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Pilot-scale multi-purposes approach for volatile fatty acid production, hydrogen and methane from an automatic controlled anaerobic process

Federico Micolucci [a, *, 1], Marco Gottardo [b, 1], David Bolzonella [c], Paolo Pavan [b], Mauro Majone [d], Francesco Valentino [d]

[a] Department of Chemical Engineering, Lund University, PO Box 124, SE-221 00, Lund, Sweden
[b] Department of Environmental Science, Informatics and Statistics, Via Torino 155, 30170, Venice Mestre, Italy
[c] Department of Biotechnology, University of Verona, Strada Le Grazie 15, 37134, Verona, Italy
[d] Department of Chemistry, "La Sapienza" University of Rome, P.le Aldo Moro 5, 00185, Rome, Italy

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ABSTRACT

A combined two-levels control method has been developed and tested on a long term operation of a two-phases pilot-scale anaerobic process for the concurrent production of volatile fatty acids, hydrogen and methane. The latter was designed for the treatment of food waste of urban origin (namely, the organic fraction of municipal solid waste). The optimized control method was set on the base of the inputs of three online probes: a pH-meter in the fermentation reactor, a pH-meter and a conductivity probe in the digestion reactor. The first control level managed the pH in the fermentation reactor while the second control level managed the ammonia concentration in the digestion reactor. This combination established the volume of the digestate to be recycled from the digestion to the fermentation reactor, optimizing the yield of volatile fatty acid (0.31–0.32 kg COD_{VFA}/kg COD_{fed}) and the specific hydrogen production (SHP; 0.070–0.074 m^3 H_2/kg TVS_{fed}) in the fermentation reactor and the specific methane production (SMP; 0.48–0.55 m^3 CH_4/kg TVS_{fed}) in the digestion reactor.

A new process configuration was also proposed and applied over the course of the long operation period. This configuration allowed to remove part of the volatile fatty acid-rich liquid stream from the fermenter effluent, maintaining the corresponding solid-rich effluent in the whole system (as feed for the digestion reactor) by using a solid/liquid separation unit. In this way, the concentration of volatile fatty acids in the digester was kept at a low level, even with high loading rates, so maintaining a satisfying efficiency of methane production and utilizing the excess volatile fatty acids (out of the system) as building blocks for other purposes.

The optimized two-levels control method for the anaerobic treatment of food waste provides new perspectives for the valorisation of such waste stream; the production of building blocks namely volatile fatty acids supports new innovative bio-refinery platforms for the production of bio-products.

1. Introduction

The European Union (EU) 2030 strategy leads to ensure sustainable growth via a new and ambitious package of measures on "Circular Economy" (ec.europa.eu, 2020). These measures aim to increase products lifetime and their recyclability benefiting both the environment and the economy.

Food waste management is part of this vision as it allows for conservative use of resources like carbon, nitrogen and phosphorous among the others. Separate collection, which allows for the production of streams of recycled materials of good quality level, is a pre-requisite of this global approach. Once this material is collected it is the perfect substrate for the application of

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* Corresponding author.
E-mail addresses: federico.micolucci@chemeng.lth.se (F. Micolucci), marco.gottardo@unive.it (M. Gottardo), david.bolzonella@univr.it (D. Bolzonella), pavan@unive.it (P. Pavan), mauro.majone@uniroma1.it (M. Majone), francesco.valentino@uniroma1.it (F. Valentino).
[1] Contributed equally to the study.

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Abbreviations

AD	anaerobic digestion
AnD	anaerobic digestion reactor
AnF	anaerobic fermentation reactor
COD	chemical oxygen demand
COD_{SOL}	soluble COD
COD_{VFA}	volatile fatty acids expressed as COD
CSTR	continuous stirred tank reactor
EU	European Union
GC	gas chromatograph
HRT	hydraulic retention time
LoQ	limit of quantification
OLR	organic loading rate
SRT	sludge retention time
RR	recirculation ratio
SHP	specific hydrogen production
SMP	specific methane production
TCD	thermal conductivity detector
TKN	total Kjeldahl nitrogen
TS	total solids
TVS	volatile solids
VFA	volatile fatty acids
WWTP	wastewater treatment plant

fermentative processes able to produce renewable biofuels in a sustainable way (Micolucci et al., 2016).

Anaerobic digestion (AD) is a well-known and robust technology, largely applied in the EU. AD can be the basis for the development of new approaches where anaerobic processes are considered as the key technology of production sites (biorefineries) rather than disposal sites (Gottardo et al., 2017).

AD treatments of organic waste with energy recovery remain the most attractive strategy for organic waste management (Fernández-Rodríguez et al., 2013). AD pathways can be divided into two main processes, the first dedicated to dark fermentation (DF) and the second step dedicated to methanogenesis. In this process configuration, it is possible to produce volatile fatty acids (VFAs) and hydrogen from the first stage and methane from the second stage (Shen et al., 2017).

Looking at the European perspective, the cumulative percentage of one stage and two stage capacity installed in 2014, was 93% in favour of the single stage (De Baere and Mattheeuws, 2015). However, two-stage AD better connects to the biorefinery concept as it allows to obtain added value streams rich in VFAs from fermentation and integrates the digestion stage to obtain methane from all the organic by-products of the biorefinery chain.

To increase the effectiveness of the hydrogenase enzymes and avoid the inhibition, or a metabolic shift to unwanted high concentrations of lactic acid and unconverted alcohols with consequent reduction of gaseous hydrogen and decrease of VFA production, it is necessary to control and keep the process parameters stable as much as possible (Mussatto et al., 2008)(Zhang et al., 2018). In particular, the pH in the fermenter must be kept over 5.0 to promote the fermentative activity and below 6.0 to avoid the proliferation of methanogenic bacteria.

To optimize VFA and hydrogen production, the pH in the fermentation process has to be controlled and the use of chemicals can be discouraged if a controlled recirculation of the digested effluent from the methanation stage is applied as an eco-friendly solution. The digestate has a buffer function for the pH in the fermentation stage (Chinellato et al., 2013), associated to the alkalinity of anaerobic digestate, which is the result of the equilibrium of carbon dioxide - bicarbonate ions. The use of the recirculation of the digestate is the most economical solution that requires particular attention to avoid an excessive accumulation of ammonia in the system with consequent inhibition of especially the methanogenic stage, due to higher pH (Gottardo et al., 2015)(Cavinato et al., 2016).

Double-stage AD in general shows resilience and high biogas yields, by comparing the two AD systems (mono-stage and double-stage), it was observed that the two-stage has a removal efficiency higher than the single stage, and consequently it has less sludge for disposal (Micolucci et al., 2018). The use of dark fermentation is an important stage to produce VFA, platform chemicals for biorefinery approaches (Micolucci et al., 2018). In the two-phases anaerobic process designed for food waste valorisation, it has been demonstrated that keeping stable the digestate recirculation flow rate by applying a fixed recirculation ratio (RR) does not guarantee the process stability for a long term period (Micolucci et al., 2014). The fastest and uncomfortable solution was to set a recirculation ratio day-by-day, decided by the operator based on the pH value in the fermenter and the weekly trend of the ammonia content in the digester, which was analytically quantified.

Micolucci et al. (2014) introduced a model able to predict the concentration of undissociated ammonia in a two-stage anaerobic digestion system through chemical (VFA) and physical-chemical parameters (alkalinity and electrical conductivity) carried out during a one-year trial. Addressing reference set-points to those parameters, which could indicate a safe range for the system operation, was a crucial step to deliver a precisely defined amount of recycled digestate considering the demands and the reactors' evolution with time. The research demonstrated that ammonia concentration was estimated by means of a multiple linear regression model with statistical software using conductivity, VFA and alkalinity. As a consequence, higher VFAs, bio-hydrogen and biomethane productions can be easily achieved.

In this paper, the optimization and testing of the control system for a long operation period is reported. From a practical point of view, this approach is particularly attractive since it allows to use simple, resilient and low-cost online probes (pH and conductivity). The experimentation was carried out at pilot scale, creating basic support to the further development of full-scale platforms. The creation of an automatized control ensuring a stable acidogenic fermentation and VFA production has been set. This proposed approach allows to constantly monitor and control the stability of the anaerobic digestion system, in an eco-friendly and economic way, bringing AD closer to biorefinery. Within a context of a biorefinery development, the proposed approach is particularly relevant since the production of a VFA-rich stream at constant concentration and with stable chemical features is a key aspect for the synthesis of added value products (Battista et al., 2020).

2. Material and methods

2.1. Inoculum and substrate

The first fermentation reactor was not inoculated with active biomass but it was filled up with a mixture of food organic waste coming from the municipality of Treviso (northeast Italy) and tap water (to increase the economic feasibility of this approach urban wastewater can also be used, without affecting the anaerobic process (Moretto et al., 2020)), to obtain a total solids content of about

8% w/w (Micolucci et al., 2018). The organic matter could be easily converted into hydrogen and organic acids through the action of fermentative bacteria (Zhou et al., 2018). The use of food waste generated an inoculum capable of producing hydrogen under short sludge retention times (STRs), due to the presence of indigenous bacterial communities and the applied thermophilic condition (55 °C). The fermentation reactor was daily fed with a liquid mixture of organic waste, sludge recycled from the second stage (methanogenic reactor) and dilution water to reach the required organic loading rate (OLR).

The methanogenic reactor was inoculated with the anaerobically digested sludge coming from the full-scale mesophilic digester of Treviso WWTP (Da Ros et al., 2017) and maintained under thermophilic temperature (55 °C) for about 1 month, as it was for the first fermentation reactor. The methanogenic reactor was daily fed with the effluent from the first stage. Food waste coming from the municipality of Treviso was used as substrate. The food waste came from a separate source-sorted waste collection and, for the experimental purposes, it was initially pre-treated by soft wet refine approach (Cecchi et al., 2011); later on, it was pre-treated in a dedicated plant by using a screw press, which allowed to obtain a pressed and a more homogeneous organic waste matrix (Micolucci et al., 2016).

2.1.1. Characteristics of the food waste

The organic waste used for this pilot-scale investigation came from the door-to-door collection was conferred in the experimental area (Treviso WWTP) weekly. The incoming waste had a relatively high organic content, in the range 88–93%, which was quantified by volatile solids content. The main characteristics of the pre-treated organic waste are presented below. In the first period (Run1), the conferred food waste was pre-treated by Soft Wet Refine approach before being fed into the reactors; Table 1 shows the chemical-physical characteristics of the food waste as a result of this pre-treatment.

Over the course of Run2 and Run3, the conferred food waste was pre-treated using a screw press which produced a squeezed and homogeneous matrix (food waste juice). Table 2 below shows the chemical and physical characteristics of the pre-treated food waste by Screw Press.

2.2. Experimental set-up

Two stainless steel CSTR reactors (AISI 304) were used for VFAs and biogas production. The first reactor was dedicated to the fermentative step (AnF; 0.2 m^3 working volume) and the second reactor (AnD; 0.6 m^3 working volume) to the methanogenic step. Both reactors were heated by a hot water recirculation system and maintained at 55 °C ± 0.1 using an electrical heater controlled by a PT100-based thermostatic probe. When applied, a centrifugation unit was used for solids/liquid separation after the fermentation process: a coaxial centrifuge equipped with 5.0 μm porosity nylon filter bag for solids removal as described below. Fig. 1 shows the flow diagram of the pilot-scale anaerobic reactors system described above.

Table 1
Physical-chemical characteristics of the wet-refine pre-treated food waste.

Parameter	M.U.	Average ± St.Dev.	Min	Max
TS	gTS/kg	244 ± 32	202	300
TVS	gVS/kg	216 ± 27	175	270
COD	gO_2/kg	230 ± 34	185	299
TKN	gN/kg	6.7 ± 1.0	4.6	11.0
P tot	gP/kg	1.1 ± 0.5	0.6	2.2
pH	–	7.8 ± 0.9	7.2	8.3
VFA	gCOD/l	< LoQ		

Table 2
Physical-chemical characteristics of the screw-press pre-treated food waste.

Parameter	M.U.	Average ± St.Dev.	Min	Max
TS	gTS/kg	213 ± 18	174	242
TVS	gVS/kg	199 ± 19	170	230
COD	gO_2/kg	211 ± 17	174	245
TKN	gN/kg	3.9 ± 1.0	3.0	4.6
P tot	gP/kg	0.57 ± 0.30	0.35	0.98
pH	–	4.9 ± 0.7	3.8	4.7
VFA	gCOD/l	5.7 ± 0.8	2.4	24

The key aspect of the process is pH control. The classic strategy to control the pH involves the continuous dosing of chemical compounds, such as sodium and potassium hydroxide; with a significant impact on running costs, thus making the whole process less attractive. To cope with this limitation, the feasibility of managing the pH of the fermenter through the recirculation of part of the digestate coming out of the methanogenic has been applied. The digestate is a solution rich in alkalinity deriving in particular from the bicarbonate ion. The recirculation ratio was daily established through a control method based on inputs from a system of online probes; the probes-system consisted in a pH meter in the fermentation reactor, a pH meter and a conductivity probe in the digestion reactor.

The managing of the pH of the fermenter by recirculation of the digestate is by far the most economical option. A low digestate dosage may not guarantee the long-term maintenance of the pH in the desired range and excessive dosage of digestate could lead the accumulation of ammonia in the digestion reactor beyond the level of tolerability of the methanogenic step with a consequent shift of the process towards fermentative reactions rather than methanogenic reactions.

The digestate recirculation method operates through two control levels: the first control level (1CL) manage the pH in the fermentation reactor while the second control level (2CL) manages the ammonia concentration in the digester reactor as detailed described elsewhere (Pavan et al., 2018).

The process was maintained in operation for about 300 days and two different configurations were tested: in the first configuration (A), the whole mass flow produced in AnF was used as feed for AnD reactor; in this way, hydrogen and methane were the main mass flows produced in the entire system (Fig. 2A). In the second configuration (B), a portion of mass flow produced in AnF was processed by solid/liquid separation unit; therefore, the AnD reactor was fed with a mixture of fermentation liquid (from the first phase of the process), and the solid-rich mass flow produced from the solid/liquid separation unit. This second configuration simulates a two-phases anaerobic digestion process optimized for VFA and hydrogen-methane production (Fig. 2B).

The overall operation period was divided into three consecutive runs, as reported in Table 3. The first configuration was tested during Run1 and Run2, with food waste obtained by different pre-treatment. Run1 was characterized by the use of organic waste after wet-refine pre-treatment and Run2 by the use of organic waste after screw-press pre-treatment. The second configuration simulating the two-phases process was tested in Run3, with the organic waste after screw-press pre-treatment.

The applied operating conditions have been summarized in Table 3. The first fermentation stage had a hydraulic retention time (HRT) of 3.3 days and an organic loading rate (OLR) of 19.0 kg TVS/(m^3d). In the methanogenic stage, a HRT in the range 12.5–12.6 days and an OLR of 4.0–4.2 kg TVS/(m^3d) were applied.

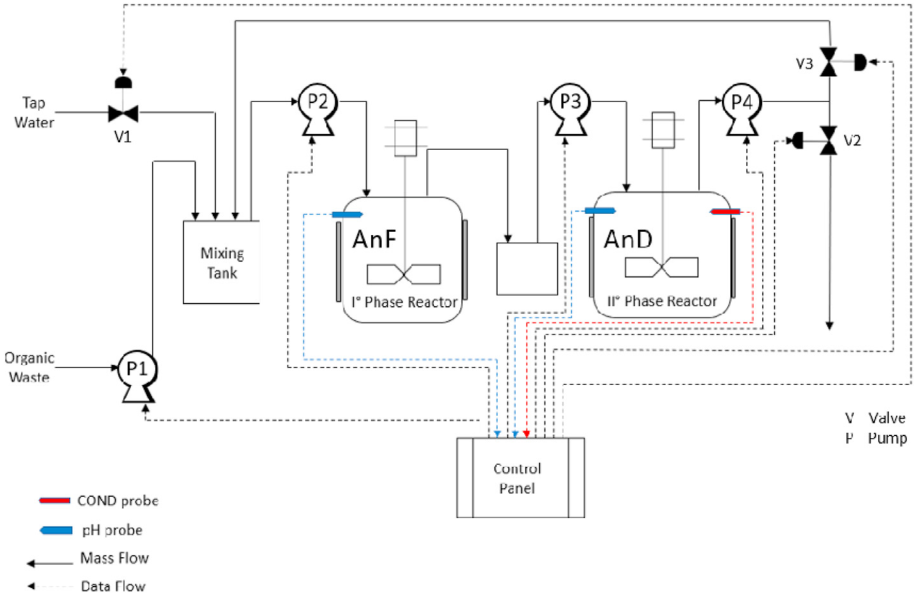

Fig. 1. Pilot-scale control device developed for food waste treatment and volatile fatty acids and hydrogen-methane production.

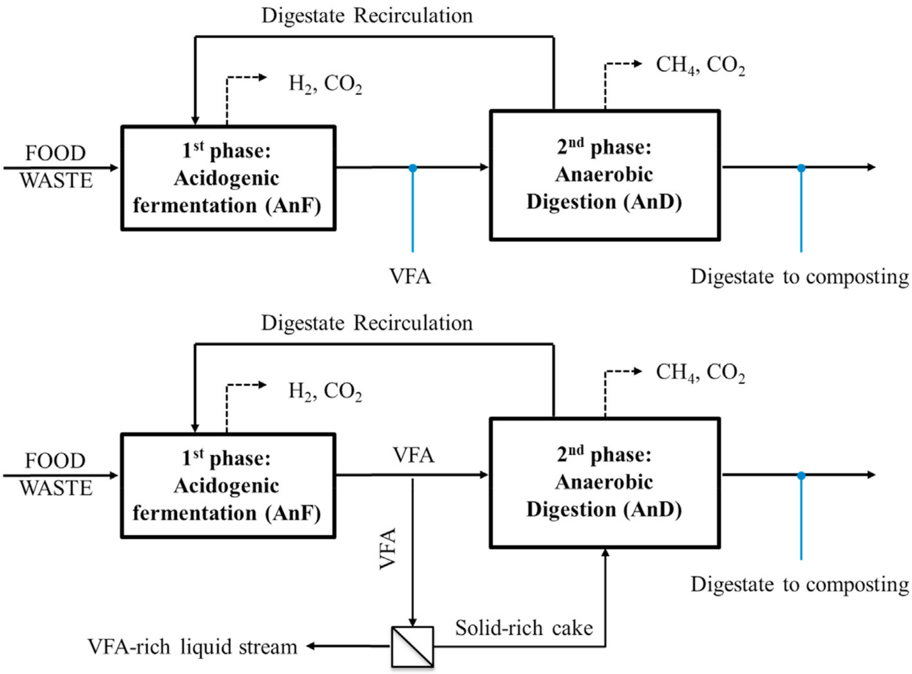

Fig. 2. A. Process configuration A for the production of bio-hydrogen and bio-methane from food waste valorisation.
B. Process configuration B for the production of VFA, bio-hydrogen and bio-methane from food waste valorisation.

Table 3
Operating conditions of the three runs having different process configurations and/or different pre-treated food waste.

Run	Food waste pre-treatment	First fermentation phase			Second digestion phase				Configuration
		HRT	OLR	T	HRT	SRT	OLR	T	
		d	kg TVS/(m^3d)	°C	d	Range d[a]	kg TVS/(m^3d)	°C	
Run1	Wet-Refine	3.3	19	55	12.6	21–31.5	4.0	55	A
Run2	Screw Press	3.3	19	55	12.6	21–31.5	4.0	55	A
Run3	Screw Press	3.3	19	55	12.5	21–31.5	4.2	55	B

[a] RR: 0.4–0.6.

2.3. Analytical methods

The effluents of the reactor were monitored three times per week, for total and volatile solids content, chemical oxygen demand, TKN and total phosphorus. The remaining parameters, namely pH, conductivity, volatile fatty acids content and speciation, total and partial alkalinity and ammonia, were checked daily. All the analyses, except for VFAs, were carried out in accordance with the Standard Methods (APHA/AWWA/WEF, 2012). The analysis of the volatile fatty acids was conducted using AGILENT 6890 N gas chromatograph equipped with a flame ionization detector (T = 200 °C), a fused silica capillary column, DB-FFAP (15 m × 0.53 mm x 0.5 μm thickness of the film). The analysis was conducted by increasing the temperature from 80 °C to 200 °C (10 °C/min), and hydrogen was the gas carrier). The samples were analysed before being centrifuged and filtered with a 0.45 μm filter.

The production of gas for both reactors was monitored by two flow meters (Ritter Company™, drum-type wet-test volumetric gas meters). The percentage of hydrogen and methane was determined by a gas chromatograph GC Agilent Technology 6890N™ equipped with a column HP-PLOT MOLESIEVE™ (30 m × 0.53 m ID x 25 μm thickness of the film), using a thermal conductivity detector (TCD) at a temperature of 250 °C. The injector temperature was 120 °C. There was a constant pressure in the injection port (70 kPa). Samples were taken using a gas-type syringe in 200 μL biogas amounts. Once the entire sample was vaporized, separation of the peaks occurred within the column at a constant temperature of 40 °C (8 min). Argon was used as gas carrier.

2.4. Statistical analysis

The study has evolved to find which model can predict the ammonia content in the anaerobic digester by the measurement of pH and conductivity (supplementary material). Throughout all the database of the experimental long term process, about 300 day-data-set in 3 RUNs of the anaerobic digestion process have been used for the statistical calculations. The statistical analysis was outwardly validated to compare and determine the best possible prediction of the parameter ammonia. These calculations and the statistical analysis were carried out with the aid of the software “R”. Firstly, the values were auto-scaled by calculating the Pearson correlation matrix, then principal component analysis and regression analysis were calculated (Gottardo et al., 2017).

3. Results and discussion

3.1. Wet Refine and Screw Press

The two pre-treated food waste were considerably different showing that the screw pressed food waste was more acid than the food waste from wet refine approach, as demonstrated from the pH value and VFA content. Despite of the major dry-volatile and COD concentrations on wet-refine food waste, the fraction of volatile and COD (based on TS) were significantly higher (t. Test, $p < 0.05$) in the screw-pressed food waste juice (Fig. 3).

By means of the squeezing pre-treatment (Run2 and Run3), the organic fraction in the food waste remarkably increased (TVS/TS ratio equal to 0.93 on average), highlighting that different types of substrate pre-treatment could generate different substrate features. As a consequence, the process performances could be affected by the different kind of food waste pre-treatment and this study has been performed to solve this open question and to understand if the system operability could be adapted to the different feedstock characteristics. Run1 and Run2 were characterized by the different feedstock pre-treatment to verify the response of the control method to the change of feedstock characteristics.

3.2. The control method for pH and ammonia regulation in the two-phase anaerobic process

The two-phases process has been followed for a long operation period and three runs have been separately characterized in terms of performances as a function of the different pre-treated food waste and process configuration. All the three runs lasted about 100 days, which corresponded to 30.0 HRTs for AnF reactor and 8.0 HRTs in Run1, Run2 and Run3 for AnD reactor. Fig. 4A shows the pH of the AnF reactor together to the adopted RR. The three runs are visible from the two vertical axes at 100 and 200 days approximately.

Fig. 4A clearly shows how the 1CL allowed to maintain the pH value above 5, the necessary condition to apply the 2CL for the determination of the accurate RR calculated by equation (I). In general, the minimum pH value to produce VFA and hydrogen from food waste is 5.2 (Lee et al., 2014). The 1CL was adopted just in the first days of running Run1 (start-up) and in the early days of Run2. The high RR of the start-up was due to the slight drop of the pH, at the beginning of the acidification process. When the screw-pressed food waste was fed to the AnF reactor (Run2), the control algorithm was activated as to recirculate the maximum ratio (1.0), since the pH in the fermenter was dropped below the value of 5.0 (days 100–110). In both cases, the 1CL allowed to quickly re-establish the desired acidity condition. This evidence is in line with what expected: the rapid increase in alkalinity in AnF, due to the rise in pH above the lower limit of 5.2 (obtained in the two start-up phases of Run1 and Run2) allowed the control to manage the process adapting the recirculation flow rate (2CL).

Regarding the AnD reactor, Fig. 4B shows the evolution of the total ammonia concentration.

The 2CL has effectively managed the ammonia content in the AnD reactor, as shown by the observed average value (analytically determined), which has settled at 0.756 g/L (the set point value assigned was 0.75 g/L). The increase in the RR in the early days of Run2 has increased the concentration of ammonia in the AnD reactor. At the end of day 110, when the 1CL was switched off, the system had dynamic recirculation according to the calculation through the modelling algorithm and the ammonia values were around 0.750 g/L for the entire duration of Run3. When the optimal working condition of the AnF was re-established (after the start-up of Run2), the production of VFA which lead as a consequence the unavoidable acidification of AnF did not create instability in the whole system. The merit of this process control strategy was due to the model. The model optimally predicted the concentrations of ammonia in the AnD reactor allowing its management in the system accurately. The control method that has been developed consists of a mathematical model (equation (1)), which evaluates the ammonia concentration from pH and conductivity values.

$$[NH_3]_{Pred} = - 682.2\ (pH_{a2}) + 235.5\ (C_{b2}) + 3874.9 \qquad (1)$$

As proof of this, Fig. 5 compares the mathematical model that have been used to predict the ammonia concentration and the analytically measured ammonia concentration values.

By comparing the measured ammonia concentration with the ammonia concentration values predicted by the model, it was possible to calculate the Standard Deviation Error in Prediction (SDEP) and the variance percentage explained by the predictive model (Q^2). These parameters were then compared with those taken from the three models reported in Micolucci (Micolucci et al.,

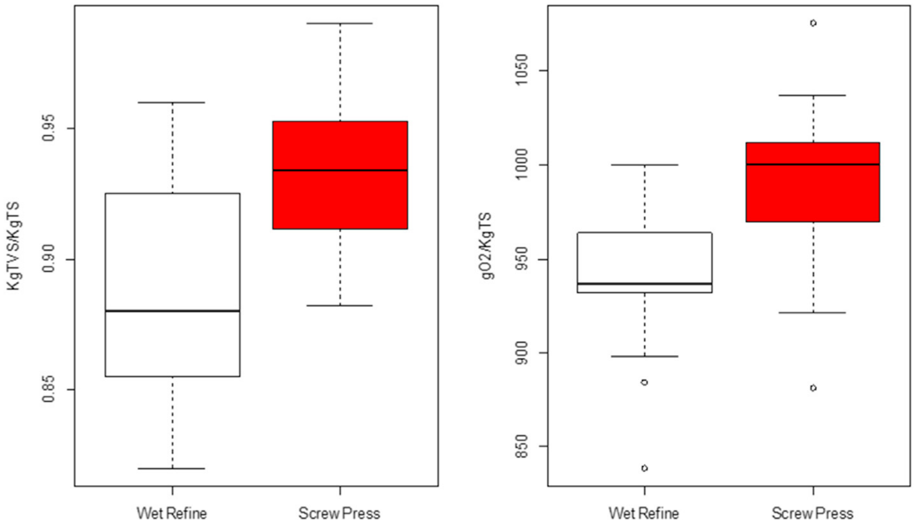

Fig. 3. Box - Plot TVS and COD on a dry basis of the food waste pre-treated with Wet Refine and Screw Press.

2014), where the ammonia concentration was defined as a function of conductivity, VFA, total alkalinity and pH, as depicted in Table 4 (Micolucci et al., 2014). Table 4 summarizes the SDEP and Q^2 values for the model described in this study and the models previously designed, demonstrating that the new model has a much better prediction capacity.

The used control system maintained a recirculation ratio of 0.52 ± 0.01, except for the start-up phase of Run1 and Run2, with a maximum and minimum value of 0.69 and 0.41. In the previous study (Micolucci et al., 2014), the average recirculation ratio was similar but its trend was characterized by larger fluctuations (as shown by the minimum and maximum values of 0.0 and 0.7) and more prolonged over time.

In Micolucci et al. (2014), the ammonia content in AnD was lower than the one recorded in the present experimentation (0.64 gNH_3/L vs 0.71 gNH_3/L) but both in the limit of possible condition for the inhibition of the methanogenic component (approximately above 0.7 gNH_3/L) (Angelidaki and Ahring, 1993). The average pH of AnF reactor was equal to 5.2, lower than the one detected in the present work (5.5 ± 0.9) and coincident with the lower limit of the control (1CL). Both parameters (ammonia in AnDpH in AnF) have made the control of the system particularly difficult to be applied, forcing the operator to continuously adjust one of the two parameters and, as a consequence, negatively affecting the other. Another previous study showed a simplified system where the only operating condition that allowed the process to remain stable was the RR, which was shifted between 0.5 and 0.7 (Gottardo et al., 2017). Given the relatively high recirculation ratio adopted, the ammonia content in AnD reactor was higher (0.8 gNH_3/L) compared to this study, and above the threshold of a possible inhibition level of the methanogenesis.

3.3. Bio-hydrogen, bio-methane and bio-products

Fig. 6 shows the trends of the three products of the process: hydrogen, VFA and methane expressed as specific hydrogen production (SHP, m^3 H_2/kg TVS_{fed}), VFA yield (COD_{VFA}/COD_{fed}) and specific methane production (SMP, m^3 CH_4/kg TVS_{fed}).

Focusing on the VFA yield and SHP, observed results in the three experimental runs remained constant. The use of different food waste pre-treatment and/or process configuration did not affect the process performances, which appeared quite robust and stable as the structured control method ensured. In the particular case of SMP trend, it was also observed a stable trend in all the three runs; however, the average value quantified in Run3 (0.48 ± 0.02 m^3 CH_4/kg TVS_{fed}) was lower if compared with Run1 and Run2 (0.55 ± 0.02 and 0.54 ± 0.02 m^3 CH_4/kg TVS_{fed} respectively). Even though methane production was not affected by the change of food waste pre-treatment (as observed for hydrogen and VFA production), the change of process configuration from A to B negatively affected methane production in AnD reactor. This was expected since the introduction of a solid/liquid separation unit between AnF and subtracted a portion of the VFA flow. The solid-rich overflow (solid cake, SC) obtained from the centrifugation (C) unit was used as the substrate for AnD reactor, together to the other fraction of VFA-rich stream not subjected to the C process unit. The mixture between the two components was adjusted to maintain the same OLR (in terms of TVS) as it was in Run1 and Run2. The lower VFA level in the SC justified the lower methane production in Run3. In return, configuration B allowed recovering stream rich in VFA (amenable for other purposes), differentiating the type of bio-products obtainable from the treatment of food waste with respect to biogas or hydrogen and methane.

Compared to previous studies, the current experimentation showed significantly higher specific production for VFA and hydrogen; in a minor extent, also the specific methane production was increased, with the exception of Run3 value (ANOVA, Duncan - Waller post-hoc test, $\alpha = 0.05$; Table 5). The lower SHP and VFA yield of AnF reactor in the two previous studies may be related to the lower pH (below 5.3) compared to the present one (5.5 ± 0.9) and in general a more stable process.

Table 5 also shows a marked improvement in the performance of the AnD second stage in the present trials Run1 and Run2 (ANOVA, Duncan - Waller post - hoc test, $\alpha = 0.05$). This was a consequence of a better fermentation process management carried out in this study, in particular for the pH value, whose fluctuations were less frequent and of minor changes compared to those previously recorded (Micolucci et al., 2014)(Gottardo et al., 2017). The importance of an efficient control for the stability of the fermentation performances and its related impact on the AnD performance (the conversion efficiency of the COD into biogas) was previously discussed [3].

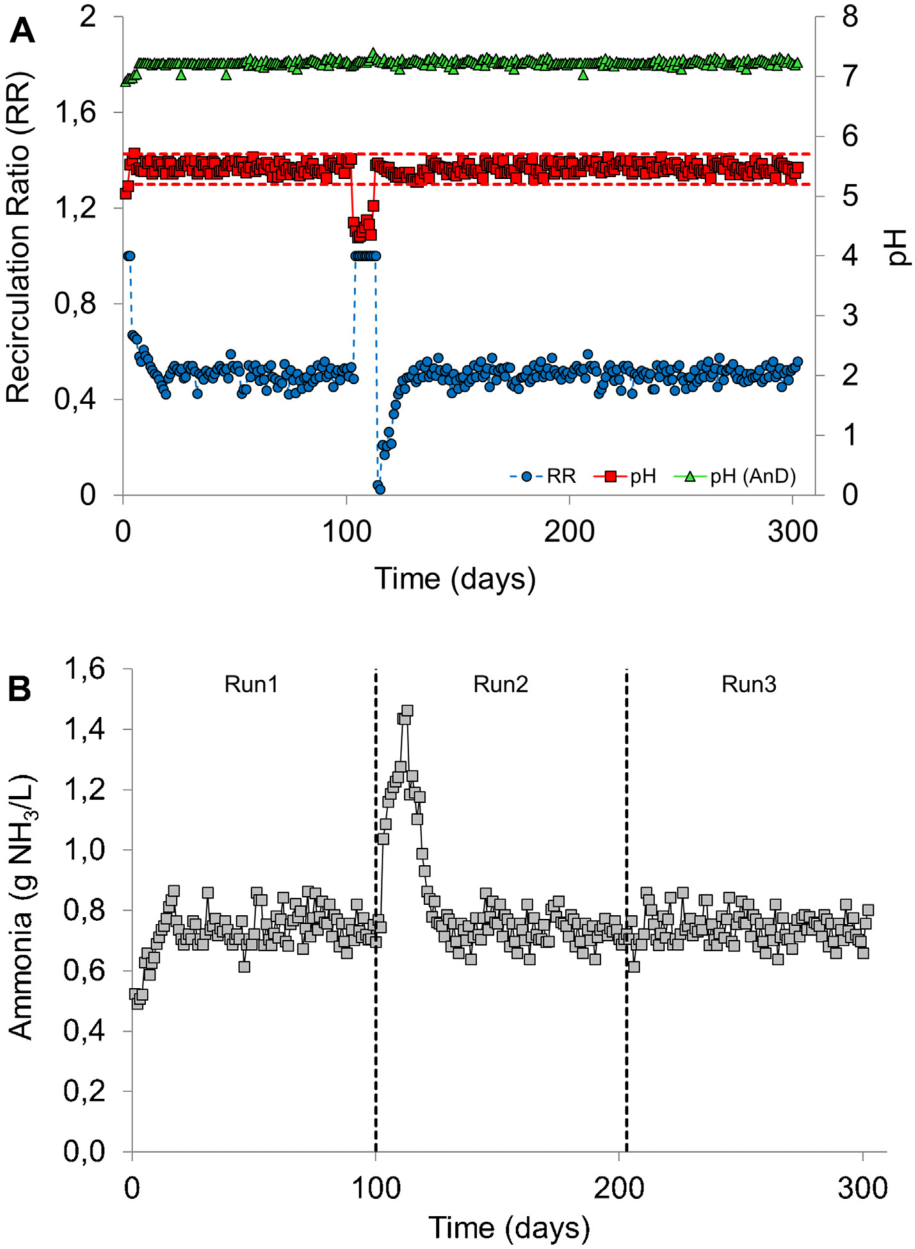

Fig. 4. Trends of pH value monitored in AnF reactor and of the adopted RR (A); Evolution of the total ammonia concentration in the AnD reactor (B).

3.4. Mass balance of the system

The mass balance assessment was performed for the process configuration B, the only one that allowed to produce hydrogen and methane and to separate a VFA-rich effluent for parallel purposes. Each data related to the characterization of the process influents/effluents produced biogas and the efficiency of the solid removal in the solid/liquid separation step has been taken into account (Fig. 7). Almost 30% of the COD fed to AnF reactor was converted into VFA with (acidification yield of 0.31 g COD_{VFA}/g COD_{fed}), in line with previous thermophilic experiences utilizing different food waste mixture (Valentino et al., 2018)(Valentino et al., 2019). A little portion of COD (~3%) was converted into the gas mixture (whose composition was 43% H_2, 51% CO_2 and 6% CH_4) and the largest part remained as particulate COD (~65%). The fermented liquid stream was equally split into two portions (30 kg/d each). One of these portions was subsequently subjected to the solid/liquid separation unit, which generated two additional streams: the low-solid mass content flow (LS) and the high solid content flow (solid cake, SC). The LS flow was characterized by an average VFA level of 21.9 ± 0.4 g COD_{VFA}/L, consisting of 25% acetic, 19% propionic, 33% butyric acid and the remaining 23% of valeric, caproic and heptanoic acids (COD basis); a COD_{VFA}/COD_{SOL} ratio of 0.81 ± 0.3 was also quantified. These characteristics made it possible to obtain a noble substrate for other purposes within the biorefinery concept, such as PHA. As recently demonstrated (Moretto et al., 2020) the quality of the substrate produced in term of %VFA supports the required loads (3.0–8.0 kg COD_{SOL}/m^3 d) and the so-called feast-famine strategy (feasible at high COD_{VFA}/COD_{SOL} ratio) in the typical sequencing batch reactor (SBR) designed for the production of PHA-producing biomass selected from waste activated sludge (Rodriguez-Perez et al., 2018). Production of 0.24 kg COD_{PHA}/d can be estimated,

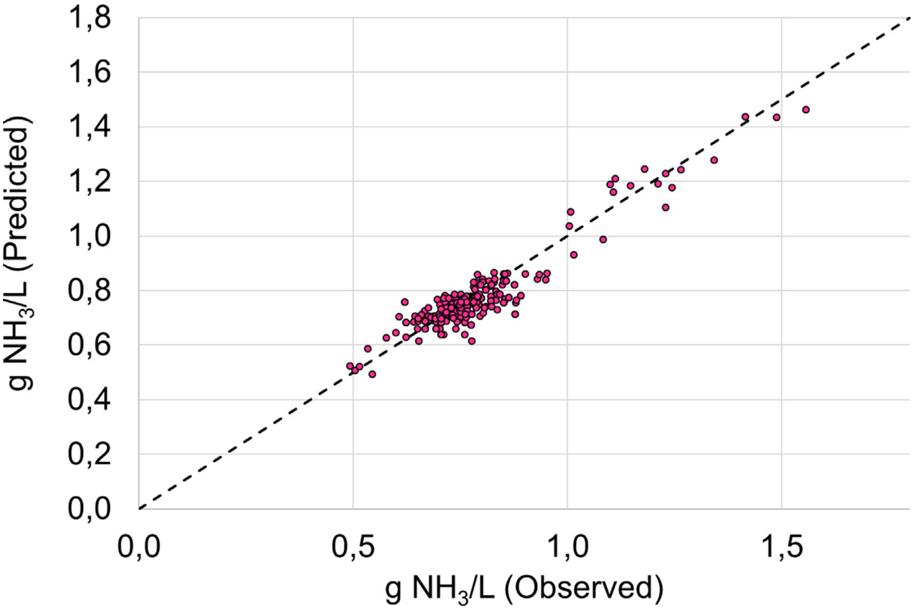

Fig. 5. Comparison of the concentration of ammonia observed and predicted by the model over the entire period of process operation.

Table 4
Comparison of the predictive performance of the model calculated in this study with the previous models discussed in Micolucci et al. (2014).

Model	SDEP	Q^2	References
First model: AMM = f (COND)	0.202	0.78	Micolucci et al. (2014)
Second model: AMM = f (COND, VFA, Total Alkalinity)	0.116	0.93	Micolucci et al. (2014)
Third model: AMM = f (COND, Total Alkalinity)	0.121	0.92	Micolucci et al. (2014)
New model: AMM = f (COND, pH)	0.048	0.99	This study

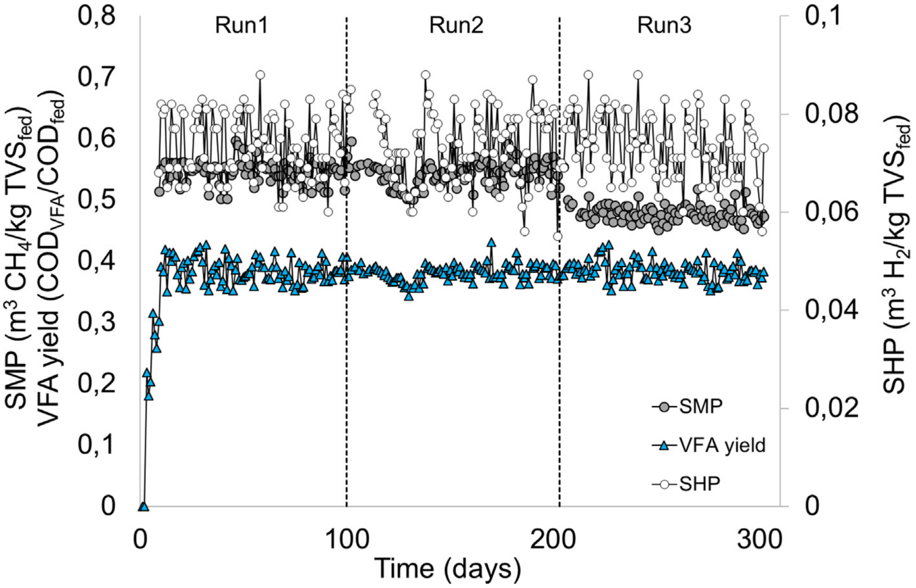

Fig. 6. Specific hydrogen and methane production (SHP and SMP), VFA yield in three runs.

based on the recently reported PHA yield of 0.35 COD_{PHA}/COD_{SOL} (Valentino et al., 2018) and the available COD_{SOL} (0.68 kg COD_{SOL}/d). The anaerobic digester was fed with a load of 4.2 kg TVS/(m^3d) and with a HRT of 12.5 d; the feed was composed by the SC stream overflow and by the portion (30 kg/d) of AnF effluent. The stability parameters confirmed the effectiveness of the approach and the robustness of the process: partial and total alkalinity (P.Alk and T.Alk) had average concentrations of 3.28 ± 0.07 and 5.25 ± 0.05 g $CaCO_3$/L; VFA/P.Alk ratio and pH were respectively equal to 0.18 ± 0.03 and 8.1 ± 0.1.

Considering the system as a whole, the biogas produced had an average composition equal to 53 ± 1% in CH_4, 41 ± 1% in CO_2 and

Table 5
Specific productions and results of the Duncan - Waller post-hoc test. For the averages with different quotes, the null hypothesis is rejected.

Specific Hydrogen Production (SHP; m^3 H_2/kg TVS_{fed})	VFA yield (kg COD_{VFA}/kg COD_{fed})	Specific Methane Production (SMP; m^3 CH_4/kg TVS_{fed})	Ammonia NH_3 (g/L)	Reference
0.060 ± 0.010	0.17 ± 0.04	0.47 ± 0.02	0.64 ± 0.06[a]	Micolucci et al., 2014
0.068 ± 0.010	0.18 ± 0.01	0.50 ± 0.01	0.68 ± 0.05[a]	Gottardo et al., 2017
0.074 ± 0.005	0.32 ± 0.02	0.55 ± 0.02	0.71 ± 0.02[b]	Run1
0.070 ± 0.010	0.32 ± 0.01	0.54 ± 0.02	0.82 ± 0.19[b]	Run2
0.074 ± 0.009	0.31 ± 0.01	0.48 ± 0.02	0.70 ± 0.06[b]	Run3

[a] methanogenesis reactor (2nd stage).
[b] AnD.

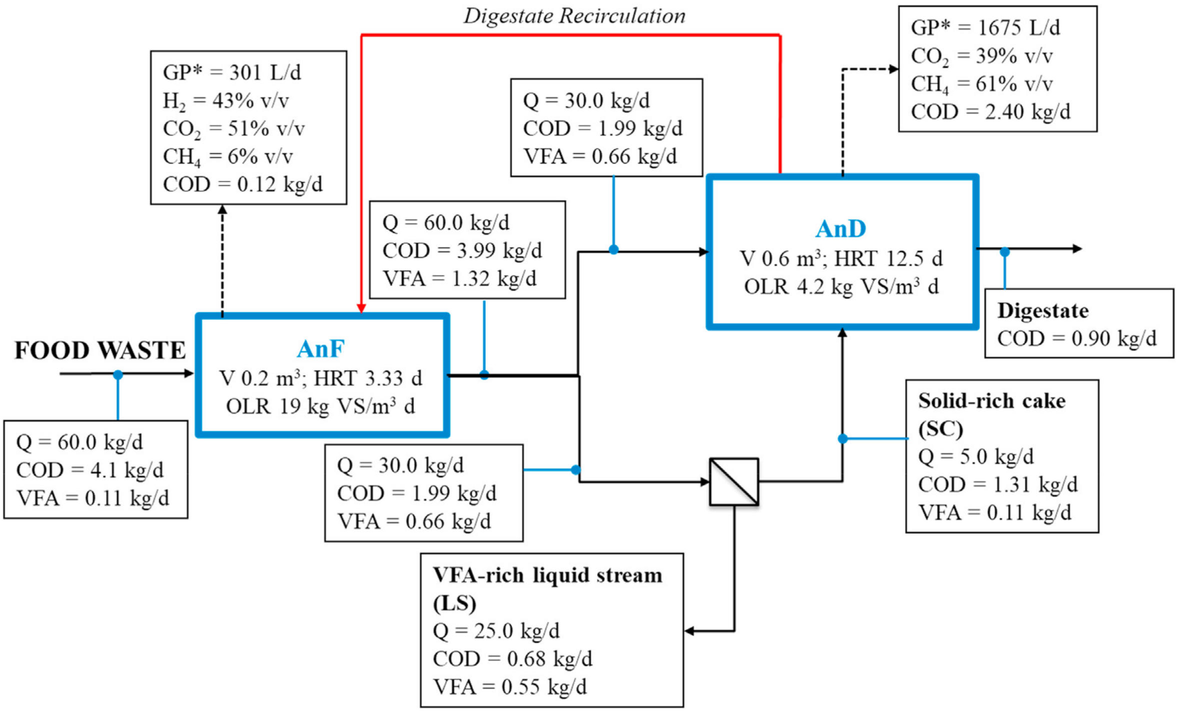

Fig. 7. Mass balance assessment for the two-stages process*
GP: gas production.

7.0 ± 0.3% in H_2. The designed pilot-scale system achieved both objectives in terms of hydrogen and methane and VFA production. As solely applied for process configuration B, the removal of a part of organic matter in a soluble form (by applying the solid/liquid separation step) from the methanogenic reactor, did not lead to a remarkable reduction in biogas production compared to the tradition two-phases process (configuration A). The availability of a valuable VFA-rich stream can give a realistic opportunity to recover bio-products (e.g. polyhydroxyalkanoates) with higher market value if compared to biogas and/or compost.

4. Conclusions

The use of chemometric tools and statistical analysis allowed to obtain an effective model and to predict some necessary parameters (e.g. ammonia concentration) for the control and the optimization of the two-stage anaerobic systems also when differently pre-treated organic wastes were treated, demonstrating the robustness of the system.

The approach proposed in this study has the following novelty features:

- automatic management of the fermentation process for the stabilization in the event of a shock, maintaining optimal process conditions (in particular, pH values above 5.2 in AnF), which normally requires manual maintenance and analysis by a person responsible for the process;
- the simplicity of the probes' utilisation for continuous online monitoring of the process parameter gives resiliency and capacity to predict ammonia concentration online of the anaerobic process;
- the estimation of ammonia concentration carried out with a mathematical validated model which has shown good capability in prediction;

The application of the proposed two-levels control system has allowed improving the production performance of the AD process in separate stages, compared to those obtained in the past (Gottardo et al., 2017)(Micolucci et al., 2014), while maintaining a high process resilience and a fast-automatic restoration capacity due to external forces stress conditions. Overall, in the frame of food waste management, this approach can be considered new know-how to change the paradigm of waste treatment plants into real bio-products production processes. In particular, for organic waste management, the implementation of controlled VFA production is one of the most pioneering strategies that could bring the applied research of the pilot platform to a more detailed technical-economical evaluation of the technology and consequential market exploitation.

CRediT authorship contribution statement

Federico Micolucci: Conceptualization, Methodology, Investigation, Formal analysis, Data curation, Software, Resources, Validation, Writing - original draft. **Marco Gottardo:** Conceptualization, Methodology, Investigation, Formal analysis, Supervision. **David Bolzonella:** Supervision, Project administration. **Paolo Pavan:** Visualization, Supervision, Project administration, Funding acquisition. **Mauro Majone:** Supervision, Project administration. **Francesco Valentino:** Conceptualization, Data curation, Writing - review & editing, Visualization.

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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