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Sieving of municipal wastewater and recovery of bio-based volatile fatty acids at pilot scale

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ABSTRACT

This study combined at pilot scale the recovery of cellulosic primary sludge from the sieving of municipal wastewater followed by the production of bio-based VFAs through acidogenic fermentation. The sieving of municipal wastewater was accomplished by a rotating belt filter which allowed the removal of around 50% of suspended solids when operated at solids loading rates higher than 30–35 kgTSS/m^2 h. The solids recovered by sieving contained around 40% of cellulose, which is a suitable raw material for the production of bio-based VFAs. Initially, fermentation batch tests of cellulosic primary sludge were carried out adjusting the initial pH of the sludge at values of 8, 9, 10 and 11, in order to evaluate the best production yields of bio-based VFAs and their composition. The highest VFAs yield achieved was 521 mgCOD$_{VFA}$/gVS occurring when pH was adjusted at 9, while propionic acid reached 51% of the total VFAs. Then, the optimal conditions were applied at long term in a sequencing batch fermentation reactor where the highest potential productivity of bio-based VFAs (2.57 kg COD/m^3 d) was obtained by adjusting the pH feeding at 9 and operating with an hydraulic retention time of 6 days under mesophilic conditions. The cost-benefit analyses for the implementation of cellulosic primary sludge recovery was carried out consideringthe anaerobic digestion as reference scenario. The economical assessment showed that the production of bio-based VFAs from cellulosic primary sludge as carbon source and/or as chemical precursors give higher net benefits instead of the only biogas production.

1. Introduction

Since more than 100 years, activated sludge process has been considered an effective technology to remove oxygen demanding compounds (COD) and nutrients (N & P) from wastewater, despite the high electrical energy consumption and generation of greenhouse gas emissions (GHGs) which contribute to the climate change (Maktabifard et al., 2018; Kampschreur et al., 2009). Currently, the role of wastewater treatment plants (WWPTs) is no longer merely evaluated based on the effluent quality, materials to recover energy and resources embedded in wastewater to improve the sustainability of the water sector towards circular economy (van der Hoek et al., 2016; Wang et al., 2015). Water is the first type of resource to recover for multiple uses, while the wastewater organics can be upgraded to reusable materials such as cellulose, methane or biopolymers (Resource Recovery Cluster, 2015). The spreading of this approach contributes to rebrand WWTPs into Water Resource Recovery Facilities (WRRFs) and to develop new processes combining existing systems with innovative technologies. Nevertheless, the use of concentrated streams has been reported as a successful strategy for resource recovery by Verstraete et al. (2009), thus making raw wastewater not suitable for this purpose due to its dilution rate.

The rotating belt filter (RBF) is a primary treatment aiming the mechanical removal and concentration of larger suspended solid by cake filtration and sieving of the wastewater. Several studies demonstrated the benefits of this system compared with conventional primary clarifiers like lower footprint, reduction of the overall organic loading rate in the biological reactor mainly due to slowly biodegradable COD which contributes to the mitigation of CO$_2$-eq emission by 36% (Paulsrud et al., 2014; Franchi and Santoro, 2015). In addition, the sludge produced using the RBF is enriched in toilet paper fibers which enables the recovery of marketable cellulosic fiber from cellulosic primary sludge (CPS) as demonstrated by CirTec which

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implemented Cellvation™ technology in Geestmerambacht WWTP (Akyol et al., 2019). The final product, called ReCell®, is a valuable raw material suitable for different industrial sectors like construction, paper and board, coatings and sustainable chemicals. The recovered cellulose is already used as asphalt additive or could replaces pure cellulose in hydraulic lime-based mortars (Palmieri et al., 2019). Moreover Zhou et al. (2019) developed a compatible sludge cellulose plastic composite including recovered CPS and demonstrated that the produced biocomposite had 15% lower manufacturing costs than conventional one, and it was 5% more eco-efficient than wood plastic composite.

Despite the anaerobic digestion of CPS gains 20% higher biogas production compared with conventional primary sludge (Ghasimi et al., 2016), the overall revenues are limited by the low value of the methane in market. The bio-based volatile fatty acids (VFAs) represent a higher added-value product because of their economic price and wide range of application, indeed, the VFAs are a suitable carbon source for Enhanced Biological Phosphorus Removal (EBPR) process or can be used as chemical precursors for a number of applications (Oehmen et al., 2007; Lee et al., 2014; Longo et al., 2017; Bhatia and Yang, 2017). The great number of studies dealing with acidogenic fermentation of sewage sludge during the last decade (Atasoy et al., 2018) showed the increase of interest for bio-based VFAs production within WRRF and the feasibility to valorize the organic matter through this bioprocess. Crutchik et al. (2018) investigated the effect of pH and temperature on VFAs production from a simulated CPS constituted by a mixture of primary sludge and toilet paper. The highest bio-based VFAs yield was obtained with an initial pH of 8 under mesophilic conditions (254 $mgCOD_{VFA}/gVSS$). The VFA concentration in the liquid fraction 14–18 gCOD/L and the dominant species were acetic and propionic acid. Consequently to these findings, the examination of the combination of RBF and acidogenic fermentation to evaluate its potential exploitation.

This study investigated at pilot scale and under real environmental conditions the integration of RBF with the acidogenic fermentation of CPS for the conversion of slowly biodegradable COD into bio-based VFAs as chemical precursors. For this purpose, the VFAs productivities were evaluated in a pilot sequencing batch fermentation reactor (SBFR) under different retention times and pH according with preliminary batch test. Finally, the observed performances were employed to analyze and compare three pathways of CPS: i) production of methane through anaerobic digestion, ii) production of bio-based VFAs as supplementary carbon source for BNR process iii) production of bio-based VFAs as chemical precursors for polyhydroxyalkanoates (PHA) recovery.

2. Materials and methods

2.1. Wastewater characterization

Municipal wastewater was characterized in terms of pH, total alkalinity, total and volatile suspended solid, total and soluble chemical oxygen demand (COD), total Kijendhal and ammonium nitrogen, total phosphorus and phosphate according to of Standard Methods (American Public Health Association et al., 1999). Granulometric distribution was evaluated on 24-hours composite samples using sieving on meshes with porosity of 350, 210, 158, 90 and 54 μm as described by Rusten and Lundar (2006).

2.2. Wastewater sieving by pilot rotating belt filter

The cellulosic primary sludge was produced by the sieving of around 400 m^3/day of municipal wastewater from Carbonera WWTP (Treviso, North of Italy) collected after a coarse bar screening (3 mm) and grit removal of the main WWTP line. The wastewater sieving was accomplished through a rotating belt filter type SF1000 (Salsnes Filter SA, Norway) with a flow capacity up to 15 l/s and a submerged sieve cloth area of 0.24 m^2. The experiments have been carried out using two mesh sizes (350 and 210 μm) in order to evaluate how this parameter affects the suspended solids removal efficiency. Different flow rates were applied in the range between 8 and 11 l/s while higher values determined overflows of the wastewater not providing reliable performances. The belt rotation speed was controlled by a PID model control in order to maintain a constant liquid level (210 mm), while the inclination of the belt was kept constant at 30°. A sludge layer formed on the mesh surface which consequently retained smaller particles than the mesh pore size of the belt, improving the TSS removal efficiency. The RBF was embedded with a cleaning system made up of an air knife and compressed hot water to remove the residual sludge cake from the belt. The CPS produced was discontinuously fed in a Sequencing Batch Fermentation Reactor through a volumetric pump (described in section 2.4), while the sieved wastewater was discharged and recirculated in the headworks of the WWTP. Two autosamplers collected the influent and effluent at regular intervals (every 5 min) to obtain representative samples for their characterization and calculation of the mass balances.

2.3. Batch fermentation tests

Around 10 L of CPS was collected from RBF and investigated through batch fermentation tests in duplicate glass bottles with working and total volume of 0.7 and 1.2 L, respectively. The bottles were placed in a temperature-controlled air chamber at 37 °C. Mesophilic condition was chosen due to the positive results reported by Crutchik et al. (2018) indeed thermophilic or hyperthermophilic temperature had a detrimental effect on total bio-based VFAs production. However, differently from the authors, in this work real CPS was evaluated under batch conditions. The role of initial pH was investigated using raw CPS (pH around 6.6) and changing the pH of substrate by adding NaOH (30% solution) to obtain an initial pH of 8, 9, 10 and 11. The pH of the sludge was not controlled during the fermentation and mixing was operated manually twice per day. The process was monitored for two weeks collecting around 50 ml of sludge per day. Then, the samples were centrifuged, and the liquid fraction was characterized in terms of pH, soluble COD, bio-based VFAs concentration and composition, ammonium and phosphate content. The performances at different pH were quantified as maximal bio-based VFAs yields (Y_{VFA}) that corresponds to ratio between produced bio-based VFAs and fed VS (Equation (1)).

$$\text{VFA yield}\left(\frac{\text{mgCOD}}{\text{gVS}}\right)=\frac{COD_{VFA\ max}-COD_{VFA\ t0}}{VS_{fed}} \tag{1}$$

Where $COD_{VFA\ t0}$ and $COD_{VFA\ max}$ were respectively the initial VFAs concentration and the concentration at the time where the highest value was observed, while VS_{fed} was the concentration of volatile solid of the CPS.

2.4. Sequencing batch fermentation reactor

The acidogenic fermentation of the CPS was carried out in a pilot-scale sequencing batch fermentation reactor (SBFR) of 2.6 m^3 of working volume. The temperature of the reactor was maintained at 37 °C using an electrical heating system located on the internal wall of the reactor. The reactor was initially filled with CPS and the feeding of the reactor was accomplished during 5 days per week by discharging a volume of fermented CPS from the reactor and replacing the same amount with CPS directly pumped from the RBF. The volumetric exchange ratio was fixed at 25% or 10% corresponding to an average HRT of 6 and 14 days, respectively (Table 1). The organic loading rate was affected by the flow rate and average inlet TS concentration as reported in Table 1. Each operational condition was tested for at least 3 HRTs and under steady state conditions.

The reactor was mixed using a grinder pump installed at the bottom of the tank aiming the reduction of the particle sizes and continuous recycling the treated sludge at the top of the reactor.

The effluent from the SBFR was collected and analyzed three times per week to evaluate the fermentation performances. The raw samples were used to determine the total and volatile solids content, while an amount of sample was centrifuged to characterize the liquid fraction. Soluble COD and bio-based VFAs were analyzed to calculate the bio-based VFAs yields and the composition of soluble organic matter. The acidification degree was determined by dividing the concentration of bio-based VFAs (Huang et al., 2018) by the concentration of soluble COD at the same retention time (Equation (2)).

$$\text{Acidification degree } (\%) = \frac{COD_{VFA}}{\text{Soluble COD}} \tag{2}$$

Ammonium and phosphate concentration in the liquid fraction (NH_4-N_{LF} and PO_4-P_{LF}) were measured to estimate the nutrient release and their potential recovery. Phosphorus and ammonium releases was calculated according to Equations (3) and (4), where TKN_{CPS} and TP_{CPS} represents the total content in the fed sludge.

$$NH_4 - N \text{ release}(\%) = \frac{NH_4 - N_{LF}}{TKN_{CPS}} \tag{3}$$

$$PO_4 - P \text{ release}(\%) = \frac{PO_4 - P_{LF}}{TP_{CPS}} \tag{4}$$

The significance of average performances reached under different operational conditions was analyzed using one-way ANOVA and Tukey post-hoc test with a significance threshold level of 5% ($p < 0.05$) The statistical analysis was conducted using Microsofts Excel/XLSTAT©-Pro (Version 7.2, 2003, USA).

3. Results and discussion

3.1. Wastewater characteristics

Composite discrete samples were collected and analyzed in order to represents the average characteristics of wastewater over the experimental periods. The average characteristics (Table S1) of the influent wastewater in Carbonera WWTP during the experimental period were: 197 mg TSS/l, 391 mg COD/l, 38 mg TKN/l with 82% due to ammonium, 5.8 mgP/l (67% phosphate). The COD/N ratio was 10.3 gCOD/gN, while the content of cellulose accounted for 18% lower than the percentage reported by other studies (35% TSS, Ruiken et al., 2013).

The particulate distribution had the typical profile showed by Rusten and Lundar (2006) where most of the particulate material was either larger than 350 μm or smaller than 54 μm (Fig. 1). Ravndal et al. (2018) showed that the larger fraction is generally composed by carbohydrates and slowly biodegradable organic matter while the fraction with dimension <54 μm has a higher content of proteins and fat materials. The effect of the mesh porosity on the TSS and COD removal efficiency using static sieve was reported in supplementary material (Fig. 1S).

Table 1
Mean operational conditions applied to SBFR in runs 1 - 3.

Parameter	Unit	RUN1	RUN2	RUN3
HRT	days	6	14	6
OLR	kgVS/m^3 d	17.7	5.7	7.5
NLR	kgN/m^{3}d	0.35	0.11	0.15
PLR	kgP/m^{3}d	0.07	0.02	0.03
pH during feeding	–	6.4	6.4	9.0

Council Directive 91/271/EEC defines that a primary treatment must be able to reduce 20% of BOD_5 and to remove 50% of suspended solids. The preliminary results showed that the required TSS removal efficiency can be met using a 54 μm opening mesh if a cake is not formed during filtration. Regarding the requirement for BOD_5 removal (equivalent to 25% removal of COD, Rusten and Lundar., 2006), it could be easier achieved using a 210 μm mesh. On the other hand, Rusten and Lundar (2006) reported that if the percentage of TSS with dimension larger than mesh porosity is higher than 20% the cake formation occurs and leads to an increasing efficiency corresponding to a mesh with smaller porosity. Basing on base of wastewater characteristics, a mesh with opening dimension of 350 μm can be used and the cake formation is expected.

Previous studies reported that particulate fraction (>0.65 μm) had higher COD/N and COD/P ratio than colloidal, polymeric, oligomeric and monomeric fraction because of the abundance of macromolecules without nitrogen or phosphorous atoms (e.g. cellulose, starch and lignin) in the large particulate fractions (Ravndal et al., 2018). In particular, a former study demonstrated that fraction with size >100 μm had a content of carbohydrates ranging between 70 and 100% of total COD due to toilet paper residues (Ruiken et al., 2013). The biodegradability of the larger solid fraction is limited by particle breakup (Dimock and Morgenroth, 2006). Indeed, the surface area to volume ratio of particles was lower than in smaller fraction, hence substrate availability is proportionally inverse to the particle size. This explanation was confirmed by Ravndal et al. (2018) that observed that the largest particle fractions of wastewater was characterized by slowly biodegradable COD.

3.2. RBF performances

The RBF operated with a belt mesh size of 350 and 210 μm under two different periods meshes while the flow rates varied between 8 and 11 l/s.

The RBF removed approx. 44% of suspended solids (range 17–76%) and 35% of COD (range 10–61%). The nutrients removal efficiencies were variable and mainly affected by the characteristics of the wastewater. For instance, a large variation of the organic nitrogen removal was observed (between 4% and 36%), while phosphorus removal ranged between 4 and 6%. The COD/N reduced to 7.8 gCOD/gN but the removed COD was constituted by slowly biodegradable fraction, therefore the effect on biological treatment may be negligible as reported by Razafimanantsoa et al. (2014) that observed similar denitrification rates using raw and sieved wastewater.

The suspended solids removal efficiency increased of 5% by means of mesh size reduction from 350 to 210 μm (Fig. 2a), although the observed difference is not significant if standard deviations are taken into account: when the mesh with dimension of 350 μm was employed the solid removal efficiency was 37 ± 15%, while with the other mesh the value was 42 ± 11%. The high standard deviation indicates that the values are spread out over a wider range and the set of data obtained using different meshes cannot be distinguished. The variation of the efficiencies values is correlated more with the suspended solid concentrations influent

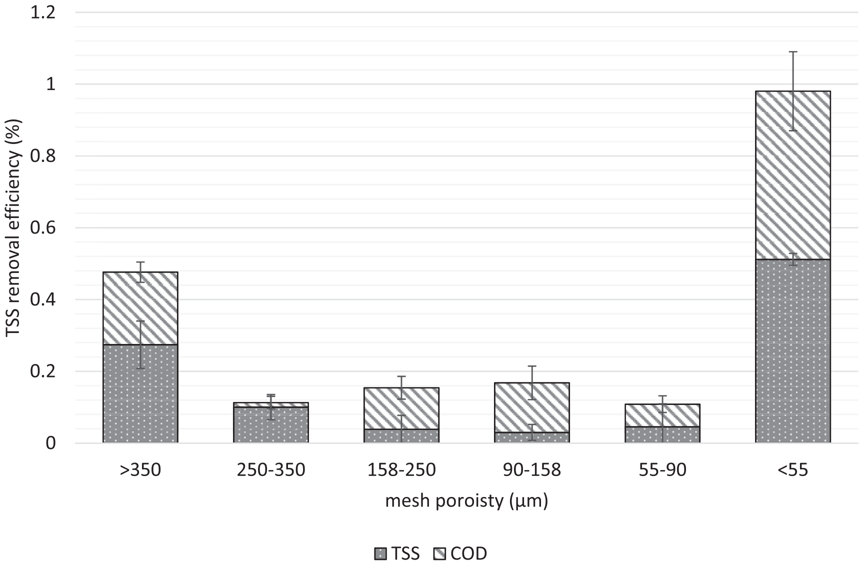

Fig. 1. Particulate size distribution of the suspended solids recovered by wastewater sieving.

rather than the change of the mesh size, which is the same conclusion reported by Rusten and Lundar (2006) from bench-scale tests.

The same authors reported that TSS removal efficiency is mainly affected by hydraulic flow although in this work the relation between TSS removal efficiency and the solid loading rate was more informative due to the high fluctuation of the wastewater characteristics (Fig. 2b). The threshold of 50% TSS removal efficiency can be achieved operating with a TSS loading rate higher than 30–35 kgTSS/m^2 h, which represent a footprint more the ten times lower compared with the conventional primary clarifier (Metcalf and Eddy, 2014).

Finally, the results of this study fitted with the model developed by Behera et al. (2018) (Fig. 2a) which explained that higher suspended solids concentration fosters the rate of cake formation over the filter mesh and consequently the removal of suspended solid with lower size.

3.3. *Production of cellulosic primary sludge*

The characteristics of the cellulosic primary sludge are summarized in Table S2 (supplementary material). The suspended solids concentration was the most variable parameter ranging between 21 and 73 g/kg. Generally, it was observed that higher TSS concentration in the wastewater affected positively the thickness of the recovered CPS. On the other hand, frequent overflow, due to high solid loading rate or mesh clogging, could reduce the solid content because of the fast belt rotation speed which does not allow the thickening of the sludge cake. The TS concentration of the CPS resulted from the sieving was quite constant during the experimental period (Table S2, supplementary material) with 92% as volatile fraction, which was significantly higher compared with the typical range 62–82% of conventional primary sludge from primary settling and 59–88% in secondary sludge (Liu et al., 2016; Miron et al., 2000; Wu et al., 2010; Metcalf).

The total solids showed a relatively high content in terms of COD (1501 mgO$_2$/gTS) and low content of TKN (from 16 to 22 mgN/gTS) and phosphorus (1.7–4.7 mgP/gTS) which were in the same range of the results reported in Ghasimi et al. (2015). The low presence of nutrients reflected by the COD/N and COD/P (55 mgCOD/mgN and 266 mgCOD/mgP respectively) which confirmed that the CPS was constituted by largest particulates like cellulose, hemicellulose and lignin more than protein-based compounds. A further investigation on CPS confirmed the high content of cellulose (41% TS) and lignin (18% TS), while lipids and proteins accounted for 9% and 13% of TS, respectively. The CPS had a protein content lower than primary conventional sludge (17–28% TS), while the lipids content was comparable (2–9% TS) (Miron et al., 2000; Thangamani et al., 2010; Wu et al., 2010).

3.4. *Evaluation of the potential bio-based VFAs production*

The fermentation batch tests were carried out adjusting the pH of the CPS to values between 8 and 11 and evaluating the effect of initial alkaline pH. The pH profiles during the batch (Fig. S2, supplementary material) followed the bio-based production which affected the activity of different microorganism groups operating the degradation of the organic matter. The fermentation without pH adjustment determined a quick drop of pH which stabilized to 4.9 (from day 1) and until the end of the batch. Tests operated with initial pH of 8–9 were still characterized by acidic values after one day of operation. Finally, when the batch was accomplished at initial pH 10, the highest alkalinity allowed to maintain neutral pH for a week which were sub optimal conditions for hydrolytic activities (Jain et al., 2015). On the other hand, methanogenic bacteria are favored by sub-alkaline pH but a weak growth could occur under acidic conditions leading the consumption of bio-based VFAS production (Feng et al., 2009). The profile of the VFAS concentration reflected the pH trends and showed that pH mainly affected the fermentation efficiencies. The bio-based VFAs concentrations were comparable until the second day in all the tests since the differences were not significant and the small variations in bio-based VFAs content could be due to the sludge heterogeneity. During days 3 and 4, the concentration of VFAs gradually increased until the maximum value with exception of the test accomplished at initial pH 11 that reached highest concentration after day 9 (Fig. 3a). As soon as the highest concentrations were achieved, a reduction of the VFAs was observed, which is associated with an increase of the

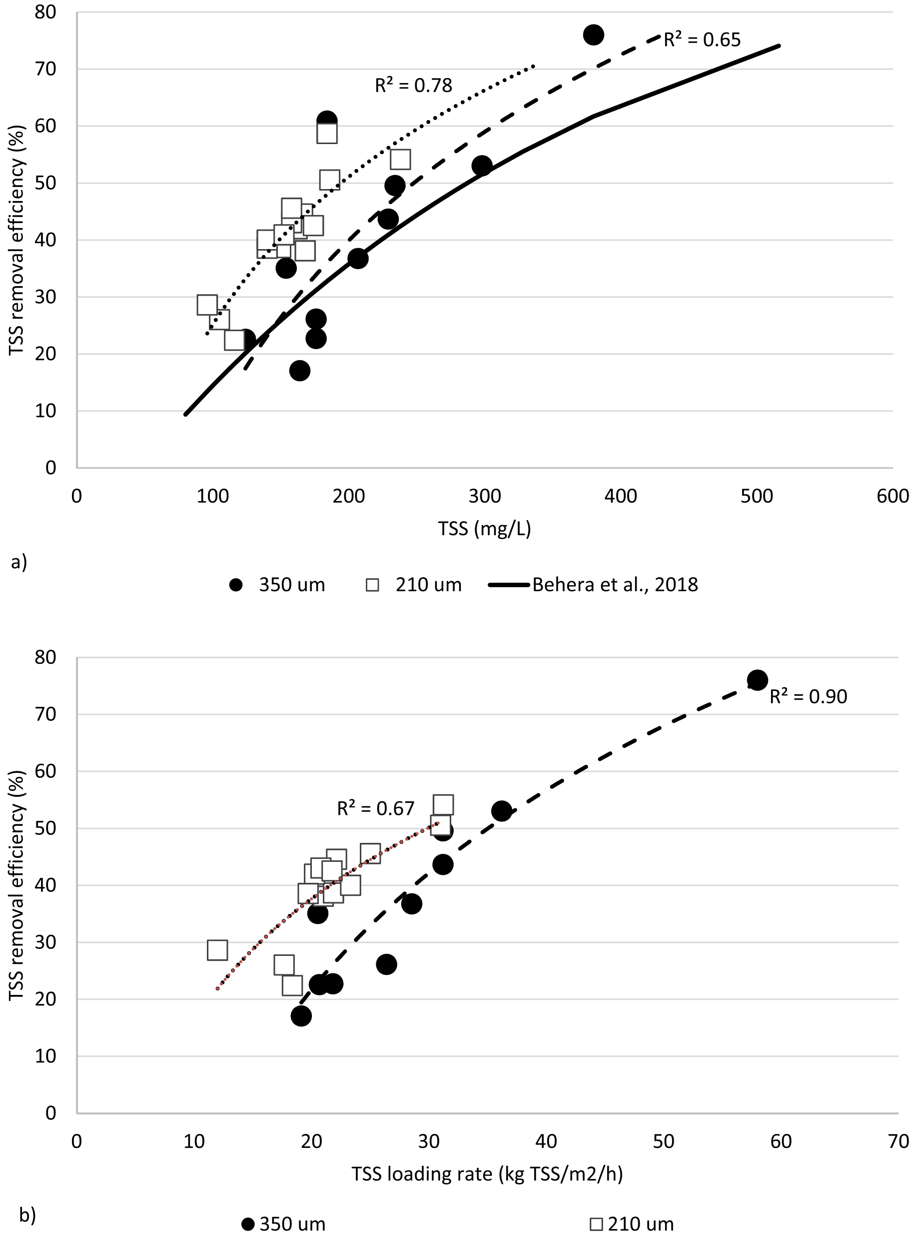

Fig. 2. a) Effect of the TSS concentration on TSS removal efficiency; b) Effect of the TSS loading rate of the TSS removal efficiency.

methanogenic activity (Dahiya et al., 2015) as aerobic oxidation and accumulation of storage polymers can be excluded under these conditions. The maximal concentration without the adjustment of pH was 7.8 gCOD/l (Y_{VFA} of 232 $mgCOD_{VFA}$/gVS) as the acidogenic fermentation was limited by acidic pH. It is well known that low pH increases the concentration of undissociated bio-based VFAs (Atkins and de Paula, 1990) that act as uncoupling compounds, thus the microorganisms consume more energy for maintaining neutral pH inside the cell and reduce their performances (Imai and Ohno, 1995; Verduyn et al., 1992).

The highest performances were obtained with initial pH 9 (17.7 g COD/l) that corresponded to a VFAs production yield of 521 $mgCOD_{VFA}$/gVS. The best performances under weak alkaline conditions can be explained by several factors such as the lower toxicity of undissociated forms of bio-based VFAs due to highest pH during the first days of fermentation and the enhanced hydrolysis process due to the alkaline pH which decrease the particle size (Lin and Li, 2018) and reduces both the degree of polymerization and cellulose crystallinity (Kim et al., 2015). The size reduction is beneficial to the biodegradability because smaller particle sizes increase the available specific surface and improve microbial attachment and enzyme activity (Vavilin et al., 2008).

Test with initial pH 11 was characterized by slow increase of bio-based VFAs concentration until day 9 related with decrease of pH value. The enhanced hydrolysis due to the initial alkaline conditions was not followed by high bio-based VFAs concentration because the pH allowed the activity of methanogens that converts part of the VFAs into methane, on the other hand the acidogenesis has

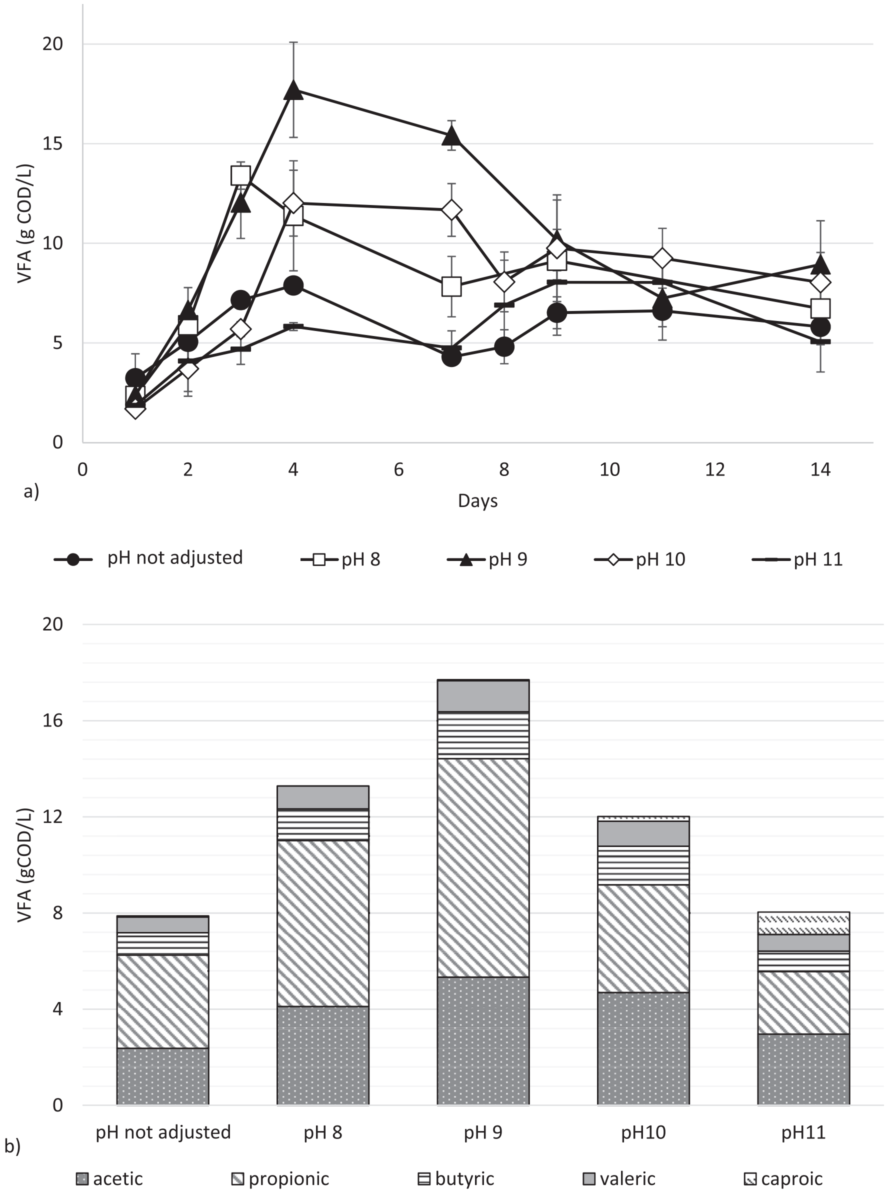

Fig. 3. a) Profile of the VFAs observed during the batch experiments at different initial pH; 3b) Composition of the VFAs obtained at maximal VFAs concentration.

higher kinetics than methanogenesis resulting in an increase of VFAs concentration and subsequent pH reduction. As soon as the pH dropped to acidic value the methane production decreased progressively until complete inhibition, while the VFAs content continued to increase.

In general, the main type of VFAs observed in the fermentation liquid at the end were acetic, propionic and butyric acids, although a significant variation in the composition were observed according with the different initial pH. The tests carried out with initial pH 6.6–9 showed a quick increase of propionic acid percentage that rise from 28 to 38% after 1 day to value higher than 48% after 3 days of fermentation. The batches with stronger alkaline conditions were characterized by slower shift in the VFAs composition: when the maximal VFAs concentration was detected (day 4 for pH 10 and day 9 for pH 11) the most abundant VFAs was acetic acid (39% and 37%) and percentage of propionic acid increased in the subsequent days, however reaching reach a comparable VFAs composition to other batches (47–48%). These results demonstrate that hydrolysis rate varied as a function of initial pH. On the other hand the acidogenic bacteria population or degradation pathway were not affected: indeed as soon as the stable acidic condition were reached the same VFAs composition was observed. The Fig. 3b show the bio-based VFAs distribution when maximal concentration was achieved, while Fig. S3 (supplementary material) reported the complete profile of VFAs observed during the fermentation batches. Wang et al. (2019) demonstrated that chemical pre-treatment of lignocellulose using CaO_2 destroyed substancially benzene rings and the double bonds, consequentely the cellulose become more bio-available for microbial hydrolysis, and many small molecular organics were produced including two alcohol-like substances, two aldehyde-like substances, one ketone-like substance, five acid-like substances, and two ester-like substances. One of these intermediates was the lactic acid (called also 2-Hydroxypropanoic acid), an organic acid which is produced by lactic acid bacteria from hexoses and pentoses as an anaerobic product of glycolysis (Abdel-Rahman et al., 2013) which could drive the production of propionic acid during the acidogenesis stage (Chen et al., 2013). Therefore the propionic acid is one of the main VFAs expected as fermentation product but its amount depends on environmental conditions. In particular the stronger alkaline conditions could determine the reversible inactivation of vitamin B12 that is an important factor in the conversion of pyruvate to propionate (Huang et al., 2018) and could explain the lower percentage of propionic acid at the beginning of tests with alkaline condition.

All the tests showed that maximum bio-based VFAs concentration was followed by a gradual decrease due to partial consumption of organic acids caused probably by methanogenesis which took place also at unfavorable conditions. The consumption of bio-based VFAs indicated that choice of HRT was also important to maximize the yield.

The release of nutrients like ammonium and phosphorus was affected by both pH and hydraulic retention time. Phosphate concentration reached the maximal release after 7 days for all the condition, however the concentration increased proportionally with the initial pH from 41 to 64 mg PO_4-P/L in batches with uncontrolled pH and adjusted at pH 11 (Table 2), respectively. The ammonium concentration increased along the tests, which negatively affected the COD/NH_4-N ratio concurrent with the reduction of the bio-based VFAs at longer time of fermentation.

Table 2 report the best performances reached with different initial pH considering both the concentration and the ratio between bio-based VFAs and nutrients. COD/NH_4-N and COD/PO_4-P ratios underline the importance to meet the compromise between bio-based VFAs production and nutrients release. In particular, the initial pH 8 led to lower bio-based VFAs concentration than using initial pH 9 but nitrogen release was significant lower, resulting in a COD_{VFA}/NH_4-N of 172 mgCOD/mgN vand 105 mgCOD/mgN respectively. Finally, the cost of the alkaline source used to control the initial pH should be taken into account because around 0.5 ml of NaOH 30% per liter of CPS must be used to increase 1 pH unit.

3.5. Sequencing batch fermentation reactor

The best conditions observed from the batch test were applied in the SBFR, which aimed at the evaluation of the bio-based VFAs productivity under real environmental conditions. The application of batch configuration is often not economic feasible because leads to construction of a higher number of reactors with smaller dimension than one single sequencing batch fermentation. The SBFR started with RUN1 applying an HRT of 6 days and feeding CPS with VS concentration as high as 71 gVS/kg, thus the OLR resulted of 17.7 kgVS/m^3d. Under these conditions, the bio-based VFAs concentration was 8,37 mgCOD/L, comparable to the value observed in the batch test without any pH control, but the VFAs production yield was lower (154 $mgCOD_{VFA}$/g VS) if compared with other studies (Crutchik et al., 2018; Li et al., 2011). The behavior could be explained by the constant acidic pH which negatively affected the activity of the acidogenic bacteria (Table 3). Although alkalinity was available in the liquor background of the CPS, it was not enough to cope the acidification due to the VFAs formation. Propionic acid was the dominant VFAs in the mixture which constituted the 46% of total bio-based VFAs, while acetic acid accounted for 30%. The specific yield in term of propionic acid was 70 mgCOD/g VS in agreement with results reported by Crutchik et al. (2018) that have operated using same conditions. Regarding the nutrients released, the ammonium concentration in the fermentation liquid was 370 mgN/L and the phosphate was 101 mgP/L corresponding to release of 24% of N and 35% of P. The acidification degree was 88% meaning that most of solubilized COD was converted into VFAs. Consequently most of organic carbon in the liquid fraction can be directly used for denitrification. Considering the low VFAs yields observed in RUN1 (around 70% of batch value), RUN 2 was carried out with longer HRT (14 d) to evaluate how this parameter effected hydrolysis and VFAs concentration. The HRT was increased while OLR decreased to 5.7 kgVS/m^3d. The bio-based VFAs yield was 137 $mgCOD_{VFA}$/gVS and the average VFAs concentration was 8,79 mgCOD/L, thus performance related to VFAs did not change significantly from RUN1 and the longer HRT did not improve the hydrolysis. On the other hand, the acidification degree increased up to 94% indicating that almost all hydrolyzed COD was converted into bio-based VFAs, thus longer HRT promoted the step of conversion of dissolved compounds into VFAs. The latter were composed mainly by acetic and propionic acids (30% and 52% respectively), thus the specific yield in terms of propionic acid was 71 mgCOD/gVS.

The ammonium and phosphate concentrations were 290 mgN/L and 70 mgP/L, and these values were slightly lower than those detected in RUN1 because of lower nutrients volumetric loading rates compared with RUN1 (NLR and PLR, Table 1). The nutrients released in RUN 1 were around 25% and 31% of the nitrogen and phosphorus influent, which did not statistically differ ($p < 0.05$, details reported in Supplementary Information) with RUN 2. During the RUN3, the pH was adjusted at 9.0 just before feeding, while the HRT was decreased at 6 days which corresponded to an OLR of 7.5 kgCOD/m^3d. The liquid fraction of pre-treated sludge showed that soluble COD was three times greater than raw CPS, while bio-based VFAs concentration did not change, on the other hand the pH increase determined a partial precipitation of phosphate. The bio-based VFAs and propionic acid yields increased significantly in the reactor (322 and 164 $mgCOD_{VFA}$/gVS, respectively) during RUN 3, despite the maximum values (>400 $mgCOD_{VFA}$/gVS) were often observed when feeding was interrupted during the week-end which gave longer retention to convert soluble compounds into VFAs. The propionic and acetic acids constituted the 51% and 28% of total bio-based VFAs, thus the profile of organic acids was not affected by initial pH of sludge confirming the batch insight. With respect to COD mass balance, initial alkaline pH allowed the solubilization of 24% of the particulate COD and the overall balance

Table 2
Performances of batch tests and effluent characteristics.

Parameter	unit	pH not adjusted	pH 8	pH 9	pH 10	pH 11
pH	–	4.84 ± 0.01	4.78 ± 0.03	4.86 ± 0.01	5.49 ± 0.53	5.62 ± 0.97
bio-based VFA	mgCOD/L	7,883 ± 197	13,397 ± 683	17,704 ± 2,389	12,016 ± 1,653	8,040 ± 2,657
Y_{VFA}	mgCOD/gVS	232 ± 6	394 ± 20	521 ± 70	353 ± 49	236 ± 78
NH_4	mgN/L	68.6 ± 2.0	77.7 ± 3.0	168.0 ± 2.0	303.1 ± 40.6	408.9 ± 39.6
PO_4	mgP/L	40.8 ± 3.2	55.5 ± 2.1	55.4 ± 1.4	52.3 ± 3.2	63.9 ± 5.4
COD_{VFA}/NH_4-N	mgCOD/mgN	114.9 ± 0.4	172.4 ± 2.6	105.4 ± 15.5	39.6 ± 40.9	19.7 ± 8.4
COD_{VFA}/PO_4-P	mgCOD/mgP	193.4 ± 10.3	241.2 ± 21.3	319.4 ± 35.5	229.8 ± 234.2	125.9 ± 31.0

Table 3

Average and standard deviation of fermentation liquid fraction and performance during each runs.

	Unit	RUN1	RUN2	RUN3
Effluent characteristics				
pH	–	4.9 ± 0.1	4.8 ± 0.1	4.9 ± 0.2
Alkalinity (pH 4.3)	$mgCaCO_3/L$	969 ± 125	918 ± 71	1447 ± 209
sCOD	mgCOD/L	9,500 ± 915	8,786 ± 1,568	11,890 ± 1,529
bio-based VFAs	mgCOD/L	8,361 ± 2,275	8,347 ± 1,754	9,975 ± 1,984
Acetic acid	%VFAs	30 ± 3	25 ± 5	29 ± 3
Propionic Acid	%VFAs	45 ± 6	53 ± 5	51 ± 5
NH_4	mgN/L	326 ± 23	290 ± 19	198 ± 25
PO_4	mgP/L	101 ± 11	70 ± 12	72 ± 15
bio-based VFAs/N	gCOD/gN	26	29	50
Process performances				
Y_{VFA}	mgCOD/gVS	154 ± 24^{a}	137 ± 33 ^{a}	322 ± 56 ^{b}
VFAs productivity	$kgCOD/m^3_{reactor}$ d	1.23	0.44	2.57
Y_{HPr}	mgCOD/gVS	70 ± 15 ^{a}	71 ± 20 ^{a}	164 ± 40 ^{b}
Acidification Degree	%	88 ± 7 ^{a,b}	94 ± 4 ^{a,}	81 ± 9 ^{b}
N Release	%	24 ± 2 ^{a}	25 ± 2 ^{a}	37 ± 4 ^{b}
P release	%	35 ± 4 ^{a}	31 ± 8 ^{a}	49 ± 9 ^{b}

Details about statistical analysis is reported in the Supplementary information.

closed with an error lower than 10% that can be explained with the neglected methane production. As compared with RUN 3, the RUN 1 and RUN 2 showed a solubilization of 9% and 8% of fed COD respectively while the balance closed with similar error, indicating that the lose due to gas emission were not affected by HRT or initial pH.

The acidic pH determined the dissolution of precipitated nutrients and overall releases were quantified as 40% of N and 51% of P, value higher than other operations ($p < 0.05$). However, the lower loading rate values (Table 1) determined concentrations comparable to RUN2 (232 mgN/L and 68 mgP/L). Although the Y_{SCFA} were similar in RUN1 and RUN2, operating at HRT of 6 days allowed the achievement of a productivity of 1.23 $kgCOD_{VFA}/m^3d$.

The average characteristics of the CPS fermentation liquid and process performances were resumed in Table 2S (supplementary material). The concentrations of bio-based VFAs depended on initial pH and the consequent yield, but also the CPS solids concentration affected the final content, thus the concentration in RUN 3 could have been further increased using higher OLR. Considering the average VS content of CPS during the overall experimentation (44.7 gVS/kg) and the operational conditions, the potential VFAs concentration could increase up to 14.4 gCOD/L resulting in a volumetric productivity of 2.57 $kgCOD/m^3$ d of bio-based VFAs.

3.6. *Economical assessment*

A cost-benefit analysis of the CPS biorefinery was performed by comparing the main revenues and related cost and potential savings due to valorization of recovered resources, like methane and VFA.

Three different scenarios were considered which differ by the type of valorization of CPS and the recovery pathway of the VFAs produced from the acidogenic fermentation (Table 4).

Specifically, the SCENARIO 1 (S1) can be considered as reference as the CPS is completely valorized to biogas/methane through anaerobic digestion, while the carbon source for biological nutrients removal is supplied from the market as acetic acid. The SCENARIO 2 (S2) introduced the acidogenic fermentation of CPS, while VFAs (after solid/liquid separation) could replace the use of external carbon source (e.g. acetic acid) for the BNR process. Finally, SCENARIO3 (S3) considered the utilization of VFAs as carbon source for PHA production through the process described by Frison et al. (2015a, 2015b). Fig. 4 resumes the economic evaluations splitted as revenues and main associated costs derived from the integration of the RBF and acidogenic fermentation in a WWTP.

In the S1, the amount of CH_4 that can be obtained from 1 ton TS of CPS is around 310 m^3, corresponding to an electrical energy of around 1150 kWh through a Combined Heat and Power (CHP) unit (considering 38% of electrical energy efficiency). Assuming a selling price of 0,16 €/kWh, the electrical energy accounts for 185 €/ton of TS that could be considered as the unique saving of this configuration when the electricity is used for internal consumption to partially fulfill the needs of the WWTPs. On the other hand, this scenario includes the utilization external carbon source for the biological removal of nutrients (nitrogen and phosphorus) released in the anaerobic digester which increase the operational costs due to the supply of acetic acid (market price around 450 €/ton, ICIS – Intelligent Commodity Intelligent Services) and the related cost for disposal of surplus sludge production. The latter accounts for 32 €/ton TS, considering 100 €/ton of sludge disposed at 25% of dry matter. The difference between the revenues for the utilization of the methane and operational costs was positive (53 €/ton TS).

According to the best operating conditions found in this work, the production of VFAs from CPS for the S2 and S3 could achieve 311 kgCOD/ton TS that can replace the external acetic acid addition and avoid the production and disposal of surplus sludge. The equivalent COD as VFAs corresponds to 96 €/ton TS as cost avoided for the purchase of external carbon source in S1. The diversion of organic matter in the sludge line with VFAs recovery leads the reduction of methane from the anaerobic digester by around 30%, which affects the revenues due to the production of electricity.

The SFBR could be maintained under mesophilic condition using the surplus heat from methane combustion in CHP without affect significantly in the electricity and heat consumption. On the other hands, the CPS conditioning to pH 9 needs the addition of alkali while the solid/liquid separation of the fermentation liquid fraction enriched of VFAs requires polyelectrolite dosage. Therefore, the cost for chemical consumption has been calculated based on NaOH and polyelectrolite consumption (10 g NaOH/gTS fermented and 35 g of polyelectrolite/kgTS separated) which accounted for 48 €/ton TS, almost 80% of the total cost in S2. However, the replace of external carbon source with bio-based VFAs leads to positive benefits of 67 €/ton TS, which is 20% higher than S1.

The S3 considered the conversion of the bio-based VFAs into PHA according with a production yield of 0.11 $KgCOD_{PHA}/kgCOD_{VFA}$ found at lab scale by Frison et al. (2015a, 2015b), resulting in 25 kg of PHA per ton of TS treated with nitrogen removal. The valorization of the VFAs to PHAs increased the potential revenue up to 90 €/ton TS treated due their selling price (between 2.2 and 5 €; /kgPHA,

Table 4
Description of scenarios considered for CPS biorefinery in the WWTP.

Scenario	Description
SCENARIO 1 (S1)	- Wastewater sieving + Anaerobic digestion of CPS + CHP unit; - Acetic acid for biological nutrients removal
SCENARIO 2 (S2)	- Wastewater sieving + Acidogenic fermentation of CPS + bio-based VFAs for biological nutrients removal; - Anaerobic digestion of the residual CPS + CHP unit
SCENARIO 3 (S3)	- Wastewater sieving + Acidogenic fermentation of CPS + bio-based VFAs for PHAs production and biological nutrients removal; - Anaerobic digestion of the residual CPS + CHP unit

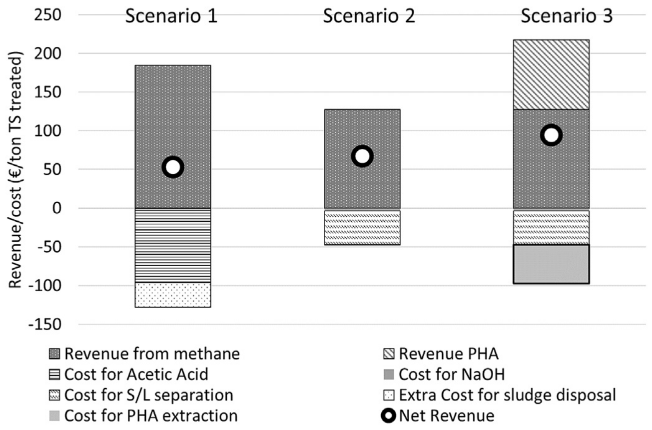

Fig. 4. Revenue vs Cost for different utilization of CPS and bio-based VFAs.

Valentino et al., 2017), although a real market is not established yet. Considering the potential value from the methane, the potential revenue in S3 were 218 €/ton SS. If the cost for the extraction and purification of PHA is assumed to be 2 €/kg of PHA recovered according with literature (Mudliar et al., 2008; Leong et al., 2017), the net revenue would still be positive, and it would reach 95 €/kg TS treated which is significantly higher than S1. Also, the latter represents the most cost-effective pathway of valorization of VFAs and agreed with the results reported in Kleerebezem et al. (2015), despite the most relevant costs associated with the bioprocess and downstream were considered in this work.

4. Conclusions

The pilot-scale RBF was applied for the sieving of municipal wastewater with a TSS content of 197 mg TSS/L and the best performances were reached operating with a solid loading rate higher than 30–35 kgTSS/m^2 h without any influence by mesh porosity. The TSS removal efficiency was 44%, while the CPS had a TS content of 48.6 gTS/kg with 92% constituted by organic matter. The batch fermentation showed a positive impact of sludge pH conditioning (pH 9) on acidogenic fermentation with potential bio-based VFA yield that increased from 232 to 521 mg bio-based VFA/gVS due to the faster hydrolysis of organic matter, where propionic acid accounted for about 50% of total bio-based VFA. On the other hand, pH higher than 9 determined a delay in VFAs accumulation because of methanogenic activity and partial consumption of bio-based VFA. The effect of HRT and pH adjustment was evaluated by pilot-scale SBFR operating with HRT of 6 and 14 days. The longer HRT did not improve significantly the bio-based VFA yield (134–154 mgCOD$_{VFA}$/gVS) but higher acidification degree was observed (94%) while the adjustment of sludge pH to value of 9 improved the bio-based VFA yield to 322 mgCOD$_{VFA}$/gVS. The nitrogen release increased from 24 to 40% when pH was adjusted and the resulted COD/N ratio was 50 mgCOD/mgN which could be considered as a valid carbon source to support the biological nutrients removal process and PHA production. This operating condition allowed to obtain a VFAs concentration up to 14.4 gCOD/L and a daily productivity of 2.57 kgCOD/m^3 d. The best average solid removal efficiency observed during the operation of the RBF and the optimized operational conditions in SBFR were considered for the evaluation of a preliminary economical assessment. The integration of RBF and acidogenic fermentation for VFAs production from CPS combine both benefits due to the diversion of organic matter in the side-stream treatment line for methane and bio-based VFAs production. Specifically, the utilization of VFAs from CPS as carbon source could be a good option to replace the dosage of external carbon source and enhance the BNR process in existing WWTP. On the other hand, the highest net benefits were found when VFAs are valorized to PHAs, although their current selling price, extraction and purification costs are still under debate due to the lack of an actual market of PHA-bioplastics from WWTPs.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.watres.2020.115633.

References

Kampschreur, M.J., Temmink, H., Kleerebezem, R., Jetten, M.S., van Loosdrecht, M.C., 2009. Nitrous oxide emission during wastewater treatment. Water Res. 43 (17), 4093–4103. https://doi.org/10.1016/j.watres.2009.03.001.

Abdel-Rahman, M.A., Tashiro, Y., Sonomoto, K., 2013. Recent advances in lactic acid production by microbial fermentation processes. Biotechnol. Adv. 31, 877–902. https://doi.org/10.1016/J.BIOTECHADV.2013.04.002.

Akyol, C., Foglia, A., Ozbayram, E.G., Frison, N., Katsou, E., Eusebi, A.L., Fatone, F., 2019. Validated innovative approaches for energy-efficient resource recovery and re-use from municipal wastewater: from anaerobic treatment systems to a biorefinery concept. Crit. Rev. Environ. Sci. Technol. https://doi.org/10.1080/10643389.2019.1634456.

American Public Health Association, American Water Works Association, Water Environment Federation, 1999. Standard methods for the examination of water and wastewater. Stand. Methods 541.

Atasoy, M., Owusu-Agyeman, I., Plaza, E., Cetecioglu, Z., 2018. Bio-based volatile fatty acid production and recovery from waste streams: current status and future challenges. Bioresour. Technol. 268, 773–786. https://doi.org/10.1016/J.BIORTECH.2018.07.042.

Atkins, P.W., de Paula, J., 1990. Physical Chemistry, vol. 5. Oxford University Press, Oxford, p. 1007.

Behera, C.R., Santoro, D., Gernaey, K.V., Sin, G., 2018. Organic carbon recovery modeling for a rotating belt filter and its impact assessment on a plant-wide scale. Chem. Eng. J. 334, 1965–1976. https://doi.org/10.1016/J.CEJ.2017.11.091.

Bhatia, S.K., Yang, Y.H., 2017. Microbial production of volatile fatty acids: current status and future perspectives. Rev. Environ. Sci. Biotechnol. https://doi.org/10.1007/s11157-017-9431-4.

Chen, Y., Li, X., Zheng, X., Wang, D., 2013. Enhancement of propionic acid fraction in volatile fatty acids produced from sludge fermentation by the use of food waste and Propionibacterium acidipropionici. Water Res. 47, 615–622. https://doi.org/10.1016/J.WATRES.2012.10.035.

Crutchik, D., Frison, N., Eusebi, A.L., Fatone, F., 2018. Biorefinery of cellulosic primary sludge towards targeted Short Chain Fatty Acids, phosphorus and methane recovery. Water Res. 136, 112–119. https://doi.org/10.1016/j.watres.2018.02.047.

Dahiya, S., Sarkar, O., Swamy, Y.V., Venkata Mohan, S., 2015. Acidogenic fermentation of food waste for volatile fatty acid production with co-generation of biohydrogen. Bioresour. Technol. 182, 103–113. https://doi.org/10.1016/J.BIORTECH.2015.01.007.

Dimock, R., Morgenroth, E., 2006. The influence of particle size on microbial hydrolysis of protein particles in activated sludge. Water Res. 40 (10), 2064–2074.

Feng, L., Chen, Y., Zheng, X., 2009. Enhancement of waste activated sludge protein conversion and volatile fatty acids accumulation during waste activated sludge anaerobic fermentation by carbohydrate substrate addition: the effect of pH. Environ. Sci. Technol. 43, 4373–4380. https://doi.org/10.1021/es8037142.

Franchi, A., Santoro, D., 2015. Current status of the rotating belt filtration (RBF) technology for municipal wastewater treatment. Water Pract. Technol. 10, 319. https://doi.org/10.2166/wpt.2015.038.

Frison, N., Katsou, E., Malamis, S., Oehmen, A., Fatone, F., 2015a. Development of a novel process integrating the treatment of sludge reject water and the production of polyhydroxyalkanoates (PHAs). Environ. Sci. Technol. 49, 10877–10885. https://doi.org/10.1021/acs.est.5b01776.

Frison, N., Katsou, E., Malamis, S., Oehmen, A., Fatone, F., 2015b. Nutrient removal via nitrite from reject water and polyhydroxyalkanoate (PHA) storage during nitrifying conditions. J. Chem. Technol. Biotechnol. 90 (10), 1802–1810.

Ghasimi, D.S.M., Tao, Y., de Kreuk, M., Abbas, B., Zandvoort, M.H., van Lier, J.B., 2015. Digester performance and microbial community changes in thermophilic and mesophilic sequencing batch reactors fed with the fine sieved fraction of municipal sewage. Water Res. 87, 483–493. https://doi.org/10.1016/j.watres.2015.04.027.

Ghasimi, D.S.M., de Kreuk, M., Maeng, S.K., Zandvoort, M.H., van Lier, J.B., 2016. High-rate thermophilic bio-methanation of the fine sieved fraction from Dutch municipal raw sewage: cost-effective potentials for on-site energy recovery. Appl. Energy 165, 569–582. https://doi.org/10.1016/j.apenergy.2015.12.065.

Huang, L., Chen, Z., Xiong, D., Wen, Q., Ji, Y., 2018. Oriented acidification of wasted activated sludge (WAS) focused on odd-carbon volatile fatty acid (VFA): regulation strategy and microbial community dynamics. Water Res. 142, 256–266. https://doi.org/10.1016/j.watres.2018.05.062.

Imai, T., Ohno, T., 1995. The relationship between viability and intracellular pH in the yeast Saccharomyces cerevisiae. Appl. Environ. Microbiol. 61 (10), 3604–3608.

Jain, Siddharth, Jain, Shivani, Wolf, I.T., Lee, J., Tong, Y.W., 2015. A comprehensive review on operating parameters and different pretreatment methodologies for anaerobic digestion of municipal solid waste. Renew. Sustain. Energy Rev. 52, 142–154. https://doi.org/10.1016/j.rser.2015.07.091.

Kim, J.S., Lee, Y.Y., Kim, T.H., 2015. A review on alkaline pretreatment technology for bioconversion of lignocellulosic biomass. Bioresour. Technol. 199, 42–48. https://doi.org/10.1016/j.biortech.2015.08.085.

Kleerebezem, R., Joosse, B., Rozendal, R., Van Loosdrecht, M.C., 2015. Anaerobic digestion without biogas? Rev. Environ. Sci. Bio/Technol. 14 (4), 787–801. https://doi.org/10.1007/s11157-015-9374-6.

Lee, W.S., Chua, A.S.M., Yeoh, H.K., Ngoh, G.C., 2014. A review of the production and applications of waste-derived volatile fatty acids. Chem. Eng. J. 235, 83–99. https://doi.org/10.1016/j.cej.2013.09.002.

Leong, Y.K., Show, P.L., Lan, J.C.W., Loh, H.S., Lam, H.L., Ling, T.C., 2017. Economic and environmental analysis of PHAs production process. Clean Technol. Environ. Policy 19 (7), 1941–1953. https://doi.org/10.1007/s10098-017-1377-2.

Li, S.-Y., Srivastava, R., Suib, S.L., Li, Y., Parnas, R.S., 2011. Performance of batch, fed-batch, and continuous A–B–E fermentation with pH-control. Bioresour. Technol. 102, 4241–4250. https://doi.org/10.1016/J.BIORTECH.2010.12.078.

Lin, L., Li, X., 2018. Effects of pH adjustment on the hydrolysis of Al-enhanced primary sedimentation sludge for volatile fatty acid production. Chem. Eng. J. 346, 50–56. https://doi.org/10.1016/J.CEJ.2018.04.005.

Liu, F., Tian, Y., Ding, Y., Li, Z., 2016. The use of fermentation liquid of wastewater primary sedimentation sludge as supplemental carbon source for denitrification based on enhanced anaerobic fermentation. Bioresour. Technol. 219, 6–13. https://doi.org/10.1016/J.BIORTECH.2016.07.030.

Longo, S., Frison, N., Renzi, D., Fatone, F., Hospido, A., 2017. Is SCENA a good approach for side-stream integrated treatment from an environmental and economic point of view? Water Res. 125, 478–489.

Maktabifard, M., Zaborowska, E., Makinia, J., 2018. Achieving energy neutrality in wastewater treatment plants through energy savings and enhancing renewable energy production. Rev. Environ. Sci. Bio/Technol. 17, 655–689. https://doi.org/10.1007/s11157-018-9478-x.

Metcalf & Eddy, 2014. Wastewater engineering: treatment and resource recovery. Public Works Engineering. https://doi.org/10.1007/978-1-349-06927-9_4.

Miron, Y., Zeeman, G., Van Lier, J.B., Lettinga, G., 2000. The role of sludge retention time in the hydrolysis and acidification of lipids, carbohydrates and proteins during digestion of primary sludge in CSTR systems. Water Res. 34, 1705–1713. https://doi.org/10.1016/S0043-1354(99)00280-8.

Mudliar, S.N., Vaidya, A.N., Kumar, M.S., Dahikar, S., Chakrabarti, T., 2008. Techno-economic evaluation of PHB production from activated sludge. Clean Technol. Environ. Policy 10 (3), 255.

Oehmen, A., Lemos, P.C., Carvalho, G., Yuan, Z., Keller, J., Blackall, L.L., Reis, M.A.M., 2007. Advances in enhanced biological phosphorus removal: from micro to macro scale. Water Res. 41, 2271–2300. https://doi.org/10.1016/J.WATRES.2007.02.030.

Palmieri, S., Cipolletta, G., Pastore, C., Giosuè, C., Akyol, Ç., Eusebi, A.L., Frison, N., Tittarelli, F., Fatone, F., 2019. Pilot scale cellulose recovery from sewage sludge and reuse in building and construction material. Waste Manag. https://doi.org/10.1016/j.wasman.2019.09.015.

Paulsrud, B., Rusten, B., Aas, B., 2014. Increasing the sludge energy potential of wastewater treatment plants by introducing fine mesh sieves for primary treatment. Water Sci. Technol. 69, 560. https://doi.org/10.2166/wst.2013.737.

Ravndal, K.T., Opsahl, E., Bagi, A., Kommedal, R., 2018. Wastewater characterisation by combining size fractionation, chemical composition and biodegradability. Water Res. 131, 151–160. https://doi.org/10.1016/j.watres.2017.12.034.

Razafimanantsoa, V.A., Ydstebø, L., Bilstad, T., Sahu, A.K., Rusten, B., 2014. Effect of selective organic fractions on denitrification rates using Salsnes Filter as primary treatment. Water Sci. Technol. 69, 1942. https://doi.org/10.2166/wst.2014.110.

Resource Recovery Cluster, 2015. State of the Art Compendium Report on Resource Recovery from Water. IWA International Water Association, London, UK.

Ruiken, C.J., Breuer, G., Klaversma, E., Santiago, T., van Loosdrecht, M.C.M., 2013. Sieving wastewater – cellulose recovery, economic and energy evaluation. Water Res. 47, 43–48. https://doi.org/10.1016/j.watres.2012.08.023.

Rusten, B., Lundar, A., 2006. How a simple bench-scale test greatly improved the primary treatment performance of fine mesh sieves. In: Proceedings of the Water Environment Federation, pp. 1919–1935.

smart-plant [WWW Document], n.d. URL. www.smart-plant.eu. http://www.smart-plant.eu/ (accessed 6.10.19).

Thangamani, A., Rajakumar, S., Ramanujam, R.A., 2010. Anaerobic co-digestion of hazardous tannery solid waste and primary sludge: biodegradation kinetics and metabolite analysis. Clean Technol. Environ. Policy 12, 517–524. https://doi.org/10.1007/s10098-009-0256-x.

Valentino, F., Morgan-Sagastume, F., Campanari, S., Villano, M., Werker, A., Majone, M., 2017. Carbon recovery from wastewater through bioconversion into biodegradable polymers. N. Biotechnol. 37, 9–23. https://doi.org/10.1016/j.nbt.2016.05.007.

van der Hoek, J.P., de Fooij, H., Struker, A., 2016. Wastewater as a resource: strategies to recover resources from Amsterdam's wastewater. Resour. Conserv. Recycl. 113, 53–64. https://doi.org/10.1016/j.resconrec.2016.05.012.

Vavilin, V.A., Fernandez, B., Palatsi, J., Flotats, X., 2008. Hydrolysis kinetics in anaerobic degradation of particulate organic material: an overview. Waste Manag. 28, 939–951. https://doi.org/10.1016/J.WASMAN.2007.03.028.

Verduyn, C., Postma, E., Scheffers, W.A., Van Dijken, J.P., 1992. Effect of benzoic acid on metabolic fluxes in yeasts: a continuous-culture study on the regulation of respiration and alcoholic fermentation. Yeast. https://doi.org/10.1002/yea.320080703.

Verstraete, W., Van de Caveye, P., Diamantis, V., 2009. Maximum use of resources present in domestic "used water.". Biores. Tech. 100 (23), 5537–5545.

Wang, X., McCarty, P.L., Liu, J., Ren, N.-Q., Lee, D.-J., Yu, H.-Q., Qian, Y., Qu, J., 2015. Probabilistic evaluation of integrating resource recovery into wastewater treatment to improve environmental sustainability. Proc. Natl. Acad. Sci. Unit. States Am. 112 https://doi.org/10.1073/pnas.1410715112, 1630e1635.

Wang, D., He, D., Liu, X., Xu, Q., Yang, Q., Li, X., Liu, Y., Wang, Q., Ni, B., Li, H., 2019. The underlying mechanism of calcium peroxide pretreatment enhancing methane production from anaerobic digestion of waste activated sludge. Water Res. 164, 114934.

Wu, H., Gao, J., Yang, D., Zhou, Q., Liu, W., 2010. Alkaline fermentation of primary sludge for short-chain fatty acids accumulation and mechanism. Chem. Eng. J. 160, 1–7. https://doi.org/10.1016/j.cej.2010.02.012.

Zhou, Y., Stanchev, P., Katsou, E., Awad, S., Fan, M., 2019. A circular economy use of recovered sludge cellulose in wood plastic composite production: recycling and eco-efficiency assessment. Waste Manag. 99, 42–48. https://doi.org/10.1016/J.WASMAN.2019.08.037.