SAFE

Sustainable water reuse practices improving safety in agriculture, food and environment

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Report on the high rate anaerobic bioreactor prototype, optimization of the operating conditions and preliminary tests results

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TASK T1.3 Decontamination *(task leader: M. Concetta Tomei)*

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ABBREVIATIONS

ACP anaerobic contact process

AF anaerobic filter

ABR anaerobic baffled reactor

AnMBR membrane anaerobic membrane bioreactors

COD chemical oxygen demand

EGSB expanded granular sludge bed

ER exchange ratio

FB fluidized bed

HRT hydraulic retention time

IC internal circulation

SRT sludge retention time

TSS total suspended solids

UASB up-flow anaerobic sludge blanket

VFA volatile fatty acid

VSS volatile suspended solids

EXECUTIVE SUMMARY

The development of innovative and sustainable technologies for domestic wastewater treatment places in the highlight anaerobic process conducted in high-rate anaerobic reactors. This report is firstly focused on the technological evolution starting from the conventional anaerobic treatment processes operated with suspended biomass and following the evolution to the anaerobic high-rate systems, which received great attention in the last years, due to their high load handling capacity and low sludge production. Different configurations have been proposed for treating industrial and also municipal wastewater, such as anaerobic contact process, anaerobic filter, up-flow anaerobic sludge blanket reactor, fluidized bed reactor, expanded granular sludge bed reactor, internal circulation reactor, anaerobic baffled reactor, and membrane anaerobic membrane bioreactors. Among them, particular attention was given to the Upflow Anaerobic Sludge Bioreactor (UASB) recognized as one of the best available technology so far. In particular, UASB was successfully implemented and established within a wide acceptance in municipal wastewater treatment plants in warm climate areas.

Within the framework of WP1 of SAFE project related to sustainable solutions as applied innovative technologies for domestic wastewater treatment, the objective of IRSA-CNR activity is to investigate and optimize the performance of a high-rate UASB bioreactor operated with granular biomass. Final goal is to verify its feasibility in terms of contaminant removal and energy and resource recovery in moderate climate areas. The proposed process has been optimized by managing different operating parameters such as wastewater temperature (25 and 35 °C) and Hydraulic Retention Time (HRT, 22, 14 and 9 h).

Experimental results confirmed the optimal performance of UASB technology applied to the treatment of domestic wastewater, with excellent COD removal efficiencies (84-94%), good biogas production (0.14-0.27 $m^3/kg_{CODremoved}$), no accumulation of VFA in the bioreactor, indicating the good stability of the anaerobic system, good effluent quality in terms of COD and suspended solids, and, finally, high nitrogen and phosphorus concentrations in the treated effluent suggesting their potential recovery for agriculture purpose.

These first results are a first step in the evaluation of the feasibility of the UASB technology as an energy generating process and cost-effective alternative for wastewater treatment which is also able to produce nutrient rich and solids free effluents with a high degree of pathogen removal, while occupying a small footprint.

1. Introduction

1.1. State of the art analysis of high rate anaerobic systems applied to wastewater treatment

In recent years, due to current concerns on global risks derived by fossil fuel dependence and step-increasing energy cost, resources depletion and increasing climate change due to green-house gas emissions, along with worries over public health and environmental protection, anaerobic technology for wastewater treatment is increasingly being investigated and consequently applied. Essentially anaerobic process, breaking complex organic substrates into biogas that is substantially composed of a mixture of methane and carbon dioxide, is a valid and sustainable alternative in comparison to the conventional aerobic treatment technologies. The rationale behind this can be summarized in some key points including the easy applicability, low space requirement, up to 90% of reduced sludge production, and a positive energy balance. Indeed, anaerobic treatment plants have the potential to become net producers of renewable energy, converting the chemically bound energy content in the organic pollutants of raw wastewater to useful energy carrier. In addition to the energy that can be recovered from methane-rich biogas, the application of anaerobic processes distinctly reduces the overall energy demand for wastewater treatment because no aeration energy is required for mineralizing the organics (Ozgun et al., 2013). Indeed, conventional aerobic process is an energy-intensive treatment, accounting for more than 50% of the total energy consumption in a typical wastewater treatment plant (Vinardel et al., 2020). On the contrary, Van Lier et al. (2015) reported a fossil fuel energy saving of ~1 $kWh/kg_{CODremoved}$ with anaerobic treatment instead of aerating activated sludge and a production of 13.5 $MJ/kg_{CODremoved}$, giving 1.5 kWh electricity (assuming 40% electric conversion efficiency).

Moreover, along with bio-energy generation, the potential recovery of resources comprised in treated effluents is a sustainable and convenient benefit of anaerobic treatments. Indeed, starting from the conventional anaerobic treatment processes to 'closing the loop' scheme, the concept of bio-refinery in wastewater treatment plants prospects to serve for water, energy and materials mining. While bio-energy is still dominating the resource recovery, recovery of value-added materials (i.e. struvite, biopolymers, cellulose) has drawn considerable attention in recent years (Akyol et al., 2020). In particular, nutrient recovery and recycling take a significant role in circular economy: recovered nutrients can be utilized as soil amendments or fertilizers for beneficial uses in agriculture. Nutrient recovery from wastewater can also reduce the maintenance cost of wastewater treatment facilities and avoid environmental impacts (Song et al., 2018).

Many of stated positive aspects of anaerobic technology, especially the ones related to resource recovery, are certainly included within the context of SAFE project: besides high treatment efficiency, the improvement of effluent quality and the possibility of water reuse are key points of the proposal targeted to solve water scarcity and to promote green agricultural practices. Among these aims, the development of sustainable solutions for wastewater treatment based on anaerobic process is the first objective of IRSA-CNR activity inside the SAFE programme.

A preliminary literature analysis has been carried out according to the work schedule planned for the 1st year with reference to high-rate anaerobic reactors, and in particular UASB technology, to confirm that these systems are powerful treatment devices with outstanding potentialities in terms of contaminant removal and energy and resource recovery (Stazi and Tomei, 2018).

1.2. Brief evolution of high rate bioreactors

All the mentioned outstanding advantages of anaerobic process drove research focus on the development of innovative and advanced technologies such as anaerobic high-rate systems, which received great attention in the last years, due to their high load handling capacity and low sludge production (Mainardis et al., 2020). Many of their configurations were firstly applied to the treatment of industrial and/or high-strength domestic wastewater and recently also to low-strength effluents with the consequent outcome of evolving anaerobic treatment into a competitive technology level (Stazi and Tomei, 2018). High-rate systems have revolutionized the sustainability of industrial wastewater treatment and could additionally provide an alternative for municipal wastewater (Trego et al., 2021).

The basic concept of high-rate systems is to overcome the low growth rates of anaerobic microorganisms, by retaining biomass within the bioreactors using advanced technologies. Key to the success of high-rate anaerobic reactors is that they allow the uncoupling of the sludge retention time (SRT) from the hydraulic retention time (HRT), thus providing a reduction of required reactor volumes and associated costs, making anaerobic treatment of practical interest for cost-effective wastewater treatment (Van Lier, 2008). SRT and HRT are both time-related concepts in the anaerobic treatment system: SRT represents the sludge age, i.e. the biomass retention time in the reactor, while HRT refers to the nominal value of the liquid (and dissolved matter) retention time. The way of uncoupling SRT from HRT gives a first classification of high-rate anaerobic systems: immobilization of sludge via granule and/or biofilm is the most traditional option; moreover, physical retention throughout a filtration barrier or a membrane or by using a secondary clarifier with sludge return, similar to the activated sludge process, can be also achieved. This last option is the less efficient; it has been applied in the early developed systems and at present has been substituted by more efficient solutions. Moreover, high-rate systems can operate with suspended or attached-growth processes including expanded/fluidized bed reactors and fixed-film processes. In the first case, the sludge is present as flocs or granules, whereas in attached growth systems biomass can adhere to a moving or fixed medium. Hybrid systems combine suspended- and attached-growth processes in a single reactor unit to exploit advantages of both types of biomass.

Depending on the applied sludge retention mechanism, various configurations of high-rate systems have been developed, such as anaerobic contact process (ACP), anaerobic filter (AF), up-flow anaerobic sludge blanket (UASB) reactor, fluidized bed (FB) reactor, expanded granular sludge bed (EGSB) reactor, internal circulation (IC) reactor, anaerobic baffled reactor (ABR), and membrane anaerobic membrane bioreactors (AnMBR). Table 1 gives an overview of above named high-rate anaerobic reactors.

Following the historic evolution of high-rate reactors, the ACP process was the first configuration of these systems in which the separation between SRT and HRT was accomplished by an external clarifier with return flow, similar to its aerobic homologue. Schroepfer et al. (1955) firstly designed ACP for packinghouse wastes, however first applications of ACP did not exert positive outcomes, because of a scarce separation of active anaerobic sludge from the treated water in the second clarifier (probably due to a high intensity of reactor mixing) and a post-treatment requirement (to comply with effluent restrictions). Recent ACP configurations faced these limitations by resorting to milder mixing conditions and degasing units before external clarifier (Van Lier et al., 2015).

In 1960s, Young and McCarty (1969) began working on the development of AF process, by introducing inert support material, like rocks or plastics, into the bioreactor on which the anaerobic organisms can adhere. This represented a significant advancement, since the filter can trap and maintain a high concentration of biological solids, thus ensuring long SRTs and low HRTs. AF technology has diverse strengths: it can be applied in up-flow and down-flow reactors, guarantees a rapid start-up and is less sensitive to shock loads, thus requiring small volumes and concomitant costs (Stazi and Tomei, 2018).

The main disadvantage of this process is the difficulty in maintaining the required efficient contact between sludge and wastewater, because of easy clogging of the packed bed (especially for wastewaters containing suspended matter). Nevertheless, in spite of the unnumbered full-scale applications to beverage, food-processing, pharmaceutical, and chemical industry wastewater from 1981 until 2007 (Van Lier, 2008), AF process was not able to be consolidating in the market in the last decades.

An important turning point for high-rate biotechnology was the biomass immobilization, even without support media, which was started to be observed in 1970's. Auto-immobilization concept consists in the immobilization of bacteria on themselves or on very small inert or organic particles present in the wastewater, forming dense bacterial conglomerates that mature in due course and naturally lead to round shaped granular sludge. First studies on sludge granulation dated prior to UASB implementation, whit different direct observations by Young and McCarty (1969) in an AF system or in an up-flow Dorr Oliver Clarigesters in South Africa (Hulshoff Pol et al., 2004). However, little attention was paid to this sludge type at the beginning, and only 10 years later, Lettinga and his co-workers developed the first granule-based high-rate reactor, i.e. an UASB system, in Netherlands (Lettinga et al., 1980). Granulation fundamentals are not completely understood at the present, despite the number of existing granulation-based treatment plants (Show et al., 2020). Granule composition was shown to modify in continuous operations, depending on the treated substrate (Na et al., 2016). In general, anaerobic granule formation is mostly observed in anaerobic bioreactors, which are operated in up-flow mode (Hulshoff Pol et al., 2004).

In UASB systems, the key of this process was to provide the physical and chemical conditions for sludge flocculation, and, as a consequence, high SRT (at high HRT loadings) with the separation of the gas from the sludge solids. Indeed, the sludge retention in this reactor is based on the formation of well settleable sludge flocs or granules, and on the application of a reverse funnel-shaped internal gas–liquid–solids separation (GLSS) device. Highly active biomass is retained in the reactor without packing material, and the reactor can be operated only with suspended entrapped biomass under high hydraulic retention times without any fear of biomass washout. Additional advantages of UASBs include low excess sludge production, robustness in terms of COD removal efficiency, ability to cope with fluctuations in temperature, pH and influent concentration, and quick biomass recovery after shutdown. The main limitation of UASB process is related to wastewaters with high suspended solids content, which hampers the development of dense granular sludge (Van Lier et al., 2015).

Due to the above-mentioned features, the application of UASB systems rapidly increased from their introduction, with excellent reported performances on different biodegradable wastewater streams (Mainardis et al., 2020). The first systems were installed for the treatment of food, beverage, and agro-based wastewaters, rapidly followed by applications for paper and board mill effluents (Van Lier et al., 2015). Moreover, subsequent successful applications of UASBs included the treatment of a wide range of effluents such as sugar, pulp and paper, dairy, chemical, potato starch, bean balancing, soft drinks, fish processing, noodle processing, yeast production, slaughterhouse, and coffee processing industries (Daud et al., 2018).

The need to optimize biomass-wastewater contact, and to overcome such problems as preferential flows, dead zones and hydraulic short cuts that might occur in the UASB reactor, led to the development of fluidized bed and expanded bed systems, considered the second generation of anaerobic sludge bed reactors, including FB, EGBS and IC technologies. The idea was to expand the sludge bed and to increase hydraulic mixing, with comparison to UASB, in order to enhance substrate-biomass interaction (Akyol et al., 2020).

The FB process is based on the growth of the biomass as a bio-layer around non-fixed or mobile carrier particles, which consist of, for example, fine sand (0.1–0.3 mm), basalt, pumice, or plastic (Heijnen et al., 1989). Firstly realized for aerobic applications in early 1970's, during mid-70's, FB have been also studied and developed for anaerobic treatment. For example, applications to different industrial wastewater, such as textile wastewater, ice-cream wastewater, and brewery wastewater, winery wastewater from grape-red and tropical fruit, currant and sanitary landfill leachate are reported (Jaafari et al., 2014). Main advantages of FB process include high efficiency and small volume requirements. In addition, a high up-flow influent velocity contributes to the positive features of this configuration, i.e. good mass transfer resulting from liquid turbulence and high flow rate around the particles, less clogging and short-circuiting because of the occurrence of large pores through bed expansion, and high specific surface area of the carriers because of their small size. However, FB disadvantage is mainly related to the long-term stable operation: indeed, the system relies on the formation of a more or less uniform (in thickness, density, and strength) attached biofilm. To maintain a stable biofilm development, a high degree of pre-acidification is considered necessary and dispersed matter should be absent in the feed. Despite that, an even film thickness is very difficult to control and in many situations a segregation of different types of biofilms over the height of the reactor occurs (Van Lier et al., 2015). Moreover, bare carrier particles may segregate from the biofilms, especially in full-scale reactors, and this necessarily requires continuous flow adjustments, for example lower up-flow velocities, which is opposite to the FB concept.

The EGSB reactor, usually considered an upgrade of the conventional UASB system, employs granular sludge, which is characterized by good settling characteristics and a high methanogenic activity, with applied volumetric loading rates and upward flow velocities that are higher in comparison to UASBs. The idea of sludge bed expansion was applied to use efficiently the entire reactor volume and to reduce dead zones in the reactor (van der Last and Lettinga, 1992). The solution consisted in applying a higher up-flow influent velocity (i.e. exceeding 6 m/h) by using an adequate height/diameter ratio and/or high effluent recirculation. These hydrodynamic conditions, together with the lifting action of gas evolved in the bed, assure an extended height of the expanded granular bed, leading to an excellent biomass-wastewater contact and a better performance and stability compared to conventional UASB reactors. Accumulation of flocculent excess sludge between the sludge granules is also prevented (Seghezzo et al., 1998). Interestingly, EGSB systems can be applied to several types of wastewaters that cannot be treated using conventional UASBs, such as wastewaters containing highly toxic but anaerobically degradable components (such as formaldehyde or dyes) or with cold (<10 °C) and dilute (COD <1 g/L) wastewaters, i.e., when specific gas production is very low and biogas mixing is absent. This is because applied recirculation of the effluent in EGSB dilutes the influent concentration, which is a non-favorable condition for anaerobic treatments but may also allow the treatment of toxic compounds or satisfactory efficiencies at low temperatures (Seghezzo et al., 1998; Stazi and Tomei, 2018). Main limitation of EGSB is that is not suitable for the removal of particulate organic matter due to the high up-flow liquid velocity, thus the influent suspended solids that are not retained by the granular bed will eventually leave the reactor together with the effluent (Sikosana et al., 2019).

Table 1 Overview of high-rate anaerobic reactors

Process	SRT/HRT uncoupling	First application	Advantage	Limitations
ACP	Secondary clarifier	1955	High efficiency for concentrated wastewaters with high SS concentrations	Poor sludge separation Post-treatment requirement
AF	Filtration	1969	High efficiency with low strength wastewater Up-flow or down-flow Rapid start-up High resilience to shock loads Low costs	Bed clogging problems Pre-treatment requirement Long-term instability
UASB	Granulation	1980	Good biomass-wastewater mixing Effective solids/liquid separation High exploitation of reactor volume Low energy consumption No packing material cost	Low performance for high-solid content wastewaters Long start-up (without available granular biomass)
FB	Biofilm	1980	High efficiency Limited bed clogging problems Small volume requirements	Long-term instability Biofilm thickness control requirement
EGSB	Granulation	1992	Excellent biomass-wastewater mixing Excellent efficiency No accumulation of flocculent excess sludge Possibility of treatment of toxic compounds Good performance at low temperatures	Scarce SS removal
AFB	Granulation	1983	No bed clogging problems Low cost Easy operation Low sludge production	Low SRT by hydrodynamic limitation Influent distribution
AnMBR	Filtration	1978	High efficiency Short start-up Small volume requirements High effluent quality	Membrane fouling High cost and energy requirements

Because the EGSB systems rely on a complete retention of the granular sludge, efficient sludge separation at the top part of the system is of the utmost importance. The various contractors supplying EGSB reactors have their own typical features for separating actively the sludge from the liquid and gas flow, applying specifically designed GLSS units (Van Lier et al., 2015). A special version of the EGSB concept is the so-called Internal Circulation (Biopaq®IC) reactor, in which the produced biogas is separated from the liquid halfway the reactor by means of an in-built GLSS device and conveyed upwards through a pipe to a degasifier unit or expansion device. Here, the separated biogas is removed from the system, whereas the sludge-water mixture drops back to the bottom of the reactor via another pipe. In fact, the lifting forces of the collected biogas are used to bring about a recirculation of liquid (and granular sludge) over the lower part of the reactor, which results in an improved contact between sludge and wastewater.

At the same time as Lettinga group developed the UASB system, ABR was pioneered by McCarty and co-workers (Bachmann et al., 1983, 1985) as a "series of UASB reactors". Indeed, although ACP, UASB, and EGSB reactors are based on a mixed to completely mixed reactor content, various designs have been tested that employ staging of the various phases of anaerobic treatment, creating a plug-flow in the waterline (Van Lier et al., 2001). An example is the two-stage process where the acidification step is completely separated from the following methanogenic step, which is realized in ABR. This system is a horizontal multi-staged bioreactor with a high biomass retention time due to the forced flow of wastewater through various compartments. Hence, hydrolytic and organic acid producing bacteria are isolated from methanogens with multiple compartments separated by a series of vertical baffles, through which wastewater moves upward and downward between the partitions along the reactor. One of its great merit lies in its ability to separate the processes longitudinally, making the reactor an easy-controlled, low-cost, and multiple-stage system (Lau and Trzcinsk, 2022). Other benefits with respect to the other high-rate anaerobic reactors include higher tolerance to hydraulic and organic shock loads and lower sludge yields. Moreover, ABR is simple to build and operate and needs reduced maintenance (Stazi and Tomei, 2018). On the contrary, a problem of concern is the hydrodynamic limitation giving constraints to the realizable SRT in the system, because the superficial liquid velocity in a baffled system is substantially higher than in a single-step sludge bed reactor. As a result, the sludge mass may slowly move with the liquid flow through the various compartments (Van Lier et al., 2015). Other limitations to pilot/full-scale applications include the requirement to build shallow reactors for maintaining acceptable liquid and gas up-flow velocities, and problems with ensuring an even distribution of the influent. Indeed, although the staged reactor concept showed very promising results on a pilot scale, full-scale reactors are very scarce for this type of high-rate reactor.

Although well-established UASB and/or EGSB technological configurations mostly meet the requirements necessary for anaerobic treatment, unfavorable conditions regarding the granule disintegration led to the development of AnMBR by coupling anaerobic treatment with low-pressure membrane filtration. The first test of the concept of using membrane filtration with anaerobic treatment of wastewater has been reported in 1970s' (Grethlein, 1978), then the first commercially-available AnMBR was developed by Dorr-Oliver in the early 1980s for high-strength whey processing wastewater treatment (Li, 1985). Afterwards, in the decade of 1990s, AnMBR research activity increased with investigations into different membrane materials, characterization of membrane foulants and development of strategies for membrane cleaning and fouling management. Also, immersed membranes started implementation (Jain, 2018).

AnMBRs offer high quality effluents free of solids and complete retention of biomass, regardless of their settling and/or granulation properties. Furthermore, AnMBRs can be used to retain special microbial communities that can degrade specific pollutants in the wastewater. Therefore, this technology may present an attractive option for treating industrial wastewaters at extreme conditions, such as high salinity, high temperature, high SS concentrations, and presence of toxicity, which hamper granulation and biomass retention or reduce the biological activity (Van Lier et al., 2015).

Recently, AnMBR has also started to be investigated for municipal wastewater treatment as a possible alternative to the conventional aerobic treatment processes (Martin, et al., 2011). In addition to achieving high effluent qualities, a shorter start-up period is required for AnMBRs in comparison to UASB systems, which is one of the major advantages in the treatment of especially low-strength wastewaters (Ozgun et al., 2013).

Despite the mentioned advantages, there are still critical obstacles such as low flux, membrane fouling, high costs that limit the extensive use of AnMBRs (Ozgun et al., 2013). Indeed, the AnMBR process inherently uses more energy to generate permeate flow through the membrane and for membrane fouling control (headspace methane is generally used for membrane scouring). To date, the AnMBR process has been successfully adopted for organic-rich industrial waste streams disposal at full-scales, such as alcohol distillery stillage, high-strength industrial wastewater, food wastes and municipal solid wastes; however, its effectiveness in the treatment of low-strength municipal wastewater has yet to be confirmed since only several pilot-scale systems have been in operation worldwide (Hu et al., 2022). Substantial work has been done in AnMBR pilot testing on domestic wastewater, including minimizing energy input using inert abrasive particles such as granular activated carbon. However, this also contributes to energy consumption due to the need to fluidize these particles. Other challenges associated with AnMBR operation include long-term fouling, which increases the energy costs, reduces the membrane lifetime, and makes the system less efficient (Rattier et al., 2022). Indeed, large-scale AnMBR applications for municipal wastewater treatment still face several challenges, including membrane fouling as well as a low energy recovery efficiency and limited considerations of resource recovery. Another major barrier for full-scale application of AnMBRs in domestic wastewater treatment is the lack of direct nutrient removal capability. It is worth noting that, from a circular economy point of view, this could be also an advantage since the lack removal of nutrients can be an opportunity of recovering them.

1.3. Resource recovery with anaerobic treatment

The development of innovative technologies in wastewater treatment recently created the concept of bio-refinery in wastewater treatment plants, placing high-rate anaerobic technology in the highlight (McCarty et al., 2011). Focus of SAFE project is certainly sustainable water reuse practices, including the opportunity offered by anaerobic treated effluent. Indeed, the potential of high-rate anaerobic reactors is significant as these systems can remove a board range of trace organic contaminants relevant to water reuse, convert organics in wastewater to biogas for subsequent energy production, and liberate nutrients to soluble forms (e.g. ammonia and phosphorus) for subsequent recovery for fertilizer production (Song et al., 2018). It is worth adding that granular biomass high-rate anaerobic systems even provides a recoverable resource and, consequently a market value, to excess sludge, because granular sludge is nowadays sold on the market for re-inoculating or starting up new reactor systems (Van Lier et al., 2015).

The starting point of this prospect arises from the fact that during the last years, wastewater treatment plants have moved from the concept of 'waste treatment', aimed at discharging treated wastewater into surface waters, to the concept of 'water resource recovery facility'. This transformation from pollutants removal to valuable resources frames wastewater management in the broader context of the circular economy (Akyol et al., 2020).

The possible recovered and safely reusable resources from wastewater to help closing the loop of their treatment include, as just cited, energy, water and other materials, but reclaimed water is without doubt the most relevant for SAFE project objectives. Indeed, water reuse is particularly important because it is considered as an effective option to address water shortage problems and water quality deterioration issues, which are both focus of SAFE.

Potential water reuses involve the recycle in industrial processes, non-potable domestic use (e.g. toilet flushing), and groundwater replenishing and, of course, agricultural and landscape irrigation. In addition to this, the option deserved by anaerobic treated effluent of nutrient recovery and recycling for beneficial purpose in agriculture as soil amendments or fertilizers has been already mentioned. In particular, ammonia nitrogen is advantageous because it predominates in anaerobic reactor effluents and can be useful for fertigation purposes.

Among the mentioned high-rate technologies for anaerobic treatment of domestic wastewater, UASB systems are recognized as one of the best available technology so far being widely employed and accounting for most of the high-rate anaerobic bioreactors currently in use (Stazi and Tomei, 2018). In particular, UASB was successfully implemented and established within a wide acceptance in municipal wastewater treatment plants, especially in tropical and subtropical regions where the temperature of the wastewater is usually above 20 °C (Lohani et al., 2016). Nutrient recycling has been demonstrated for UASB technology and the nutrient recovery has been realized through and/or Ca-P precipitation.

It is noteworthy that, besides the sustainability assessment of the resource recovery approach, this is also economically feasible in terms of production costs and market values. However, despite the above-mentioned potential of high-rate anaerobic systems and notwithstanding the important research activities and the developed technologies for resource recovery from wastewater, there are some bottlenecks for the market uptake and for their application. Main challenges include the dilute nature and seasonal temperature range of municipal wastewater, salinity build-up when diluted wastewater is pre-concentrated, and inhibitory substances (e.g. free ammonia and sulphide) (Song et al., 2018). Moreover, other issues, including social-technological planning and design methodology to identify their potential end-use and market requirements, still limit the efficient implementation of these technologies applied for simultaneous wastewater treatment and resource recovery.

In particular, the Italian legislation for water reuse, at present, regulates only the irrigation purpose and limited specific indications and legislations were clearly promoted for fertigation objective as highlighted even by EU Innovation Deal on sustainable wastewater treatment (EC, 2017). Many lacks and heterogeneity among countries have been identified both in the legal definition of the term discharge and for quality standards provisions adopted for wastewater effluents to be used for agriculture. Moreover, recognition of the economic and environmental benefits of water reuse within reclaimed water pricing have to be implemented (Akyol et al., 2020). On the contrary, for phosphorous and ammonia salts, more detailed studies and programs have been developed at European level to overcome regulatory barriers. However, the quality, the purity and the characteristics of the recovered materials change based on the implemented treatment process, while the different market sectors request inlet materials with diverse standards on the basis of the final productive application.

For this reason the certification of the technologies, which also has to include the main properties of the recovered products, seems necessary to couple the recovery processes to the industrial sectors. In this context, the European criteria of 'End-of-waste' could be identified as possible legislative solution to support the resources recovery application in the wastewater treatment plants. In fact, the approach of Waste Framework Directive (EC, 2008) specifies when certain waste ceases to be waste and obtains a status of a product or a secondary raw material.

The achievement of the end of "waste" status has to be supported by the following several conditions:

- the substance or object is commonly used for specific purposes;
- there is an existing market or demand for the substance or object;
- the use is lawful (substance or object fulfills the technical requirements for the specific purposes and meets the existing legislation and standards applicable to products);
- the use will not lead to overall adverse environmental or human health impacts.

Starting from this point, specific regulations, centered on the end of waste concept, could be implemented to support the regulatory framework of the resources recovery. This approach can justify and encourage the technological investments in the wastewater treatment plants to economically address and support the resource recovery and to promote the circular economy in the water sector.

Finally, public perception and social acceptance are insufficiently developed for all the described materials. Therefore, specific formative and public dissemination activities have to be strongly supported.

2. Materials and methods

2.1. Implementation and testing of an UASB prototype at lab-scale for treating urban wastewater

In the reference framework of WP1 of SAFE project related to sustainable and nature-based solutions as applied innovative technologies for wastewater treatment, objective of IRSA-CNR activity is to investigate and to optimize the performance of a high-rate UASB bioreactor operated with granular biomass. Final goal is to confirm that this technology is one of the most powerful treatment for domestic wastewater with outstanding potentialities in terms of contaminant removal and energy and resource recovery. The proposed process can be optimized by managing different operating parameters such as wastewater temperature, HRT (Hydraulic Retention Time), SRT (Sludge Retention Time), recycle flowrate, OLR (Organic Loading Rate), etc.

The experimental activity was carried out at IRSA-CNR according to the work schedule planned for the first year, and consisted of design, installation and management of an UASB reactor operated with granular biomass in sequential mode i.e. as a sequencing batch reactor (SBR) to treat a synthetic domestic wastewater. The sequential mode was chosen based on the advantages of these systems over continuous ones, such as the relative ease of operation, better solids retention, absence of primary or secondary settling, system flexibility and stability, better quality control, and low operational costs.

A detailed description of the set-up of the experimental apparatus, a summary of the experimental plan and, finally, an overview of the achieved results are reported in the following sections.

2.2. Set-up of experimental apparatus

The UASB system consisted in a laboratory-scale jacketed glass reactor (whose dimensions are listed in Table 2) with a working volume of 0.9 L whose operation was managed by a dedicated computer connected to the bioreactor through an interface (Figure 1a) able to control automatically the timing of each operating stage and the feeding/discharging of the bioreactor. A specialized software developed under Labview-Windows XP environment managed the reactor-computer connection.

Each working cycle consisted of the following operating stages: feeding phase, reaction phase (operated with liquid recirculation from the top to the bottom of the reactor), sedimentation and effluent discharge. The time duration of each of these stages was 0.2, 3-10.34, 0.5, and 1.3, respectively. Moreover, it is worth highlighting that the reaction phase lasted differentially in accordance with the scheduled HRT values for the different experimental stages, as also specified in Table 2. For example, for a HRT value of 9 h, the corresponding reaction phase lasted 3.83 h.

Figure 1 Picture of the (a) interface and (b) biogas measurement device connected to the lab-scale UASB system implemented at IRSA-CNR.

Table 2 Operating conditions of UASB system

Parameter	*Unit*	*Value*
Total height	cm	77.5
Working volume height	cm	6.5
Internal diameter	cm	3.5
Internal section	cm^2	9.6
Working volume	L	0.9
Exchange ratio[a]	-	0.56
Up-flow velocity	m/h	8
Temperature	°C	25-35
HRT	h	9, 14, 22
Working cycle duration[b]	h	3.83, 7, 11.17
- Feeding phase	h	0.2
- Reaction phase[b]	h	3, 6.17, 10.34
- Settling phase	h	0.5
- Effluent discharge phase	h	0.13

[a] Exchange ratio = Fed volume/working volume
[b] different values are related to different tested HRTs

The equipment of the bioreactor included the following devices:

- 3 tanks (5 L capacity) for the storage of the feed solution and the treated effluent and one for safety overfilling tank;
- 3 peristaltic pumps (interfaced to computer) to manage the working sequence of feeding and discharge and for internal liquid recirculation;
- 2 gas bags (0.5 L each one) to equalize the pressure and to maintain anaerobic conditions during the operating phases;
- a thermo-cryostat connected to the reactor jacket to control the temperature;
- a wet-gas meter (Figure 1b) connected in the place of gas bags during the reaction phase to measure the amount of produced biogas

Figure 2 shows a schematic representation and a picture of lab-scale system implemented at IRSA-CNR, while all the operating conditions applied during the experiments are summarized in Table 2.

2.3. Operation of UASB prototype

Once UASB installation was completed, preliminary experimental activity was devoted to verify the effluent recycle flow-rate able to maintain the up-flow velocity at the pre-fixed level and to prove the system feasibility regarding to the uptake and quantification of the produced biogas. Regarding the up-flow velocity, the value of 8 m/h has been selected, after preliminary hydraulic experiments, to guarantee an effective expansion of the sludge bed, by ensuring the effective contact between granular biomass and wastewater, and by avoiding simultaneously the biomass dragging in the effluent. Different up-flow velocities have been tested within the range of 3.5-10 m/h before identifying the optimal value chosen for the subsequent experimental campaigns. That rage of values has been set according the literature (Abbasi et al., 2012) by considering that up-flow velocities up to 10 m/h can be applied with granular biomass to achieve an effective expansion of the sludge bed.

Therefore, the bioreactor was inoculated with a microbial consortium of granular biomass coming from a wastewater treatment plant of brewery industry (inoculum volume 0.27 L, volatile solids 0.25 g_{VSS}/L) and fed with a daily prepared synthetic wastewater whose composition has been selected based on a previous literature review. The composition of the synthetic urban wastewater is constituted by peptone, soluble starch, sodium acetate, yeast extract, ammonium chloride, urea, and potassium dihydrogenphosphate, according to the proportions suggested by Kayranly and Ugurlu (2011) and listed in Table 3. Sodium bicarbonate was added to the solution in order to maintain suitable pH buffering and other mineral species to ensure the presence of microelements for microorganisms growth (See Table 2). Prepared solutions were daily analysed for COD, total nitrogen (N_{TOT}), ammonia nitrogen ($N\text{-}NH_3$) and total phosphorus (P_{TOT}): an average characterization of the influent solution was reported in Table 3.

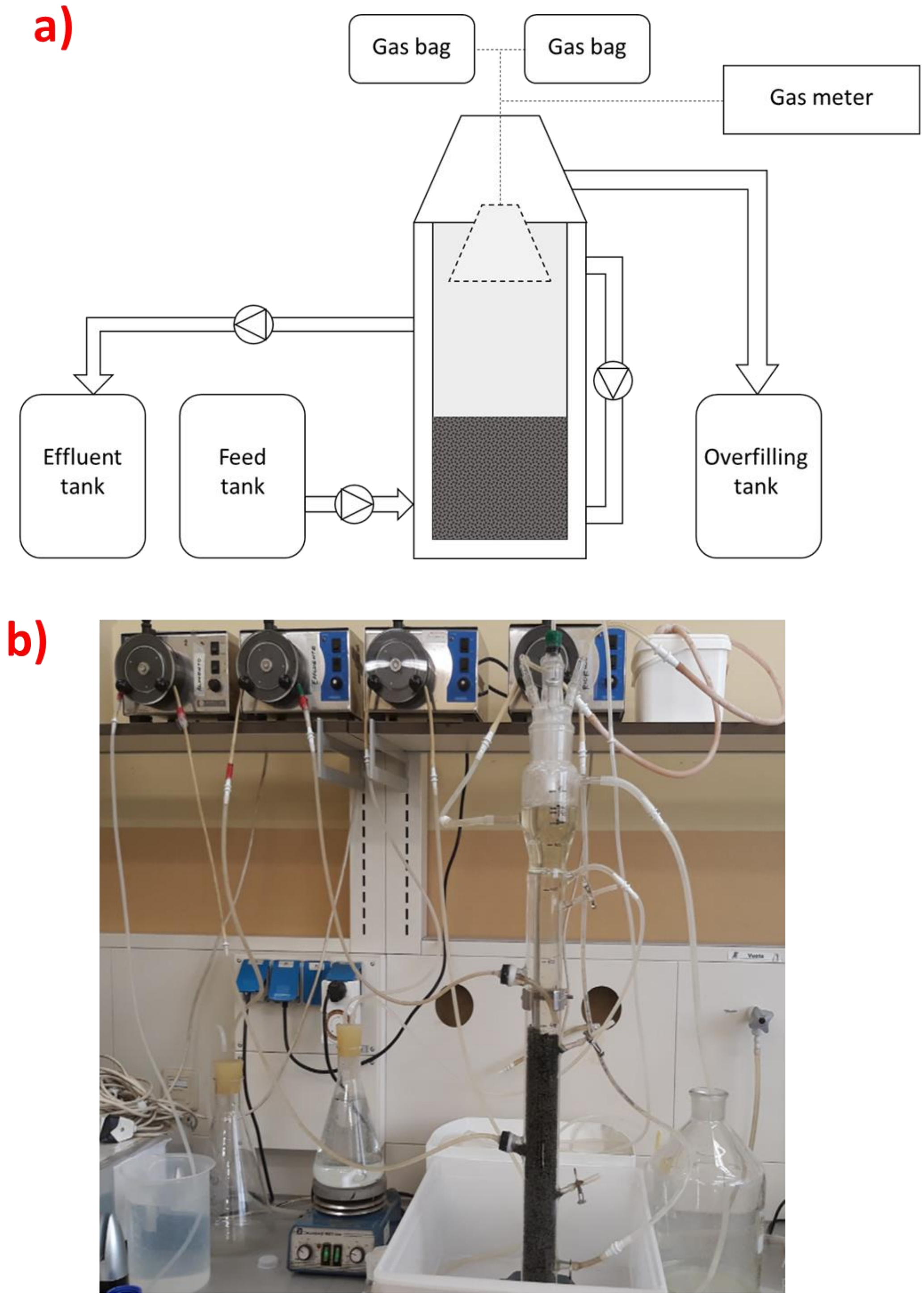

Figure 2 Schematic representation (a) and picture (b) of lab-scale UASB system implemented at IRSA-CNR and used during the experimentation.

Two experimental campaigns at different temperatures have been carried out at values of 35 ± 1 and afterwards 25 ± 1 °C, according to the experimental roadmap reported in Figure 2. For the start-up of the bioreactor, there was a short acclimatization period of around 2 weeks at HRT of 43 h corresponding to one working cycle/d, with a reaction phase of around 23 h to reach near steady-state performance.

Afterwards, UASB system was run with different conditions of HRT (see Figure 3), whose values have been controlled by the length of the reaction phase. For example, tested HRTs were 22 and 14 and 9 h and they achieved by varying the time duration of the reaction phase (and the number of cycles/d as a consequence) from 670 (2 cycles/d), to 420 (3 cycles/d) and then to 230 minutes (5 cycles/d), respectively.

Table 3 Composition and characterization of tested municipal wastewater

Parameter	***Unit***	***Value***
pH	-	8-8.6
Influent COD	mg_{COD}/L	436-564
N_{TOT}	mg/L	50-56
$N-NH_3$	mg/L	9.5
P	mg/L	5
Peptone from casein[a]	mg/L	260
Starch	mg/L	130
Sodium acetate	mg/L	181
Yeast extract	mg/L	40
Ammonium chloride	mg/L	36.2
Urea	mg/L	21.4
Potassium dihydrogenphosphate	mg/L	22
$NaHCO_3$	mg/L	1000
$CaCl_2 \cdot 2\ H_2O$	mg/L	47.5
$MgSO_4 \cdot 7\ H_2O$	mg/L	52.5
$FeSO_4 \cdot 7\ H_2O$	mg/L	40

[a] instead of milk powder, with respect to Kayranly and Ugurlu (2011)

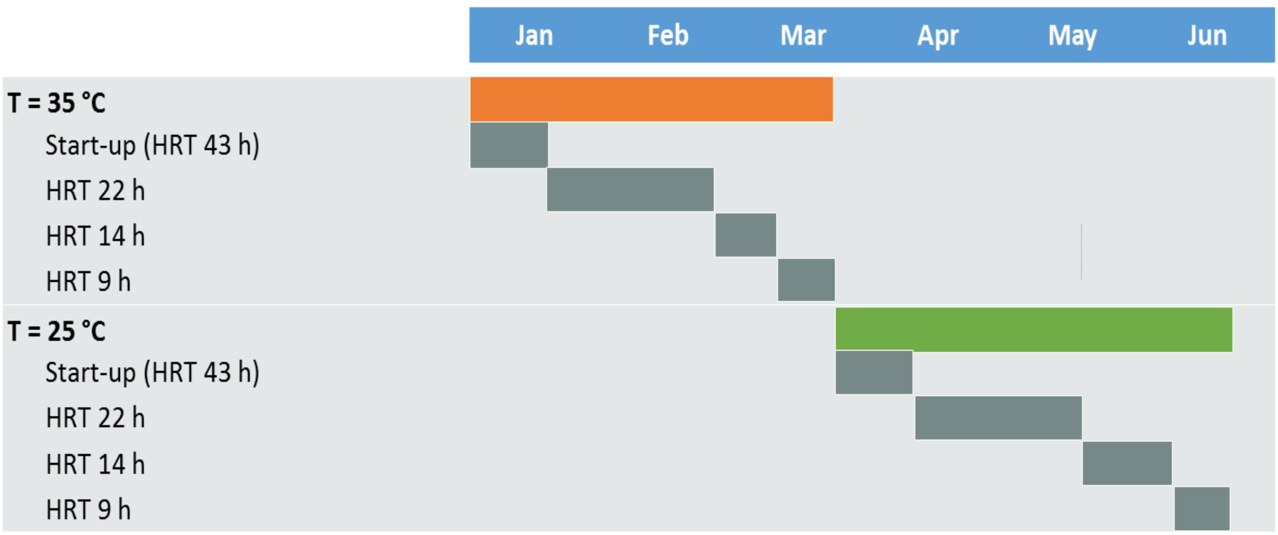

Figure 3 Time plan of operation of UASB system. The number inside the circles indicate the time duration of each experimental phase.

2.4. Process performance evaluation of UASB prototype

Process performance of the UASB was evaluated as follows: treated effluent was periodically monitored at interval times of 2–3 days according to Standard Methods (APHA, 2012) with analysis that included pH, COD, volatile fatty acids (VFAs), N_{TOT}, N-NH_3, P_{TOT}, TSS and VSS in the treated effluent.

A pH-meter (pH700, EUTECH Instruments) was employed for pH measurements of the synthetic WW and of the reactor effluent. Cell tests (Merck) and spectrophotometric determination (Spectroquant Nova30) were used for COD, N_{TOT}, and P_{TOT} measurements. Analysis of VFAs were performed by using a Perkin-Elmer Auto System gas chromatograph according to the procedure described by Cruz Viggi et al. (2017). N-NH_3 concentration was determined spectrophotometrically, by adding Seignette salt and Nessler reagent producing yellow colored complex whose absorbance was measured at a wavelength of 420 nm.

Furthermore, the volume and composition of the produced biogas was analyzed periodically. The amount of biogas was measured using a wet-gas meter, as previously mentioned, while the methane fraction in the biogas was determined by gas chromatography (Perkin Elmer AutoSystem equipped with a Carboxen 1000 (Supelco) column and a TCD detector).

3. Results

3.1. Progress status and achieved results

A summary of general performance of the UASB system is listed in Table 4 and displayed in Figure 4. Generally, the reactor exhibited excellent removal efficiencies of the organic matter for all tested HRTs for both temperatures, confirming the optimal performance of this technology.

In particular, at 25 °C, COD removals of 84-91%, 90-92 and 85-92% and biogas production values of 0.14-0.27, 0.16-0.17 and 0.15-0.25 $m^3/kg_{CODremoved}$ were observed for 22, 14 and 9 h HRT, respectively. Regarding experimental results obtained at mesophilic conditions, outstanding performance have been also detected with COD removals of 93-94, 89-94 and 89-91% and biogas production of 0.22-0.25, 0.21-0.23 and 0.14-0.21 $m^3/kg_{CODremoved}$, for 22, 14 and 9 h HRT, respectively. Data obtained for both tested temperatures confirmed that treatment efficiencies showed no substantial differences for different HRTs; hence, the lowest one, i.e. 9 h, is the most advantageous being of the same order of values of aerobic processes.

Table 4 Long-term experimental results referring to the entire experimental periods

Temperature	HRT	COD removal	Biogas production
(°C)	**(h)**	**(%)**	**$m^3/kg_{CODremoved}$**
25	22	84-91	0.14-0.27
	14	90-92	0.16-0.17
	9	85-92	0.15-0.25
35	22	93-94	0.22-0.25
	14	89-94	0.21-0.23
	9	89-91	0.14-0.21

Concerning the biogas production, it is observed that, contrary to expectations, lower organic matter removal does not imply lower biogas production. This phenomenon can be due to the entrapment of gas bubbles inside the granular biomass, resulting in an imperfect theoretical correlation between COD removal and biogas yield. The entrapment of gas bubbles is a phenomenon commonly detected in anaerobic granular bioreactors, even at steady state conditions, which can be caused by several hydrodynamic and biological factors such as granule aggregation, increase of granule size due to biomass growth, preferential gas flowing paths in the granular bed. It is worth noting that a commonly reported problem related to UASB treatment of municipal sewage, especially under suboptimal conditions, is the limited internal mixing due to low biogas production, a major contributing factor to effective mass transfer, resulting in hindered liquid–biomass contact (Cecconet et al., 2022). This phenomenon could be overcome by increasing the recirculation rate, within the limits of the values causing granules disintegration, thus allowing a more effective fluidization of the sludge bed.

Regarding the quality of treated effluent, data are shown in Figure 4 with similar values in terms of COD, VFAs, TSS, $N\text{-}NH_3$ and $P\text{-}PO_4$ at all HRT tested conditions and for both temperatures. Satisfactory results have been obtained in terms of COD, TSS and VFAs; in particular, both COD and TSS complied Italian regulation for discharge standards in receiving water bodies (Decree Italian Law 152/2006). On the contrary, nitrogen and phosphorus concentrations, even if non-limiting for anaerobic biomass, exceeded the effluent standards, but without any negative effect over the system stability. It is worth noting that high effluent concentrations of N and P lead to eutrophication problems: consequently, these elements should be removed or, alternatively, recovered for their reuse/recycle (Meesschaert et al., 2021).

Indeed, the best option for these nutrients is their recovery for fertilizing purposes and the actual quality of these effluents, free from solids and rich in nutrients, makes it potentially suitable for agricultural reuse, or for nutrients recovery. According to the SAFE prospect, this option is completely included in the paradigm shift that is currently underway from an attitude that considers wastewater streams as a waste to be treated, to a proactive interest in recovering materials and energy from these streams.

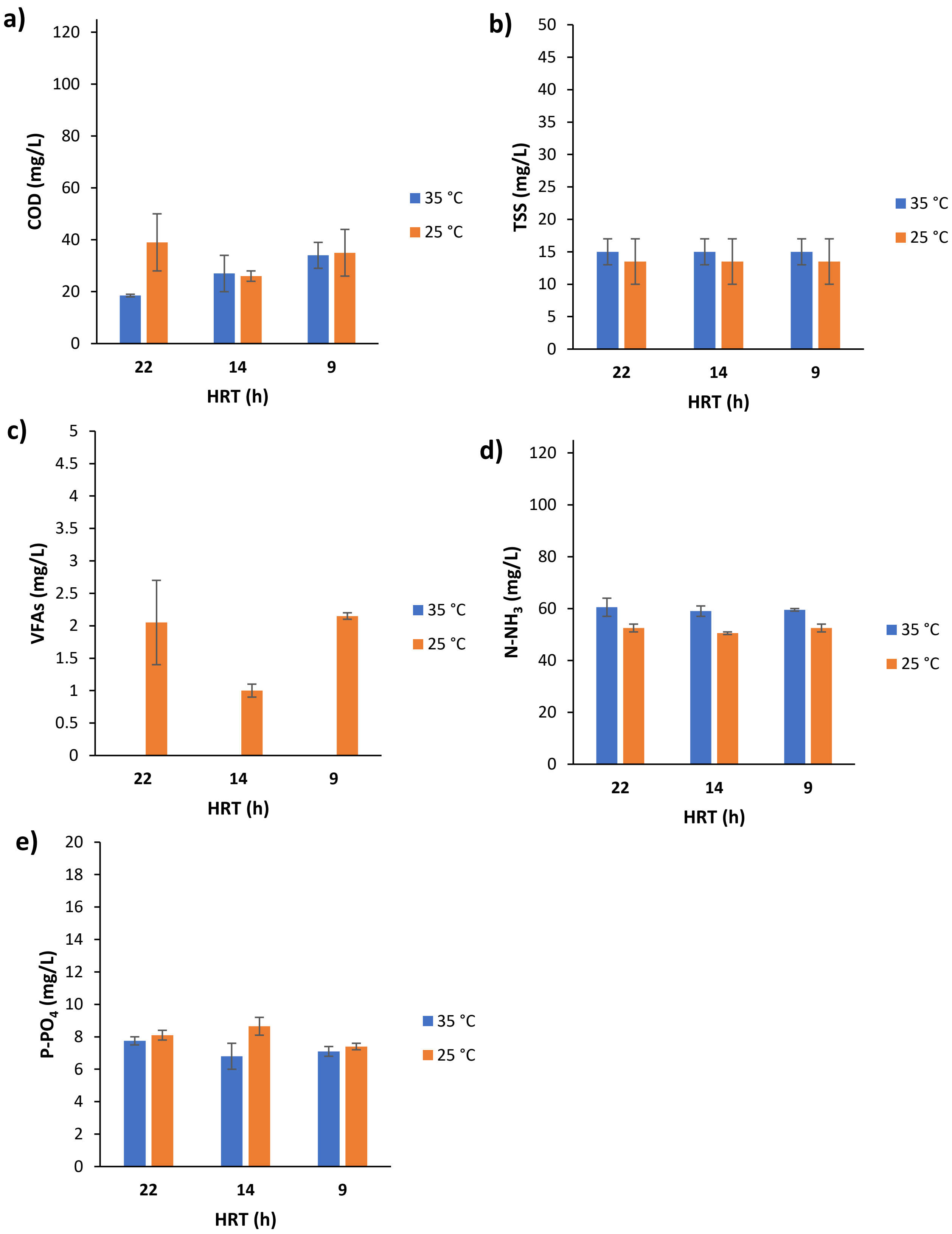

Figure 4 Experimental results of UASB system for 25 and 35 °C. Data are reported as average value referring to the entire experimental periods and the bars are standard deviations.

4. Conclusions

Long-term experimental results of the first laboratory campaigns performed at 25 and 35 °C confirmed the optimal performance of UASB technology applied to the treatment of a synthetic domestic wastewater, by pointing out the following findings:

- good COD removal efficiencies (84-94%)
- good biogas production (0.14-0.27 $m^3/kg_{CODremoved}$)
- no accumulation of VFA in the bioreactor, indicating the good stability of the anaerobic system
- good effluent quality in terms of COD (< 50 mg/L) and suspended solids (< 20 mg/L)
- high nitrogen and phosphorus concentrations in the treated effluent suggesting their recovery in agriculture.

These preliminary results are a first step in the evaluation of the feasibility of an UASB as an energy generating process and cost-effective alternative for wastewater treatment which is also able to produce nutrient rich and solids free effluents with a high degree of pathogen removal, while occupying a small footprint.

A final positive remark of obtained results is related to the sequential mode of operation of the installed system: it is worth noting that the relevance of obtained data is high for practical application because SBRs, which are characterized by high flexibility given by modularity and low footprint, can be proposed for domestic wastewater treatment plants characterized by a marked variability of the influent load i.e. seasonal variation in touristic areas.

Ongoing research efforts are focused on the investigation of the temperature effect, in particular testing of psychrophilic conditions has been scheduled with the main objective of demonstrating that UASB is technically feasible for municipal wastewater treatment, as related to biogas production and organic matter removal, also for temperatures below the optimal range for the process.

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