropriate material (preferably membrane diffusers), connected with a blower that will furnish air. The system is design for be maintained easily: an injection point of acid solution for cleaning pips is provided;
    - mixing system, composed by 2 lateral mixers installed with a proper inclination in order to guarantee the complete homogenization of activated sludge;
    - emptying wells to be able to easily empty the reactor in case of maintenance;
    - loading and unloading pumps;
    - pH detection probe;
    - OD detection probe (dissolved oxygen);
    - temperature detection probe;
    - RedOx potential detection probe; (alternatively, multi-analysis control units are available)
    - Level probe (low frequency radar meter) and detection system with an overflow control for the level of the tank (i.e. the pumps that feed the tank will be automatically turned off when, due to some malfunctioning, the permitted level is exceeded);
- End-product storages.

Two identical storage tanks are sized to accommodate each 90 d of produced slurry, 180 d on total, as required by legislation. The calculation for storage tanks dimensioning are reported in Table 53.

Table 53 Calculation for dimensioning the storage tank.

Storage Tank 1,2		
Material	Concrete, Above ground	
Shape	Circular	
Cover	yes	
Type of cover	HDPE cover	
V1,2 - Volume for storage at least 90 d (m^3)	2497,5	(LFPS/360*180)/2
D3- Tank's diameter (m)	23,8	
A1- Tank's area (m^2)	444,9	
h-Tanks high only for storage (m)	5,6	
H- Tanks high construction (m)	6,0	

Each storage tank is covered with HDPE sheets that rest on a frame of polyethylene pipes that traces the shape of the same storage tank. The choice to cover the storages derives from the need to limit at best of ammonia and greenhouse gases emissions. With this kind of solution, it is possible to limit emissions up to 80% for NH_3 and up to 90% for greenhouse gases compared to the non-covering of tanks. If an existing storage tank has to be used, the hard coverage sometimes cannot be adopted. Nevertheless, there are other valid alternatives to limit emissions, such as the application of flexible covers (light materials, plastic pellets, floating straw tiles), as well as other measures that can be applied (for example acidification of sludge, the cooling of sludge, etc.). Nevertheless, in this case the tank will be sizing in a different way for taking in account the volume of rain that can enter the tank. The different characteristics of the pig slurry, the liquid fraction obtained and the stored slurry that will be sent to subsequent treatments are reported in Table 54.

Table 54 Characteristics of farm produced slurry considering the breeding type and type of manure removal.

	Volume (m^3)	TKN (Kg/m^3)	TAN (Kg/m^3)	DM (Kg/m^3)	VS (Kg/m^3)	K (Kg/m^3)	P (Kg/m^3)
Pig Manure	9990	3,6	2,0	108,5	83,8	3,2	2,0
Liquid fraction	6968	2,6	2,4	38,8	22,3	3,7	0,5
In the storage	9990	3,1	2,3	104,3	79,6	3,2	2,0

A possible solution for calculating the volume of biological reactor is to define the quantity of nitrogen that need to be removed and nitrification speed. An example of calculation of nitrification speed is reported in Table 55. In that specific case, is necessary to remove 16335,4 kg/year – 44,75 kg/d (option B, 12705,3 kg/year – 34,81 kg/d) of TKN for reaching removal rate equivalent to 90% of TKN present in the slurry (option

B is 70%). In this *preliminary project*, instead, the volume defined is INDICATIVE and will be calculated imposing the hydraulic retention time.

Table 55 How calculate nitrogen removal speed (in red cells the imposed values, in grey the calculated values).

v_T	Nitrification speed	$(v_N)_{20}$*TKN/(TKN+K_{TKN})*O.D/(O.D+$K_{O.D}$)*$\theta^{(T-20)}$	
$(v_N)_{20}$	Maximum Nitrification speed at 20°C	3	KgTKN/(KgSSv*d)
θ	Constant of temperature adjustment	1,07	-
T	Temperature	12	°C
TKN/(TKN+K_{TKN}) (option a)	-	0,996	-
TKN/(TKN+K_{TKN}) (option b)	-	0,999	-
O.D/(O.D+$K_{O.D}$)	-	0,71	-
θ^(T-20)	-	0,58	-
K_{TKN}	Semi-saturation constant (TKN)	1,00	mgTKN/L
$K_{O.D}$	Semi-saturation constant (O_2)	1,00	mgO_2/L
O.D	Dissolved Oxygen	2,50	mgO_2/L
TKNe (option a)	TKN a@ output	260,5	mgTKN/L
TKNe (option b)	TKN b@ output	781,5	mgTKN/L
$v_{T\ a}$	Nitrification speed	1,24	KgTKN/(KgSSv*d)
v_{Tb}	Nitrification speed	1,25	KgTKN/(KgSSv*d)
X,tank	Biomass concentration	3,00	KgSST/m^3
SSV/SST	-	0,70	-
$v_{T\ a,1}$	Nitrification speed	2,61	KgTKN/(m^3*d)
$v_{Tb,1}$	Nitrification speed	2,62	KgTKN/(m^3*d)

Fixing HRT of 15 d, the tank volume will be equal to 300 m^3. Considering a concrete tank with circular shape, and high of 4,5 m, the diameter is equal to 2,5 m. The tank is slightly oversized in order to guarantee possible changes in the quantity of manure to be treated, as well as in the need to modify the process parameters to cope with unexpected fluctuations in the characteristics of the slurry. Furthermore, in order to be sure that the designed reactor is adequate for plant engineering needs, it is necessary to define the SRT, the production of daily sludge, the nitrifying biomass in the system and the biomass activating the system.

Considering that, the oxygen consumption is equal to 4,6 kg O_2 per kg N oxidized, it is possible to calculate approximately the required oxygen quantity to be supplied to the activated sludge. Obviously, this quantity must be increased with active-respiration contribution (that is connected to the oxidation and synthesis reactions) and with the endogenous respiration contribution.

Once the quantity of oxygen necessary for nitrification has been defined, the oxygenation capacity of an aeration system must also be **estimated** in order to choose the proper aeration system. In that case, considering owned data, the quantity of O_2 required is equal to 206 kgO_2/d and to 160 kgO_2/d respectively for removal rate of 90% and for 70%. The calculations are reported in Table 56.

To summarize, in order to size properly and correctly an aeration system it is necessary to define:

- operating conditions;
- oxygen required by the system;
- oxygenation capacity for the real operating conditions;
- the variation of the oxygenation capacity during the different periods of the year;
- definition of the maximum value of oxygenation capacity;
- the appropriate equipment.

Table 56 Calculation of oxygen demand, without a specific definition of required parameters. These represent only estimation.

	Option I	Option II
Removal efficiency	-90%	-70%
Vin m^3/d	19,09	19,09
TKN removed (Kg/d)	44,75	34,81
$KgO_2/KgNox$	4,60	4,60
KgO_2/d	205,87	160,12
Aeration system transfer yield kgO_2 / kWh (0,8-1,3)	1,05	1,05
kWh required/day	196,1	152,5

Start-up of the plant

Only 1 reactor (n=1) for processing 19 m^3/d of slurry is projected.

This SBR is drawn from the storage tank marked with the number 7 in Figure 63. The SBR reactor will be initialized by means of an activated sludge inoculum coming from a well-run plant. In particular, the activated sludge will be bringing from a plant finalized to the treatment of livestock manure that discharge effluent into surface waters, or into sewers; however, if this solution will not be applicable, the new reactor will be inoculated with activated sludge from a wastewater treatment plant of civil or mixed slurry treatment.

Table 57 Division of cycle's duration in different phases.

Phase	**Duration (h)**
Cycle length	8,0
Fill	1
Reaction (total)	5,25
Denitrification phase	*2,00*
Nitrification phase	*3,25*
Sedimentation	0,75
Discharge	1,00
Idle Phase	-

A cycle length equal to 8 hours is imposed: this value was used both for laboratory tests (Chapter 5), both in some analysed researches (Chapter 3). The cycle setting is reported in Table 57.

The general subdivision of the operational phases of the SBR reactor will include: static filling phase, the anoxic phase (in that way denitrification occurs using the internal carbon compounds and, theoretically, avoids the addition of external carbon), oxic phase, sedimentation, discharge of liquid fraction and discharge of sludge (sometimes). In order to feed and empty the biological system, two volumetric pumps will be installed. The flow rate of each pump is the same, as consequently the charge and discharge time. The selected pump will have a flow rate equal to 6,5 m^3/h: in that way the charge and discharge time will be equal to 1 h.

Alternative for P-recovering

As explained in chapter 7, the processes aimed at the recovery of elements whose concentrations are quite high in livestock slurry are taking hold. For this reason, as alternative to one of the separation processes, and in particular to the *separation phase 2 (centrifuge)*, is represented by a coagulation/flocculation process, using as chemicals aluminium sulphate and a natural polymer. This kind of treatment allows removing and recovering a semi-solid fraction rich in phosphorus and heavy metals. This fraction can be ceded to an industry dedicates in P-recovery or use it on the land that need P and micronutrients. In addition, the high removal solid reachable with this treatment allows obtaining more clarified slurry, that can permit the better operation of SBR. An solid/liquid separation option could be the one proposed in Figure 65.

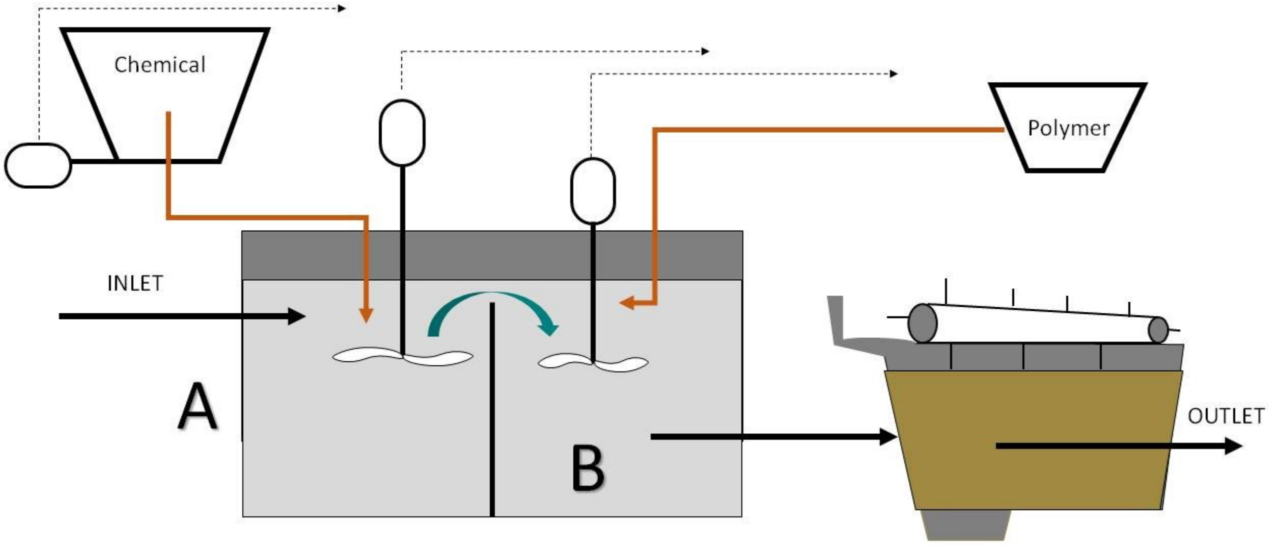

Figure 65 Coagulation/flocculation phase that can substitute the separation using centrifuge. Dot lines represent the control wire.

The proposed treatment line is only one of the possible solutions for a correct management of livestock manure, that allows to respect law requirements, to protect environmental, to limit emissions and try to recover nutrients (phosphorus in particular) from livestock manure.

8.1. References

Santonja, G. G., Georgitzikis, K., Scalet, B. M., Montobbio, P., Roudier, S., & Sancho, L. D. (2017). Best Available Techniques (BAT) Reference Document for the Intensive Rearing of Poultry or Pigs.

GENERAL CONCLUSIONS

9. Conclusions and future perspectives

Livestock manure treatment plays a key role in protecting environment when the amount of nutrients contained in that product is higher than the field capacity to receive them.

Many of the treatments commonly used are aimed at “only” removing nutrients (N and P) from livestock manure. However, is worth considering the application of treatments that allow also producing valuable by-products: the animal slurry, therefore, if properly treated, becomes a resource rather than a waste. Currently, in the context of livestock manure treatment, nitrification/denitrification process aimed to only removal of nitrogen is widely applied.

Nowadays, the treatment plants sizing is often done by applying standard procedures and not considering specific situations. This choice leads to the implementation of systems that do not allow achieving the desired results in terms of removal efficiencies and costs.

Nevertheless, scientific literature reports the possibility to achieve very high removal efficiencies if the treatment plants are properly designed.

A very important stage along treatment chain is represented by the pre-treatment application for removing coarse and fine solids from slurries before the biological process. Furthermore, it is necessary an excellent knowledge of the applied process, its development and control in order to obtain the expected performances, as also attested by scientific literature and by pilot/lab scale experiments carried out.

The use of RedOx, pH, dissolved oxygen probes etc. represents a necessity for a successful process control. Consequently, the maintenance and the proper use of these tools is required.

Thinking about the future of SBR systems applied for livestock manure treatment, one possibility could be to merge biological process applied in SBR with other kind of chemical-physical processes focused on recover of nutrients. For example, since during biological process nitrogen can be lost during aeration phase, if an SBR system is coupled with gas capture system (maybe also from storage along the treatment line), the nitrogen losses during aeration phase and during slurry storage can be collected and recovered. In this way, the desire removal rate will be reached, the environment will be protected and the costs will be reduced maybe selling the recovered products. This kind of solution will also reduce other problems connected with biological

treatment plants, as NH_3 and GHGs emissions that usually required the application some mitigation techniques.
It may be possible to apply the same principle and combine biological process already present in a farm with other type of technology. It could be promising to combine SBR or other biological process with recovering of phosphorus, which is considered more scarce resource than nitrogen. In this way, even if the nitrogen would be "lost" as N_2, the phosphorus would be recovered.

Considering the final goal of livestock manure treatment for nutrients removal, in the future it will be necessary to face with further environmental pollution problems. This fact will lead to the imposition of increasingly stringent regulatory limits that will be applied not only to civil and industrial wastewater, but, probably, to other pollution's source.
Besides nitrogen and phosphorus, it will be mandatory to limit the environmental risk connected also with emerging pollutants as antibiotics or heavy metals present in the animal livestock manure. In fact, livestock manure generated by confined animal feeding operations represent a major source of antimicrobials and antimicrobial resistance genes (ARGs) in the environment. Also in this case, the commingling and the integration of several treatments could represent a solution for these emerging problems.

ACKNOWLEDGMENTS

“Ringraziamenti”

Grazie. Gracias. Merci.

a voi, che mi avete fatto ridere durante questi tre anni,

a voi, che mi avete permesso di imparare,

a voi, che mi avete dedicato il vostro tempo,

a voi, che mi avete ascoltato,

a voi, che mi avete supportato, sempre,

a voi, che mi avete trasmesso la passione per ciò che fate ogni giorno,

a los compañeros de IRTA, os extraño muchissimos,

agli amici - a los amigos, viejos y nuevos, (il tesoro più grande),

ai colleghi..diventati amici,

ai *miei* ingegneri,

alle mie Amiche,

alle Amiche.. diventate colleghe,

a mio padre,

a mia madre,

a chi amo,

“Il miglior riconoscimento per la fatica fatta non è ciò che se ne ricava, ma ciò che si diventa grazie ad essa”

J. Ruskin