

Process Performance of Thermophilic Anaerobic Co-Digestion of Municipal Sewage Sludge and Orange Peel

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ABSTRACT

In the present study, the process performance of anaerobic co-digestion of municipal sewage sludge and orange peel (OP) was evaluated. The experiment was conducted in batch mode under thermophilic conditions (55 °C). It involved adding 1.5 and 3.0 g of OP to reactors R2 and R3, respectively. In R1 (control reactor), the mono-digestion of sewage sludge was conducted. The obtained results indicated that the application of OP led to deterioration of process efficiency. Decreased methane and biogas productions were noticed at both doses of OP. The average values of methane production were 518.9, 416.8 and 458.6 mLCH₄·g⁻¹VS in R1, R2 and R3, respectively. The declining tendency on the biogas and methane rates was also observed. The application of OP resulted in degradation of the stability parameters. The negative effect of OP application was related with the presence of inhibitors such as ammonia nitrogen, volatile fatty acids, limonene and phenol, importantly their increased contents were observed in R2 and R3. Moreover, the thermophilic conditions are not recommended for the anaerobic co-digestion of those substrates, because they might accelerate the inhibition phenomenon.

Keywords: anaerobic digestion, biogas production, batch reactors, limonene, orange wastes.

INTRODUCTION

Currently, the food industry is recognized as a main economic sector that contributed to progressive climate change. It is responsible for a major greenhouse gas emission, waste generation, as well as significant water and energy consumption. Moreover, the constantly increasing population promote a substantial development of this sector (Lund-Durlacher and Gossling, 2021). An important part of this industry is represented by juice processing, which is dominated by orange beverage (Ortiz et al., 2020; Chen et al., 2022). It is estimated that about 70% of global cultivated oranges are used in the manufacturing of juices and other preserves (Terzioglu et al., 2021). Within their processing, about 50% of fruit becomes waste, mostly represented by orange peel (Verma et al., 2020; Carranza-Mendez et al., 2022). It mainly consists of a hemicelluloses, cellulose, lignin, soluble sugars, pectin, flavonoids and essential oils (Yaradoddi

et al., 2022). Moreover, it is characterized by low pH, a significant amount of organic matter and tendency towards rapid deterioration due to high moisture content and presence of fermentable sugars (Ruiz and Flotats, 2014; Tsouko et al., 2020; Carranza-Mendez et al., 2022). Generally, it is considered as a by-product with low economic value, the effective management of which has been a serious problem for many production facilities. This issue is particularly important for low- and middle-income countries, where oranges are produced and processed (Ortiz et al., 2020; Isibika et al., 2021). Moreover, improperly managed OPs might result in water and soil contamination. Therefore, to reduce the negative impact on environment, several attempts have been made to reuse this waste (Hasan et al., 2020; El Gheriany et al., 2020). OP has been applied as animal feed, in the production of biofuels and bioplastics. Furthermore, it has been used in the generation of high added-value compound, such as cellulose, essential oils, enzymes, pectin,

monoterpenes and flavonoids. However, those applications are related with significant costs; additionally, some of them are innovative and have not been implemented on a technical scale. Therefore, a significant amount of OP is still being landfilled (Shan et al., 2016; Carranza-Mendez et al., 2022). Regarding both economic and environmental factors, the application of this by-product in anaerobic digestion process (AD) might be a solution. This technology is known as one of the most sustainable ways to convert various organic wastes into energy and valuable digestate (Suarez et al., 2022). OP might be applied in this technology; however, the presence of organic matter can lead to the accumulation of volatile fatty acids (VFA), thus inhibiting the AD (Bong et al., 2018). On the other hand, OP indicated a significant content of compounds, such as hemicellulose, cellulose, and lignin. It is commonly known that their hydrolysis is a speed limiting step of AD (Li et al., 2017). Moreover, this by-product has low pH, which is unfavourable for the growth of AD microorganisms. However, the major problem in effective AD is the presence of limonene that exhibits antioxidant properties and might lead to inhibition of the biological activity within AD even at low concentration (Bouaita et al., 2022). The above-mentioned factors make it a substrate that is particularly difficult to use in an anaerobic bioconversion. Therefore, in recent years, many studies have been dedicated to improving the AD of this waste, mainly focused on the application of pre-treatment methods to remove limonene, involving steam distillation and ethanol extraction (Martín et al. 2010; Ruiz et al., 2016), biological methods based on fungi enzymes (Akao et al., 1992), centrifugation or aeration of feedstock (Lane, 1983). Moreover, to improve the mono-digestion efficiency of this substrate, the application of advanced reactors and various operating strategies (Wikandari et al., 2014; Tayibi et al., 2021; Awasthi et al., 2022) or even two-stage process (Lukitawesa et al., 2018) and solid state AD (Srilatha et al., 1995) have been reported. However, these methods are related with additional operational and investments costs, often presenting low efficiency. Another path is the application of an additional substrate with complementary composition to OP. This strategy is known as co-digestion, it has been commonly used to overcome the difficulties of mono-digestion of particular substrate. It allows for obtaining differentiated microbial community, improving buffering capacity, balancing C/N ratio in the feedstock, diluting of toxic compounds and

providing the necessary micro- and macro-nutrients for AD (Jiang et al., 2022). The main advantages of this method are increased biogas production and improved process stability. However, successful implementation of this strategy largely depends on selecting a suitable component as well as determining the appropriate operational conditions, such as volumetric ratio, type of inoculum and temperature (Kunatsa and Xia, 2022). Thus far, OP has been co-digested with cow dung (Mandal and Mandal, 1997), organic fraction of municipal solid waste (Forgács et al., 2012; Bouaita et al., 2022), glycerol (Martín et al., 2012), food waste (Anjum et al., 2017), and marine seaweed (Negro et al., 2020). However, many of them resulted in low biogas production and/or showed significant process instability. Importantly, those examples concerned different temperature regimes. It should be noticed that under thermophilic conditions, the inhibitory concentration of limonene is higher than for mesophilic ones (Martín et al., 2010; 2013). Importantly, thermophilic regime is related with the higher operational cost; therefore, this fact should be also considered (Fagbohunge et al., 2016). Nevertheless, under these conditions, a number of benefits might be obtained e.g. enhanced biogas yields, improved volatile solids reduction as well as pathogens destruction. Furthermore, anaerobic digesters operated at this temperature are characterised by reduced reactor capacity, compared to mesophilic regime. On the other hand, the AD at this temperature indicates greater instability associated mainly with VFA accumulation and ammonia inhibition (Moerland et al., 2022). The previous studies demonstrated that application of OP to municipal sewage sludge (SS) might result in enhanced biogas production and may constitute a profitable solution for both wastewater treatment plants (WWTP) and orange processing companies (Serrano et al., 2014; Martínez et al., 2018). However, these studies were performed under mesophilic conditions. Different trend might occur under thermophilic conditions; therefore, the research in this area should be carried out.

The main objective of this study was the evaluation of the efficiency of municipal sewage sludge and OP co-digestion under thermophilic conditions. The process performance was examined based on biogas/methane production, organics removal and content of nutrients. Moreover, the process stability and effect of inhibitors were also analysed.

MATERIALS AND METHODS

Inoculum and substrates

The inoculum was taken from the mesophilic anaerobic digesters located at Hajdów WWTP. After collection, it was screened; then, 1.4 L of this sample was added to each reactor. The acclimatisation of biomass to thermophilic conditions was achieved after 60 days. The detailed characteristic of this sample is presented in Table 1.

In this study, SS was the main substrate. This sample was also obtained from Hajdów WWTP. It was prepared as a mixture of thickened primary and waste sludges, taken

separately. Under laboratory conditions, those were sieved (to remove the particles above 5 mm) and mixed at volumetric ratio of 60:40 v/v. The 0.4 L of prepared mixture was added to laboratory reactors.

OP was used as an additive to main substrate – SS. This sample was obtained from a lab-scale juice processor. Before supplying the reactors, fresh OP was shredded with a blender to obtain the particles below 5 mm. The composition of OP used in this study is listed in Table 1.

Operational set-up

The experiment was performed in a batch mode under thermophilic conditions (55 ± 1 °C). The schematic diagram of the reactors and adopted operational parameters is shown in Figure 1. In this study, to compare the obtained results, the control reactor (R1) was provided; therein the mono-digestion of SS was performed. In reactors R2 and R3, OP was added in the amount of 1.5 and 3.0 g, respectively.

To perform batch test, the BioReactor Simulator (BPC[®]) was used. Each reactor had a total capacity of 2.0 L and is equipped with mechanical stirrer. To keep the established temperature regime, the digesters were placed in a water bath. The volume of generated biogas was monitored continuously, involving additional measuring device working according to the principle of liquid displacement and buoyancy. The data was collected and recorded using web-based software based on a cloud solution.

Table 1. Composition of the orange peel and inoculum used in the experiment (the average values and standard deviation are given)

Parameter	Unit	OP	Inoculum
COD	g L ⁻¹	46.7±2.1	24.7±1.7
SCOD		10.1±0.5	2.3±0.4
ALK		nd*	3551±57
VFA	mg L ⁻¹	907±7.7	301±14.4
TP		483±17.1	456±17.1
TN		93.4±2.4	2033±14.7
N-NH ₄ ⁺		2.06±0.05	49.1±7.4
P-PO ₄ ³⁻		403±15.7	54.7±5.4
Phenol		65.8±3.6	13±3.7
pH	-	3.74±0.01	7.89±0.01
VS	g kg ⁻¹	231.35±0.47	14.82±0.7
TS	g kg ⁻¹	242.5±0.5	24.87±0.64

*nd – not detected.

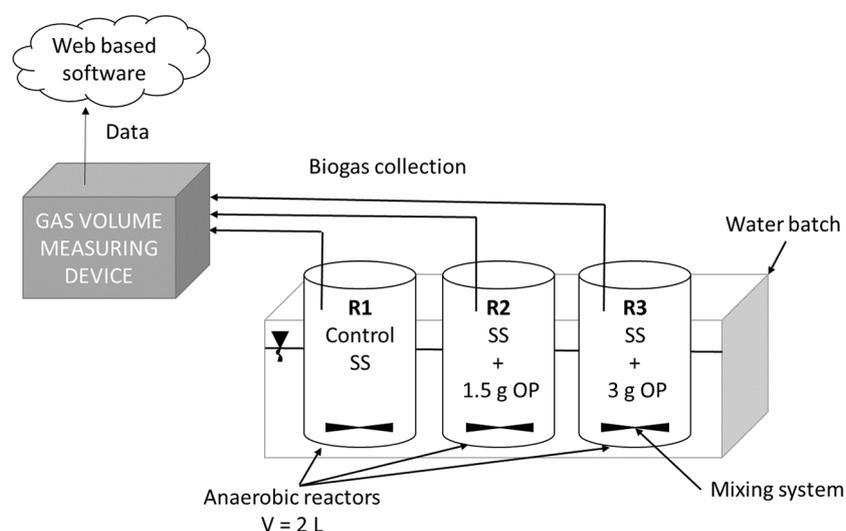


Figure 1. The scheme of operational set-up

Analytical methods

In all applied materials, the following parameters were controlled: total chemical oxygen demand (COD), TS (total solids), VS (volatile solids), total nitrogen (TN) and total phosphorus (TP). In supernatant, soluble chemical oxygen demand (SCOD), VFA, alkalinity (ALK), pH, ammonia nitrogen (NH_4^+-N) and orthophosphate phosphorus ($\text{PO}_4^{3-}-\text{P}$) were monitored.

The composition of feedstock and digestate was controlled according the same scheme, additionally the limonene, p-cymene and phenols were analysed. The composition of substrates and inoculum was established once before the experiment. Each measurement was prepared three times and the presented results are the average value. Most of analyses were performed using standard cuvette tests and UV–VIS DR 3900 (Hach, Loveland, CO, USA). Both TS and VS were established according to the Standard Methods for the Examination of Water and Wastewater (APHA, 2005). The HQ 40D Hach-Lange multimeter was used to monitor the pH values. Both TS and VS were established based on the procedure presented in the Standard Methods for the Examination of Water and Wastewater (APHA, 2005).

To evaluate the limonene and p-cymene contents, 0.5g of the sample was diluted with MilliQ water; then, it was mixed for 30 min at temperature of 35 °C. The prepared sample was extracted using SPME PDMS 100 μm for 10 min. Then, it was injected into a gas chromatograph (Trace GC Ultra PolarisQ, Thermo Electron, Italy) using helium as a carrier gas with the flow of 1.2 mL min^{-1} equipped with a Supelco Equity 5MS capillary column (30 m \times 0.25 mm ID \times 0.25 μm). The inlet temperature was 40 °C, then it was raised by 5 °C \cdot min^{-1} to reach the level of 250 °C kept for 3 min. The composition of biogas was controlled by the same gas chromatograph. For the analyses, divinylbenzene (DVB) packed columns (RTQ-Bond) were applied. The operational parameters were: 50 °C for the injector and 100 °C for the detector. Helium was used as carrier gas, with a flow rate of 1.5 mL \cdot min^{-1} .

The influence of substrate application on process performance the removal efficiencies (η) of VS, TS, COD were established. For this purpose the following equation was used:

$$\eta = \frac{F - D}{F} \cdot 100\% \quad (1)$$

In turn, for SCOD and $\text{PO}_4^{3-}-\text{P}$ a release rate (η_r) was determined.

$$\eta_r = \frac{D - F}{F} \cdot 100\% \quad (2)$$

where: F – content of corresponding parameter in the feedstock, mg L^{-1} or g \cdot kg^{-1} ;

D – content of corresponding parameter in the digestate, mg L^{-1} or g \cdot kg^{-1} .

In the preset study, the biogas and methane production rates (GPR/MPR) were also evaluated using equation:

$$\text{GPR/MPR} = \frac{\text{GP/MP}}{t} \quad (3)$$

where: GP/MP – cumulative biogas/methane production (mL $\text{CH}_4 \cdot$ g^{-1} VS);

t – digestion time (21 \cdot d).

RESULTS AND DISCUSSION

Removal efficiency of organic compounds

To evaluate the influence of co-substrate application on process performance in terms of organic compounds, the following parameters were analyzed in feedstock and digestate: VS, TS, COD and SCOD. Moreover, the removal efficiencies or release rate of the afore-mentioned parameters were also determined. As is shown in Figure 2, the application of both doses of OP has no evident effect on the TS and VS contents in the feedstock, comparable results to control reactor was found. A different trend was observed with regard to COD and SCOD (Figure 2c, 2d). In this case, the supplementation of feedstock resulted in improvement of the afore-mentioned parameters, increasing with the dose of OP. The observed changes resulted from the composition of the OP rich in these components (Table 1). For COD, enhancements of 26 and 29% were found in R2 and R3, respectively. In turn, for SCOD these were 24.3 and 62.5% in R2 and R3, respectively.

Only at minor dose of OP the VS removal increased from 61.2% (control) to 65.9% (R2). A similar trend occurred also for TS (Fig. 2b). In turn, the application of addition of a higher dose of this substrate led to a slight decline of both TS and VS removals. It might be related with possible overloading of reactor, as well as increased concentration of inhibitors such as ammonia nitrogen, limonene and phenols (Table 2). In turn,

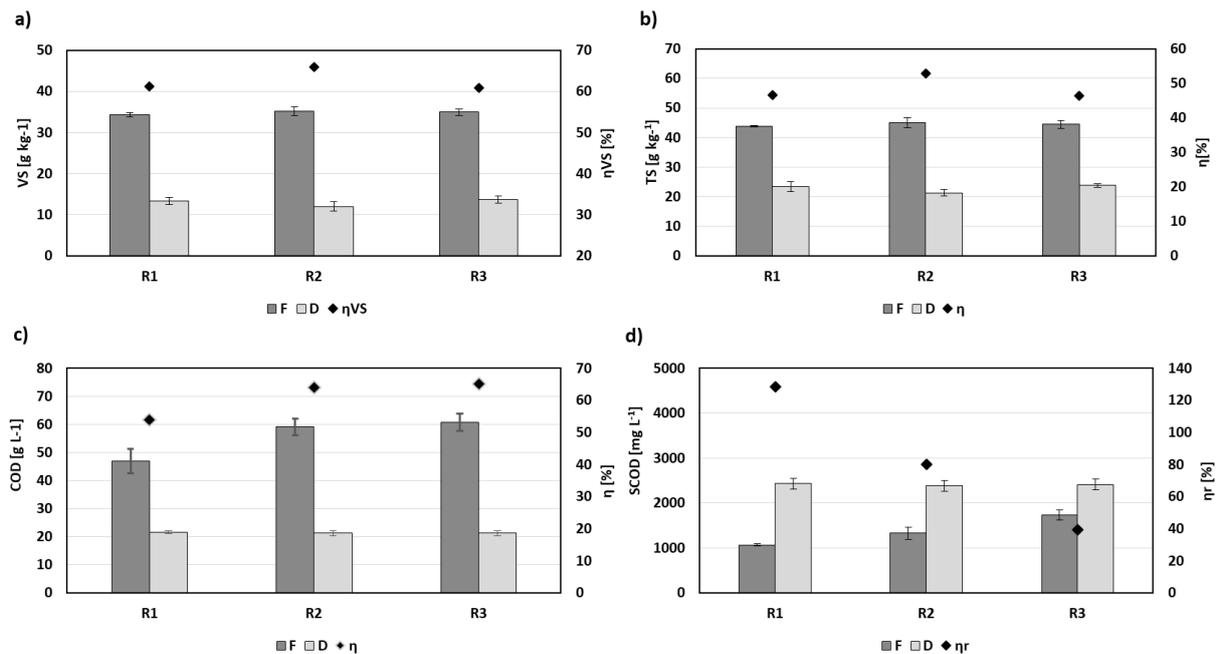


Figure 2. Concentration of VS, TS, COD and SCOD, in feedstock (F) and digestate (D), as well as related removal efficiencies (the average values and standard deviation are given)

with regard to COD at both co-substrate doses, enhanced removal efficiencies were achieved. A different declining tendency was observed for SCOD; therein, the application of OP led to reduction of release rate by 37.5 and 69.2% in R2 and R3, respectively.

Process stability, contents of inhibitors and nutrients

The pH value, ALK, VFA as well as VFA/ALK ratio were used to evaluate the process stability (Table 2). The implementation of feedstock in OP contributed to deterioration of feedstock quality in terms of these parameters. In co-substrate presence, the reductions of pH values and ALK content were found. Simultaneously, the increase in VFA concentration was observed. The observed changes resulted from characteristic of OP (Table 1). These changes are highly unfavourable, because they might lead to disturbances of process performance (Chen et al., 2008). The enhanced content of VFA might inhibit the activity of methanogens, resulting in poor biogas production (Zhang and Wang, 2021).

Within AD, the pH value, as well as the ALK and VFA contents increased. It should be noticed that as compared to control, the VFA content in co-digestion series was significantly enhanced by 36 and 54% in R2 and R3, respectively.

Nevertheless, the inhibition of the cellulolytic activity is observed at VFA concentration above 2000 mg·L⁻¹ (Siegert and Banks, 2005). Moreover, the previous studies indicated that under thermophilic conditions, the improved VFA production is observed (Hao and Wang, 2015; Chen et al., 2017).

It is worth mentioning that in all series, the ALK content in digestate was established at high level providing in this way a sufficient buffering capacity within AD. Importantly, in all reactors, the VFA/ALK ratio was at a low level below 0.2–0.3, indicating stable process performance (Ayodele et al., 2022). However, an increase of this ratio was observed in OP presence. The average values were 0.09, 0.12 and 0.13 in R1, R2 and R3, respectively. Additionally, the observed pH values in all series were within a preferred range for AD (pH 7.2–7.8) (Romano and Zhang, 2008). However, as compared to SS mono-digestion, slightly decreased values of pH were found in co-substrate presence. To conclude, the aforementioned parameters were within the required level in all series, indicating process stability. However, a negative effect on these indicators was observed in OP presence.

Ammonia nitrogen is known as a major inhibitor of microbial activities in AD. It is known that especially the excess free ammonia blocked acetate metabolism, leading to the accumulation of VFA, finally resulting in AD failure (Liu et al.,

Table 2. The characteristics of feedstock (F) and digestate (D) in corresponding series (the average values and standard deviation are given)

Parameter	Unit	Feedstock (F) / Digestate (D)	Series		
			R1	R2	R3
ALK	mg L ⁻¹	F	845±42	831±27	818±18
		D	5706±78	5672±52	5713±82
VFA	mg L ⁻¹	F	258±2.5	309±1.7	421±4.2
		D	486±3.7	661±5.1	749±8.7
TP	mg L ⁻¹	F	428±7.8	425±3.9	455±5.2
		D	563±12.5	564±15.7	601±13.4
TN	mg L ⁻¹	F	1519±51	1695±29	1847±45
		D	1761±25.7	1845±36.1	2106±33.5
N-NH ₄ ⁺	mg L ⁻¹	F	55.2±2.5	55.4±1.7	58.9±4.7
		D	1039±10.5	1089±11.2	1267±4.52
P-PO ₄ ³⁻	mg L ⁻¹	F	50.1±1.2	54.1±3.4	50.1±2.4
		D	82±7.5	80.6±7.2	121±8.7
Phenol	mg L ⁻¹	F	2.95±0.2	5.03±0.3	7.45±0.15
		D	27.9±1.5	23.4±1.7	33.8±3.4
pH	-	F	6.5±0.01	5.59±0.02	5.96±0.01
		D	7.8±0.01	7.72±0.01	7.68±0.02
limonene	ppb	F	29.3	1760	1832
		D	22.3	440	982
p-cymene	ppb	F	13.8	165.8	173.1
		D	63.9	66.6	71.5

2021). This compound is particularly problematic during processing of the wastes with significant nitrogen content; in this case, high concentrations of ammonia nitrogen will appear within the AD process. It should be pointed out that the application of OP increased the N content in feedstock; major changes were observed at 3.0 g of co-substrate. Moreover, the thermophilic conditions might also promote the inhibition by this indicator. This fact is related with enhanced activity of microorganisms at high temperatures (Zhang et al., 2019; Cai et al., 2021). As it is shown in Table 2, the NH₄⁺-N released within AD; the growths of 18.8, 19.7, 21.5 – fold were observed in R1, R2 and R3, respectively. Importantly, its concentration in digestate increased with the OP dose. Moreover, in all series the NH₄⁺-N concentration exceeded the limit value of 550 mg·L⁻¹ that might lead to disturbance in AD performance (Yan et al., 2020). In the present study, the observed ammonia inhibition resulted in decreased methane production rate (Table 3) as well as formation of the AD intermediates, such as VFA (Table 2) (Rajagopal et al., 2013).

However, the main problems with the application of citrus wastes in AD is related with high content of essential oils, mainly limonene (Ruiz

and Flotats, 2016; Calabro et al., 2020). The previous studies indicated that its inhibitory effect is related with p-cymene generation during transformation of citrus wastes within AD. The antimicrobial activity of those components is related with its interaction with the cell membrane (Bakkali et al., 2008; Ruiz and Flotats, 2014, Ruiz et al., 2016). The previous studies reported that the enhanced temperature improved the endurance of cells against limonene (Karatzas et al., 2000). In turn, some of them stated that under thermophilic conditions diffusion of essential oils is higher, which might lead to faster process disturbance. Currently, there is no clear tendency whether a higher temperature in AD of citrus wastes is beneficial. Moreover, the inhibitory effect of essential oils is dependent on a several factors, such as pH, water activity, adaptation of microorganisms, and type of inoculum (Ruiz and Flotats, 2014). In the present study, as compared to control, the application of OP resulted in a significant increase of both limonene and p-cymene contents in the feedstock. Importantly, in co-digestion series during AD, both components were degraded. The removal efficiencies of limonene were 23.9, 75 and 60.9% in R1, R2 and R3, respectively. Increasingly, the

p-cymene content was reduced by approx. 60% only in R2 and R3. A different trend occurred in the control reactor; therein, the increased concentration of this component was observed. It should be noticed that under thermophilic conditions in both co-digestion series, there was no release of this compound, which is observed under mesophilic conditions (Ruizet et al., 2016).

Another challenging aspect in AD of OP is high phenols content in co-substrate presence. Their toxic influence concerned mainly functional disturbance of acetate-utilising methanogens. The introduction of OP resulted in significant increase of this component in the feedstock. Additionally, in all co-digestion series within AD, the release of this component was observed. The last fact could be particularly problematic, in the case of application of digestate as fertiliser. The increased content of this component might affect the microbial functions in soil resulting in its poor productivity (Levén et al., 2012). It should be pointed out that thermophilic conditions are more susceptible to various inhibitors, leading to low process performance (Zhang et al, 2022). Moreover, the majority of consortia responsible for phenol degradation are isolated as mesophilic (Levén et al., 2012).

Nitrogen and phosphorus nitrogen are essential nutrients for anaerobic microorganism. These compounds are also crucial in the application of digestate as soil amendment (Han et al. 2019, Zhang et al., 2018; Chen et al., 2020). The supplementation of feedstock in lower dose of OP did not influence the composition of both feedstock and digestate. In terms of these compounds, comparable results to control were achieved (Table 2). Minor changes were observed for R3 fed by higher dose of OP. Therein, the application of OP resulted in a slight increase of the TP content in both feedstock and digestate. In turn, as compared to control, the TN concentration was enhanced by 21 and 19% in feedstock and digestate, respectively. Moreover, it should be noticed that the highest release of $\text{PO}_4^{3-}\text{-P}$ was observed in R3.

Biogas production

In the present study, the biogas/methane productions as well as their rates were analysed (Table 3). As compared to control, decreases of both biogas and methane yields were observed in OP presence. However, lower yields were found in R2 supplied by lower dose of OP. Therein, the biogas and methane yields were reduced by 18.4 and 25.5%, respectively. In turn, minor drops by 9.1 and 11% for biogas and methane yields were observed in R3, respectively. In this experiment, relatively low values of methane yields were achieved (Table 3). The thermophilic mono-digestion of pre-treated OP by steam distillation resulted in production varied between 400–600 $\text{mLCH}_4 \cdot \text{g}^{-1} \text{VS}$ (Martin et al., 2010). In turn, under mesophilic conditions, in co-digestion of OP, SS and biochar the average values were 500–704 $\text{mLCH}_4 \cdot \text{g}^{-1} \text{VS}$ (Martinez et al., 2018).

Additionally, the cumulative biogas and methane production curves (Fig. 3a,b), as well as daily biogas production (Fig. 3b) were also analyzed. In co-substrate presence, the biogas/methane productions were reduced as compared to the SS mono-digestion, also confirming the process inhibition. Moreover, it should be noticed that both biogas and methane profiles differed between reactors. Major changes were observed for R3. In R1 and R2 the curves were similarly, therein two phases of biogas production might be distinguished related with an additional peak in its production appeared between 9 and 13 day (Fig. 3c). An analogous trend was observed in co-digestion of SS and brewery spent grain under thermophilic conditions (Lebiocka et al., 2019). In turn, in R3, after the first 4 days of rapid biogas production a steady phase was noticed.

The declining tendency in OP presence was observed also with regard to the biogas and methane production rates (Table 3). As previously, the less beneficial results were obtained in R2. As compared to control, in this case the GPR and MPR were decreased by 18.4 and 20.2%, respectively. It should be pointed out that such unfavourable results, with

Table 3. The results in terms of biogas/methane productions as well as methane content

Parameter	Unit	R1	R2	R3
GP	$\text{mL} \cdot \text{g}^{-1} \text{VS}$	707.8	577.9	643.3
BP	$\text{mLCH}_4 \cdot \text{g}^{-1} \text{VS}$	518.9	416.8	458.6
GPR	$\text{mL} \cdot \text{g}^{-1} \text{VS} \cdot \text{d}^{-1}$	33.7	27.5	30.6
MPR	$\text{mL CH}_4 \cdot \text{g}^{-1} \text{VS} \cdot \text{d}^{-1}$	24.7	19.8	21.8
Methane content	%	73.31±0.01	72.13±0.03	71.29±0.18

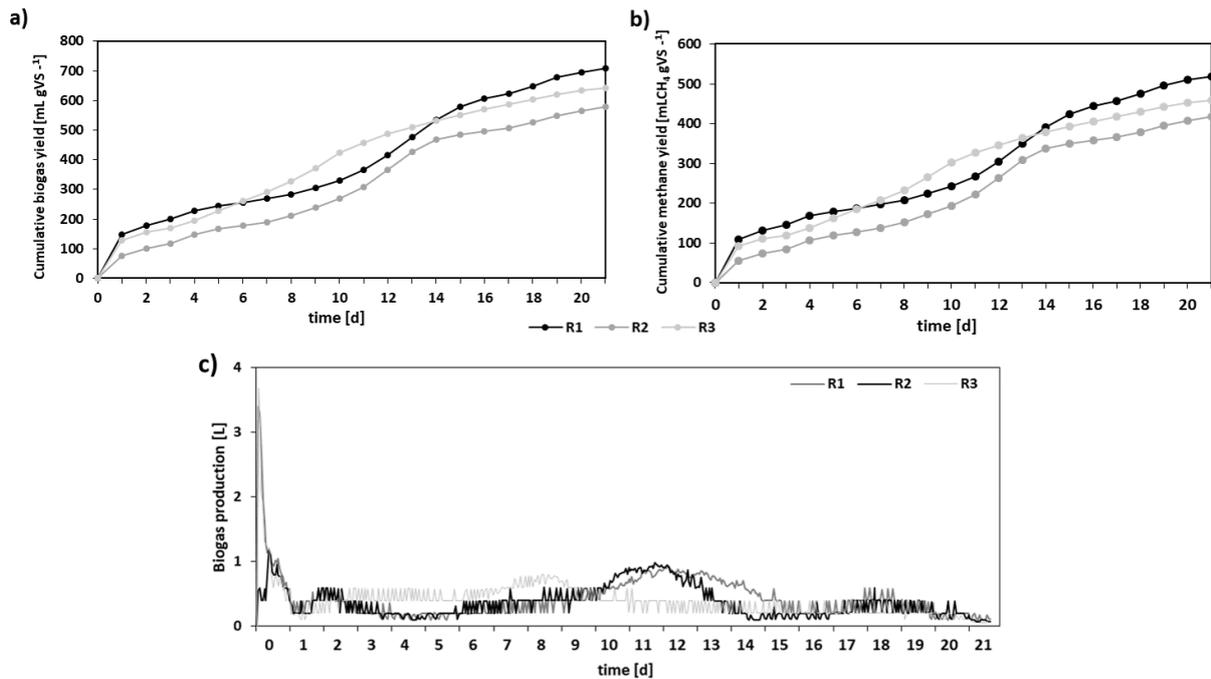


Figure 3. Cumulative biogas (a) and methane (b) yields as well as daily biogas production (c)

regard to the methane parameters, resulted from its diminished content in biogas in OP presence. This effect is related with supplementation the feedstock in OP reached in carbohydrates (Nielfa et al., 2015).

In the present study, several factors contributed to the reduced biogas production in OP presence. It is widely known that the inhibition of both ammonia nitrogen and the accumulation of VFAs they are main two main causes of process instability and low biogas yields (Shi et al., 2020). It should be noticed that in the conducted experiment, relatively high values of these parameters were achieved (Table 2). Moreover, the ammonia inhibition was previously observed in the case of OP mono-digestion (Serrano et al., 2014). Several authors also indicated that AD of various organic wastes with a high concentration of nitrogen and other toxic compounds is more susceptible to inhibition and less stable under thermophilic conditions than under mesophilic ones (Martín et al., 2013). Another aspect is related with limonene presence and its antioxidant properties that might disturb the biological activity. Bouaita et al. (2020) indicated that in the co-digestion of OP and organic fraction of municipal solid waste, an accelerated limonene inhibition was found under thermophilic conditions.

However, the effective co-digestion of selected substrates under thermophilic conditions might be achieved. To eliminate the toxic effect of OP, the appropriately low dose should be provided. Another

aspect is selecting an adequate component to two-component mixture of OP and SS that would provide sufficient buffering capacity and improve the C/N ratio in the feedstock (Calabrò et al., 2020). Therefore, further studies in this field should be conducted.

CONCLUSIONS

In the present study, process performance of thermophilic anaerobic co-digestion of municipal sewage sludge and orange peel was evaluated. The obtained results indicated that the application of OP led to deterioration of process efficiency. At both doses of OP, decreased methane and biogas productions were observed; moreover, the negative effect on GPR and MPR was also visible. Only at minor dose of OP, the VS and TS removals were enhanced as compared to control. In turn, at higher dose of OP, the declining tendency was noticed on those parameters. Additionally, an application of OP adversely affected the stability parameters. The observed changes in co-substrate presence were related with the presence of AD inhibitors such as ammonia nitrogen, VFA, limonene and phenol. The implementation of OP caused the increase of their contents in reactors, leading to poor process efficiency. Moreover, the thermophilic conditions are not recommended for anaerobic co-digestion of those substrates, because they might accelerate the inhibition phenomenon.

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