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Effects of mesophilic and thermophilic anaerobic digestion of sewage sludge on different polymers: Perspectives on the potential of the treatment to degrade microplastics

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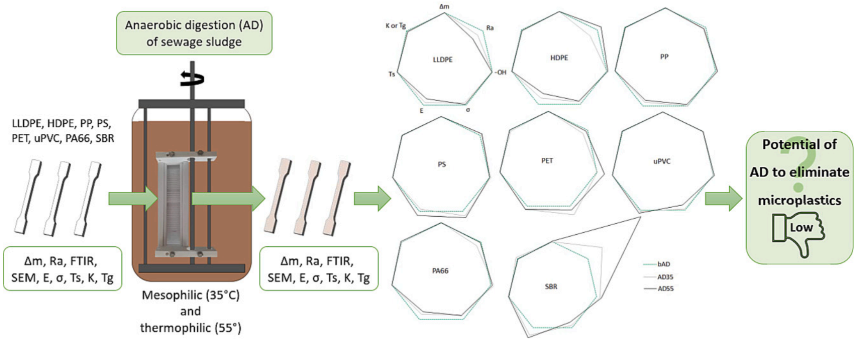
HIGHLIGHTS

- Evaluation of anaerobic digestion (AD) removal of microplastics in sewage sludge.
- Microplastic abundance after anaerobic digestion is pondered.
- Plastic degradation caused by meso- and thermophilic anaerobic digestion is studied.
- No substantial degradation due to slow and limited degradation mechanisms.
- Meso- and thermophilic AD is not a way to eliminate microplastics from sludge.

GRAPHICAL ABSTRACT



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ABSTRACT

Sewage sludge is produced during municipal wastewater treatment and can be further treated to be used for soil applications due to its high nutrient and carbon content. Anaerobic digestion is often used to manage sewage sludge. However, sewage sludge has a high load of microplastics that can be transferred to the soil, causing a burden to the environment. Some researchers suggest that anaerobic digestion could be used as a method to remove microplastics from sewage sludge, while others have shown the opposite. In this study, a variety of commodity polymers (LLDPE, HDPE, PP, PS, PET, uPVC, PA66 and SBR) are tested under mesophilic (35 °C) and thermophilic (55 °C) anaerobic digestion to evaluate their degradation after the process. As 1 mm thick sheets of polymers were used, in terms of diffusion they were considered to correspond to microplastics. Different characterization methods were used to access the visual, chemical, mechanical and thermal changes caused by anaerobic digestion. The results showed evidence of polymer degradation, for example, surface smoothening of LLDPE, HDPE and PP, embrittlement of PS and uPVC, hydrolysis of PET, plasticization of PA66, and surface cracking of SBR. However, although some changes in properties happened, anaerobic digestion could not comprehensively degrade the studied polymers. Therefore, this study suggests that anaerobic digestion of sewage sludge, at the conditions tested, is not able to be used as a method to eliminate microplastics from the sewage sludge before it is added to the soil.

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1. Introduction

Wastewater treatment plants (WWTPs) receive significant load of microplastics (MPs) (Xu et al., 2021). The treatment stages at the WWTPs consist of screening, grit and grease removal, primary settling, biological treatment, secondary settling and, in some cases, tertiary treatment and disinfection (Stuetz, 2009). It has been reported that most of MPs are retained in the sewage sludge, generated from the primary and secondary settling (Talvitie et al., 2017). The sewage sludge can be treated, e.g., via anaerobic digestion (AD) or composting, and further used, for example as a fertilizer (Piehl et al., 2018). Hence, utilisation of treated sludge is considered an important mean of transport for MPs to the environment. MPs in the sewage sludge is a class of pollutants that is not yet regulated (Hudcová et al., 2019). However, their deposition on the soil raises concerns because it has been demonstrated that MPs can compromise the soil safety and their presence is a serious risk for the biota and nutrient cycle (Koyuncuoğlu and Erden, 2021).

To date, the complete separation of MPs from the sewage sludge is not a feasible alternative (Tang, 2023). Conversely, it has been suggested that sewage sludge treatments could be used to destroy the MPs within the sewage sludge and thus, avoid their release into terrestrial systems (Vuori and Ollikainen, 2022). AD is widely used for sewage sludge treatment. It is a biological process in which a microbial community decomposes organic matter of the sewage sludge in the absence of oxygen. The decomposition process happens through hydrolysis, acidogenesis, acetogenesis and methanogenesis reactions and produces biogas (Botheju and Bakke, 2011). It is commonly operated under temperature ranges of 30 to 40 °C (mesophilic process) or 50 to 60 °C (thermophilic process) (Xu et al., 2020). Generally, AD is considered an economical and environment friendly method (Monfet et al., 2018). Moreover, it has been suggested it could degrade and, ultimately, eliminate microplastics from the sewage sludge (Tang, 2023; Mahon et al., 2017; Nielsen et al., 2019; Malakhova et al., 2023). Mahon et al. (2017) observed a reduction in the abundance of MPs in the sewage sludge after AD, Nielsen et al. (2019) noticed biodegradation of polypropylene (PP) under thermophilic conditions and Malakhova et al. (2023) demonstrated the weight loss and even destruction of PP and polyvinyl chloride (PVC). However, it has been also proposed that AD has no potential to remove MPs content from the sewage sludge by Vuori and Ollikainen (2022). Nonetheless, they have not provided consistent discussion regarding this assumption since their study had another focus than MPs' degradation.

The degradation processes of polymers are dependent on the physical, chemical and biological conditions of the surroundings. For example, dynamical forces, availability of oxygen, temperature, light incidence and types of microorganisms are some factors that should be considered (Liu et al., 2022). Certainly, the degradability of polymers depends also on their intrinsic characteristics, such as, synthesis process, chemical composition, molecular weight, hydrophobic character and additives (Singh and Sharma, 2008).

According to Liu et al. (2022), there is a lack of research done on degradation of polymers under AD, and MPs treatment in aerobic conditions is more often reported. The present study aims to investigate the potential of mesophilic (35 °C) and thermophilic (55 °C) AD to degrade different polymers. For that, a laboratory-scale semi-continuous bioreactors were used that are widely used to simulate a larger scale AD process (Cazaudehore et al., 2022). Temperature was the parameter chosen to be studied because it has a major effect on the degradation of polymers as it controls the diffusion rate of water and other substances into the polymer structures and affects the reaction rate of polymer degradation (Ehrenstein and Pongratz, 2013). Furthermore, higher temperature increases the activity of microorganisms and also the rate of biochemical reactions (Xu et al., 2020; Cazaudehore et al., 2022). According to Cazaudehore et al. (2022), studies on polymer degradation under thermophilic conditions is much less studied than under mesophilic conditions. Additionally, to the best of our knowledge, the investigation of how temperature affects anaerobic degradation of MPs has not been done. Therefore, this study intends to provide a better understanding about the matter and also the perspectives of AD as a possible method for elimination of MPs from the sewage sludge to make it safer for further use.

2. Materials and methods

2.1. Feedstock and inoculum for anaerobic digesters

The sewage sludge, after the thickening stage, used as feedstock and the anaerobically digested sewage sludge used as inoculum at 35 °C were collected from Viinikanlahti WWTP (Tampere, Finland). The anaerobically digested sewage sludge used as inoculum at 55 °C was obtained from Topinoja biogas plant (Turku, Finland). The total solid (TS) and volatile solid (VS) contents of the sewage sludge were 4.3 %-TS and 3.0 %-VS, respectively, and for the anaerobically digested sewage sludge of Viinikanlahti and Topinoja samples were 3.0 %-TS and 1.7 %-VS and 8.3 %-TS and 4.6 %-VS, respectively.

2.2. Polymer samples

Polymer samples were cut from 1 mm thick sheets of linear low density polyethylene (LLDPE, FB4230, Borealis, Austria), high density polyethylene (HDPE, CG9620, Borealis, Austria), PP (HC205TF D, Borealis, Austria), polystyrene (PS, 143E, Basf, Germany), polyethylene terephthalate (PET, RAMAPET R182, Indorama, Indonesia), unplasticized PVC (uPVC, ETRADUR, Etra, Finland), polyamide 66 (PA66, Zytel 101F NC010, DuPont, United States) and styrene butadiene rubber (SBR, Etra standard SBR, Etra, Finland). The selection of the polymer types was done based on their occurrence on the sewage sludge (Casella et al., 2023; Hassan et al., 2023).

The sample form, namely the 1 mm thick sheet, can be considered to be comparable with MPs' dimensions and thus diffusion times. Therefore, the results are judged relevant for a microplastic study.

LLDPE, HDPE, PP and SBR sheets were cut with a pneumatic press (Elastocon, Sweden) into dogbone shape samples with 35 mm overall length, 12 mm gauge length and 2 mm gauge width. PS, PET, PVC and PA66 sheets were fracturing when the pressure of the pneumatic press was applied, so they were cut into rectangular samples with 35 mm length and 2 mm width using a lever cutter. For the AD, 7 (LLDPE, HDPE, PP and SBR) and 13 (PS, PET, PVC and PA66) samples were used.

2.3. Anaerobic digestion reactor set-up and operation

AD was performed in four completely stirred tank reactors (CSTRs) operated in duplicate under mesophilic (35 °C; reactors R1 and R2) and thermophilic (55 °C; reactors R3 and R4) conditions. Each condition is referred in this study as AD35 or AD55. The reactors were inoculated by filling them (4 L) with anaerobically digested sewage sludge. The reactors were fed with sewage sludge in a semi-continuous mode five days a week to obtain a hydraulic retention time (HRT) of 20 days and organic loading rate (OLR) of 2.1 ± 0.2 g-VS/L·d. Upon each feeding cycle 280 mL of digestate was removed from the reactor and used for analyses, after which 280 mL of sewage sludge was added to the reactors. In each reactor, the sewage sludge was constantly stirred at 85 rpm. The reactors were run for 40 days (2 HRT), after which the polymers were added to the reactors and kept there for 40 days. LLDPE, PP, uPVC and PA66 were added in the reactors R1 and R3 and HDPE, PS, PET and SBR were added in the reactors R2 and R4.

2.4. Sample holder

The polymer samples were loaded on four sample holders that were attached into the mechanical impellers of the reactors. The holders were 3D printed with polylactic acid and each of them was able to carry 40

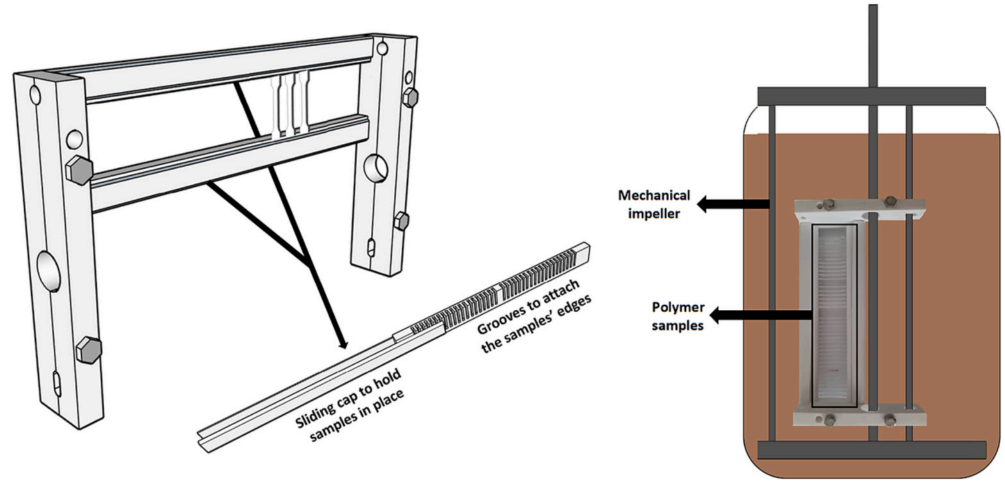

Fig. 1. Sample holder design and set up. The polymeric samples had both of their ends attached to grooves printed on the holder.

polymer samples. The sample holders were used to avoid the polymer samples from being discarded when the sewage sludge on the reactor's content was replaced. Moreover, they were designed in a way the polymer samples would be constantly in contact with the sewage sludge as it would flow between the samples during stirring. The final design of the sample holder and how the polymer samples were inserted in it is shown in Fig. 1.

2.5. *Biofilm removal from polymer's surface*

After removing polymeric samples for the reactors, the biofilm that could potentially form on the polymer's surface was washed off with 2 % sodium dodecyl sulphate (SDS, Acros Organics, United States) for 4 h, followed by washing with distilled water (Orr et al., 2004). Samples were subsequently dried at 50 °C for 24 h. It was performed for 3 samples of LLDPE, HDPE, PP and SBR and for 6 samples of PS, PET, uPVC and PA66. This procedure was indicated in this study as AD35-SDS or AD55-SDS.

2.6. *Analytical methods to assess the anaerobic digestion*

The characteristics of fresh sewage sludge were determined after obtaining fresh batch of sewage sludge (three times during the reactor runs). Furthermore, the digested sludge from the reactors as well as the biogas volume and methane content were characterized 3–5 times a week. TS and VS were gravimetrically determined according to standard methods (APHA 2540). The pH was measured with a WTW pH -3110. The methane content was analyzed with gas chromatograph equipped with thermal conductivity detector as described by Mönkäre et al. (2015). The volume of biogas collected in gas bags was measured with water displacement method. The methane production results were converted to STP conditions. From the digested sewage sludge, soluble chemical oxygen demand (sCOD) was analyzed according to Finnish standard methods (SFS 5504) and volatile fatty acids (VFAs) with gas chromatograph equipped with flame ionisation detector as described by Kokko et al. (2018). Before sCOD and VFA analyses, the samples were filtered (0.45 μm, Chromafil Xtra PET). The results for methane production are given as weekly averages to compensate for the feeding that was done five days a week.

2.7. *Analytical methods to assess polymer degradation*

2.7.1. *Mass change*

The mass change of the polymer samples was monitored by gravimetric method before anaerobic digestion (bAD), i.e. the original samples' weight, and after AD35 and AD55. The samples removed from the reactor at the end of the AD were washed with distilled water to remove the excess of debris on the surface. The mass of the samples was also measured after the SDS washing. Samples were dried to constant weight for 24 h at 50 °C. The mass change percentage was calculated using Eq. (1):

$$\Delta m = \frac{m_{final} - m_{original}}{m_{original}} \times 100\% \qquad (1)$$

Where $m_{original}$ is the mass of the polymer sample before AD, m_{final} is the mass of the polymer sample after AD and SDS washing at the indicated temperature.

2.7.2. *Visual analysis*

Polymer degradation as well as the presence of biofilm on samples after AD and after SDS washing were assessed by using scanning electron microscopy (SEM) JSM-IT500 (Jeol, Japan) and compared with the original films. All samples were coated with a 4 nm platinum-palladium layer to improve their conductivity.

2.7.3. *Roughness*

Measurements of the average roughness (Ra) of bAD, AD35 and AD55 were performed using the profilometers Alicona InfiniteFocus G5 (Alicona Imaging GmbH, Austria) for LLDPE, HDPE, PP, PET, PA66, SBR and Wyko NT1100 (Veeco, USA) for PS and PVC, as they were transparent materials. Ra is a parameter commonly used for surface texture analysis and it measures the deviation of a surface from a mean height (Whitehouse, 1996).

Measurements were done using 20× magnification. Moreover, for each condition (bAD, AD35-SDS or AD55-SDS), two samples were used and, in each, three regions were selected to obtain a standard Ra value: the middle of the sample's surface (0,0), (−3,0) and (3,0). In total, an area of 1.96 mm^2 (Alicona) and 0.21 mm^2 (Wiko) was studied for each sample type. The standard Ra value for each region was calculated using the device's software.

2.7.4. *Functional groups*

The functional groups on the surface of bAD, AD35 and AD55 were analyzed by Fourier transform infrared spectroscopy (FTIR) using the Spectrum One spectrometer (PerkinElmer, United States) equipped with an attenuated total reflectance sample holder. The scanning range was 600 to 4000 cm^{-1} and the resolution was 4 cm^{-1}. Measurements were done in triplicate.

2.7.5. *Mechanical properties*

Tensile test was performed on bAD, AD35 and AD55 samples to obtain tensile strength (σ) and elastic modulus (E), which represents, respectively, the maximum stress the material can bear before breaking and the resistance of the material to elastic deformation. Tests were done using the Instron 5967 (Instron, United States). For LLDPE, HDPE, PP, PS, PET, PVC and PA66 the test speed was 2 mm/min and for SBR, 10 min/min. The rectangular samples were first manually cut into dogbone shape using a rotary power tool and the final dimensions were 35 mm overall length, 17 mm gauge length and approximately 1 mm gauge width. The tests were performed at least in triplicate. Before testing, samples were conditioned at 23 °C and 60 % relative humidity for at least 24 h.

2.7.6. *Thermal properties*

2.7.6.1. *Thermal stability.* The thermal stability (T_s) of bAD, AD35 and AD55 samples were measured by thermogravimetric analysis using the TG 209 F3 Tarsus (Netzsch, Germany). The T_s indicates the maximum temperature the polymer withstands without thermally decomposing. A decline in T_s is considered as a sign of polymer degradation (Groenewoud, 2001). The temperature range was 30 to 600 °C, the heating rate was 20 °C/min and the test atmosphere was nitrogen. Measurements were done in duplicate. The data was analyzed with Netzsch Proteus 70 software.

2.7.6.2. *Degree of crystallinity and glass transition temperature.* The degree of crystallinity (K) of the semi-crystalline polymers and glass transition temperature (T_g) of the amorphous polymers were determined by differential scanning calorimetry using the DSC 214 Polyma thermal analyzer (Netzsch, Germany). Semi-crystalline polymers have a combination of highly ordered (crystalline) and random (amorphous) regions (Ehrenstein and Pongratz, 2013). Amorphous polymers lack long-range order of molecules and T_g represents a measurement of their

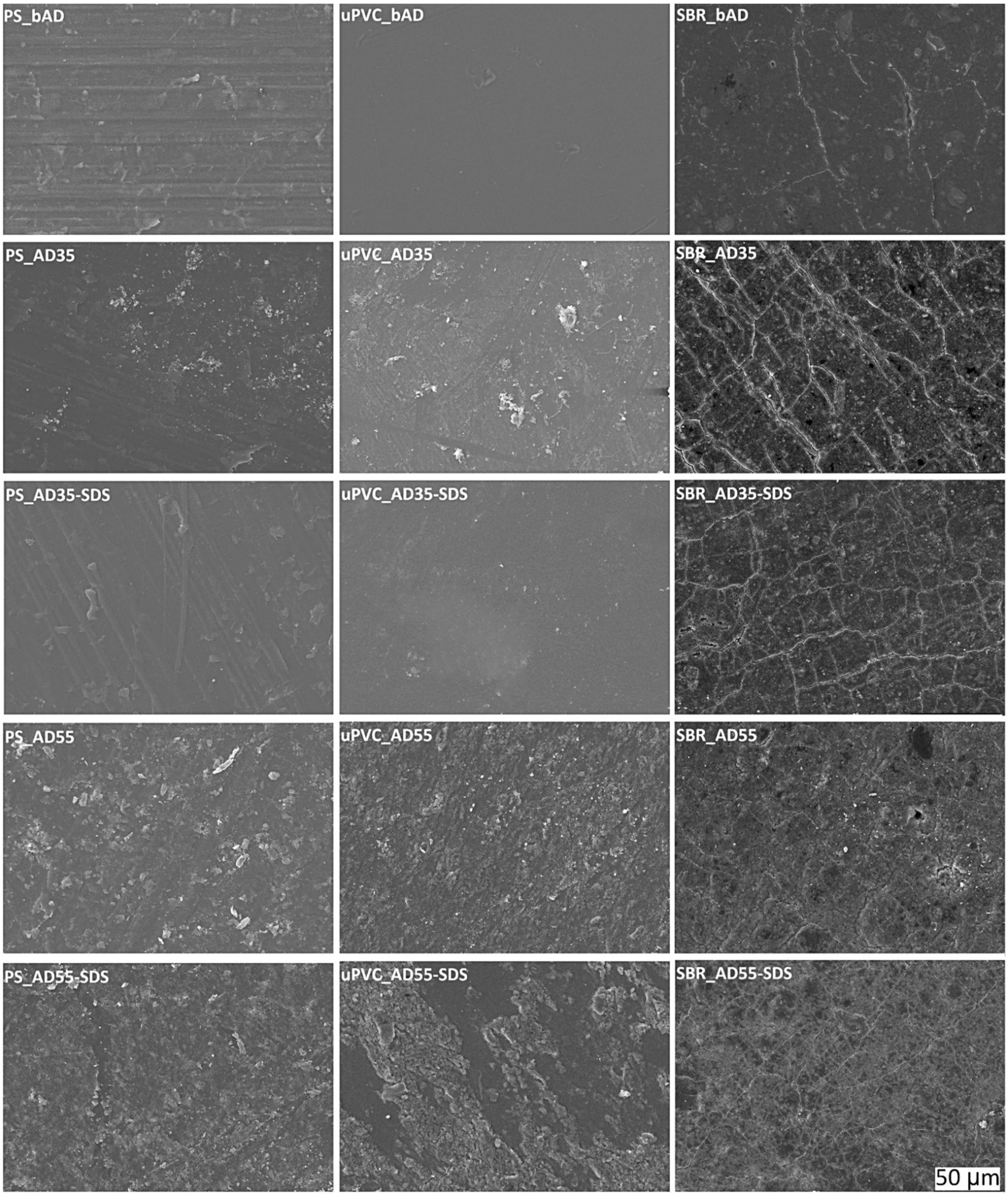

Fig. 2. SEM images of PS, uPVC and SBR at different conditions (pristine, after anaerobic digestion at 35 or 55 °C and SDS washing).

molecular mobility (Ehrenstein and Pongratz, 2013). Measurements were done in duplicate, at a heating rate of 10 °C/min, in nitrogen atmosphere and with two heating cycles. The temperature range depended on the polymer tested and it was set above the melting temperature for the semicrystalline polymers, below the T_g for amorphous polymers as well as below the degradation temperature of the polymers (Table S1). The data was analyzed with the software Netzsch Proteus 70. The determination of K was done using Eq. (2):

$$K = \Delta H_{meas}/\Delta H_{lit} \tag{2}$$

Where ΔH_{meas} is the measured melting enthalpy and ΔH_{lit} is the literature value for completely crystalline material. ΔH_{lit} were obtained from the device's manufacturer (Table S2).

3. Results

3.1. Results on anaerobic digestion

After the first HRT, the weekly average methane content in the biogas was between 49 % and 56 % in AD55 reactors as well as in one AD35 reactor (R1) (Fig. S1). In the other AD35 reactor (R2), the methane content dropped to ca. 35 % during on day 38 after which it slowly recovered to values between 50 and 56 %. The weekly average methane yield in AD55 was 273 ± 44 L-CH_4/kg-VS (Fig. S1). As with methane content, the methane yield in R1 was higher (303 ± 31 L-CH_4/kg-VS) than in R2 (187 ± 84 L-CH_4/kg-VS) (Fig. S2). The pH in the AD55 reactors was slightly higher (pH 7.2–7.6) than in AD35 reactors (pH 7.0–7.3). The organic removal was good throughout the reactor runs as the VS removal was 48 ± 1 % and 52 ± 2 % in AD55 and AD35 reactors, respectively. The sCOD content in the digestate was rather low, 1.2–2.1 g/L, in AD35 reactors and decreased in AD55 reactors from ca. 5.5 down to 2.0 g/L after the first HRT (Fig. S3). Furthermore, the concentration of VFAs was below detection limit in the digestate after day 24 (results not shown), which implies stable operation. Thus, in addition to the different temperature, AD55 reactors had a bit higher pH and sCOD content than AD35. Otherwise, there were no major changes between the two reactors that perhaps could have affected the polymer degradation.

3.2. Results on polymer degradation

From the SEM images (Fig. 2, S4-S8), after 40 days of mesophilic and thermophilic AD, the formation of a continuous biofilm along the surface of the polymer samples was not observed. Instead, in localized regions of the samples, structures that could resemble an early stage of biofilm formation were noticed (Fig. S9) (Awadh et al., 2021; Ganesh Kumar et al., 2021). Selke et al. (2015) have also reported that negligible biofilm was formed on their PE and PVC sheets exposed to AD at similar temperatures. The absence of biofilm could be due to the very smooth surface of the samples and the constant movement of the sewage sludge inside the reactor, which could hinder the microbial adhesion to the surface. Also, the polymer samples were not the sole carbon and energy source for the microorganisms, on the contrary, sewage sludge is a feedstock very rich in organic matter, which is easier available than the carbon in the polymers (Ganesh Kumar et al., 2021).

The material on the surface of the samples after AD35 and AD55 could also be considered as sewage sludge residues of inorganic and organic origin. Interestingly, comparing to the other polymer types, PS and uPVC contained significant amount of sewage sludge residue on their surfaces, and visually there was more of this residue after AD55 than after AD35 (Fig. 2). It was noticed that, after AD55, significant amount of this residue remained on their surface after SDS washing.

Majority of the polymer types did not show significant mass change in the AD at either of the temperatures (Fig. 3a), only SBR had mass change above 1 % after AD55. As residues of sewage sludge remained on some polymer surfaces after the SDS washing, it contributed to the increase of mass after AD. Some of the polymers experienced changing in colour, for example LLDPE and PA66, as shown in Fig. S10, indicating the absorption of some component from the sewage sludge (Zhang et al.,

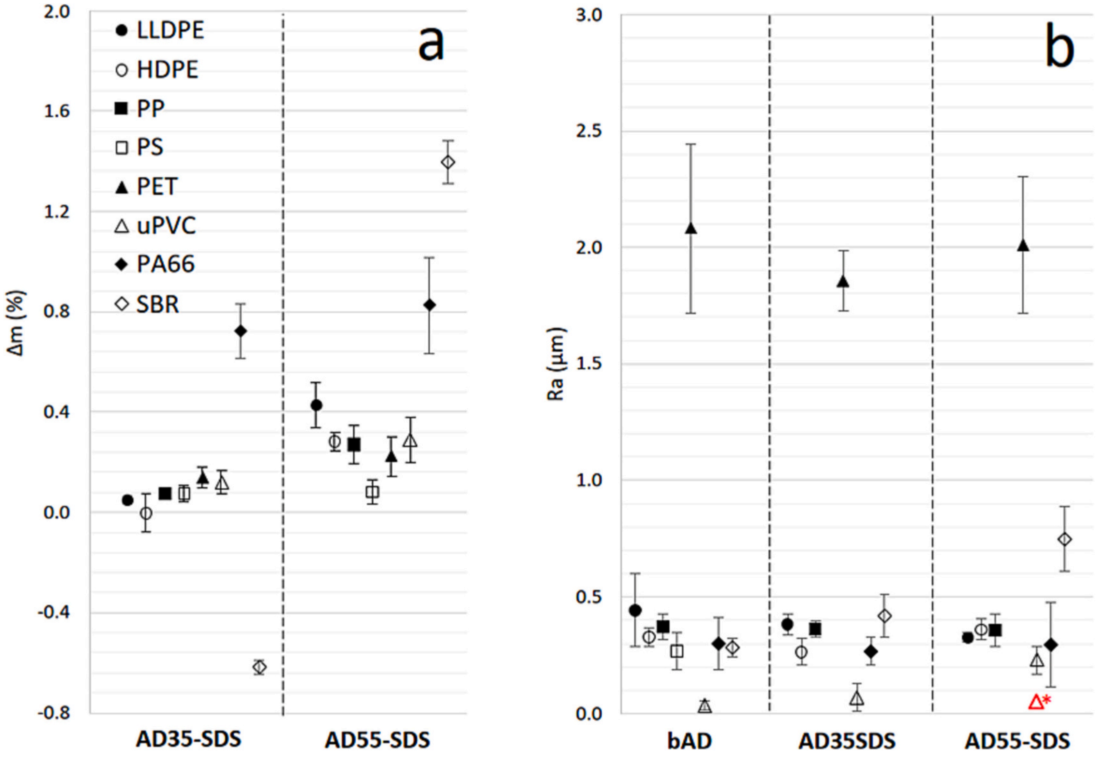

Fig. 3. (a) Mass change and (b) average roughness of polymeric samples after AD35 and AD55.
** Ra value of uPVC AD55-SDS after isopropanol cleaning.*

2018). Mass changes can also be related to the release of soluble additives and impurities on the polymers (Zhang et al., 2022).

The Ra values of AD35-SDS and AD55-SDS compared to bAD are indicated in Fig. 3b. Differently from Alassali et al. (2018), where an increase in roughness on the PE samples under AD was noticed, there was a decrease in the roughness of LLDPE and HDPE in this study. The smoothening can also be noticed in the SEM images. Regarding PP, similarly to Nielsen et al. (2019), it was also noticed a smoothening of the surface. For LLDPE and PP, $Ra_{AD55} < Ra_{AD35}$. Although not clearly indicated by Ra values, SEM images show that the surface's appearance of PS after AD55 seemed rougher than bAD. Also, PA66's surface presented eroded regions. In the case of uPVC, no significant increase in Ra was noticed after AD35 but it increased around 5 times after AD55. After AD, SBR became rougher, with $Ra_{AD55} > Ra_{AD35}$, due to the development of cracks on the surface as seen in the SEM images. From AD35 to AD55, it did not seem that the number of cracks increased on the SBR's surface, instead it got a 'cauliflower' aspect after AD55.

The FTIR spectra represented are related to the samples after SDS washing (Figs. 4 and S11). Although there were not significant changes between the FTIR spectra before and after SDS washing, the selection of the spectra after SDS washing was based on Zhang et al. (2022). According to them, the FTIR results could be possibly interfered by functional groups related to the release of additives or biofilm. Thus, to reduce misinterpretation, they suggested studying the washed polymer surface.

Compared to bAD, for majority of the samples, it was noticed some

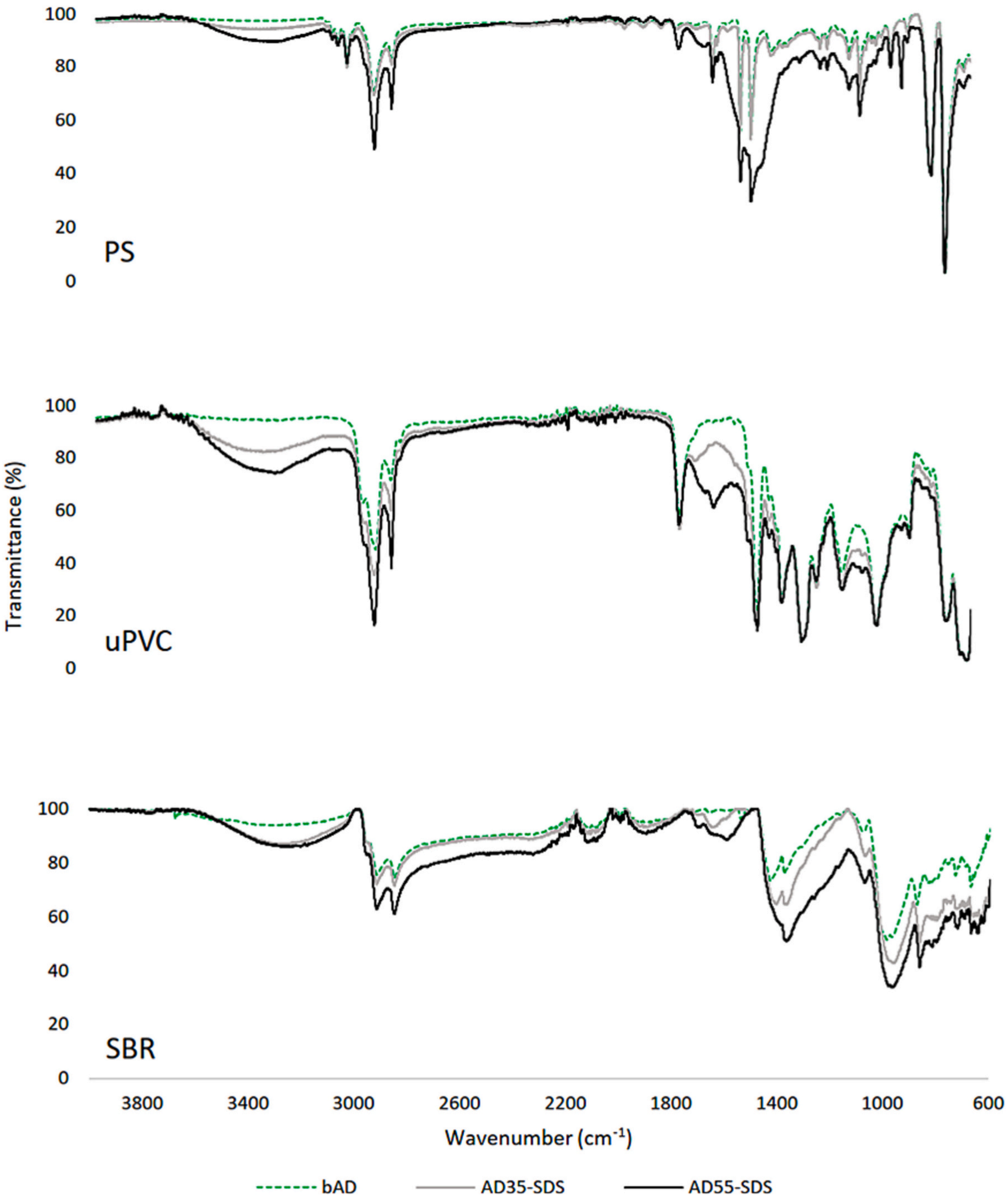

Fig. 4. FTIR spectra of samples that showed noticeable changes on the functional groups on the surface.

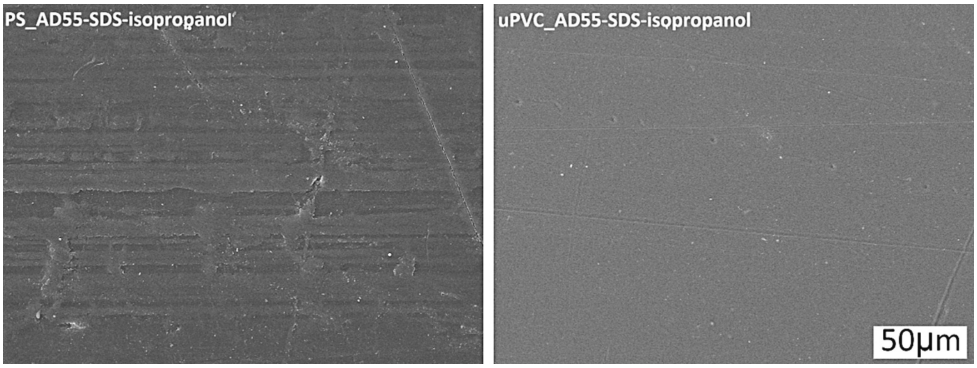

Fig. 5. The surfaces of PS and uPVC AD55 after SDS washing and isopropanol cleaning.

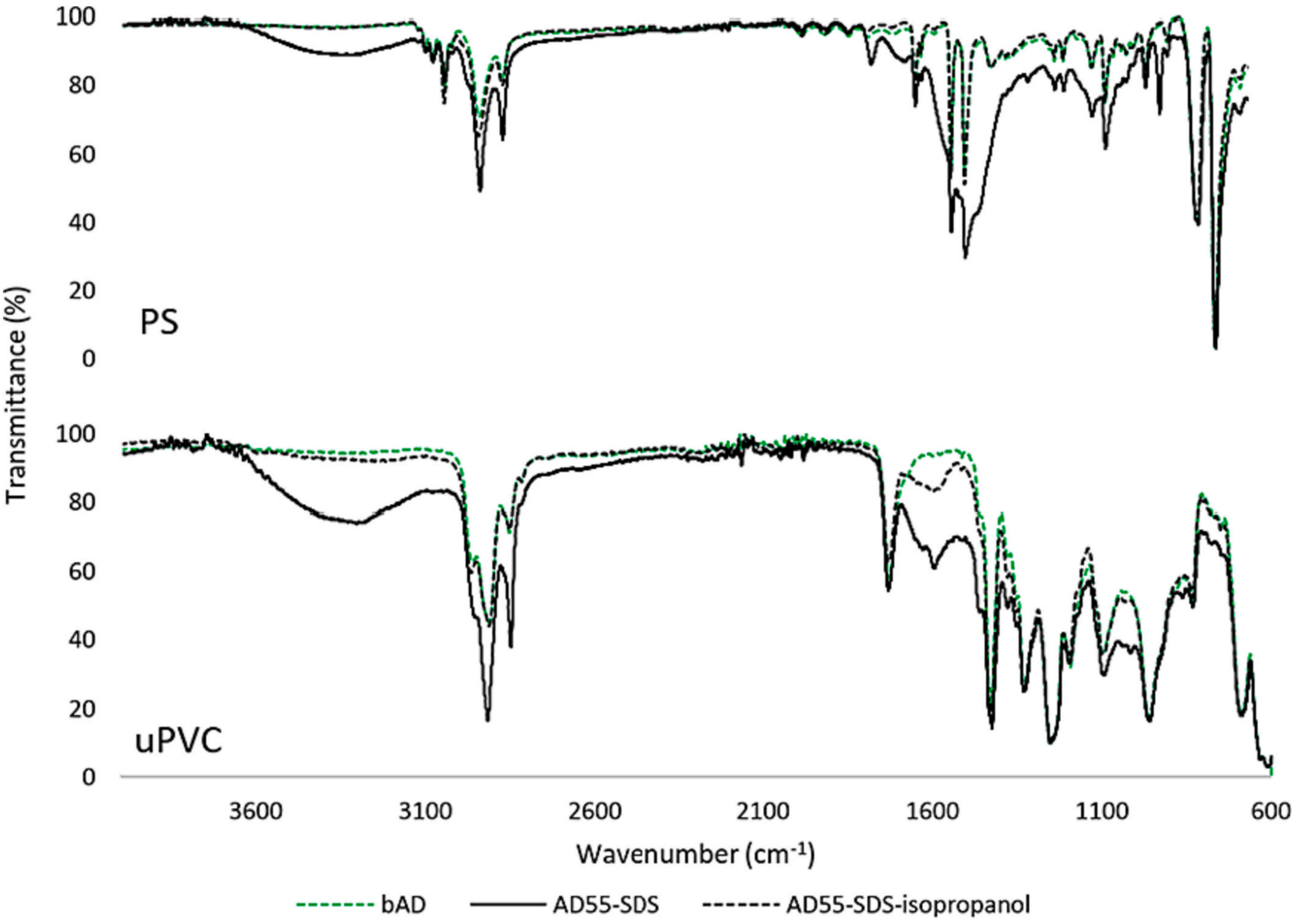

Fig. 6. The FTIR spectra of PS and uPVC after SDS washing and isopropanol cleaning.

increase in the intensity of the peaks related to hydroxyl group (OH) on the 2500–3000 cm^{-1} range. Besides it, HPDE did not exhibit any peaks that could be typical for degradation products. For LLDPE, there was a slight increase on the peak intensities related to unsaturated carbon-carbon bond (C=C) at 800, 860 and 1630 cm^{-1}, and methyl group (CH_3) at 1080 and 1370 cm^{-1} after AD35 and AD55. PP also showed small increase related to CH_3 at 1080 and C=C at 1630 cm^{-1}. PET experienced an increase in the intensity of peaks related to methylene group (CH_2) and CH_3 in the region between 2800 and 2950 cm^{-1}. For PA66 there was a decrease of the peak's intensity at 800 cm^{-1} related to C—H and a gradual decrease of intensity in the region between 1000 and 1200 cm^{-1} related to CH_3 adjacent to the nitrogen and carbonyl group (C=O), both in the amorphous region (Gonçalves et al., 2007), in the bAD > AD35 > AD55 order.

The polymers that had the most significant spectral changes are presented in Fig. 4. For PS, peaks at 1370, 1451 and 1492 cm^{-1}

Table 1

Tensile stress and elastic modulus of bAD, AD35 and AD55 samples.

	bAD	AD35	AD55
	σ (MPa)		
LLDPE	27.6 ± 0.7	26.1 ± 1.5	27.1 ± 0.6
HDPE	29.4 ± 1.7	27.8 ± 0.6	27.4 ± 0.4
PP	36.4 ± 1.4	36.4 ± 0.9	35.4 ± 3.3
PS	26.1 ± 2.5	28.8 ± 2.5	29.9 ± 1.8
PET	45.4 ± 3.4	54.0 ± 7.1	49.2 ± 8.7
uPVC	55.0 ± 4.9	53.6 ± 2.6	48.9 ± 3.0
PA66	58.5 ± 4.2	52.7 ± 4.9	53.3 ± 4.4
SBR	4.0 ± 0.2	3.8 ± 0.2	3.5 ± 0.7
	E (MPa)		
LLDPE	154.7 ± 2.4	144.7 ± 4.2	132.1 ± 2.6
HDPE	774.6 ± 46.5	668.7 ± 35.0	588.5 ± 15.9
PP	961.9 ± 44.6	888.0 ± 29.3	846.2 ± 104.2
PS	1399.6 ± 58.0	1326.3 ± 208.1	1528.9 ± 90.7
PET	989.9 ± 38.7	1061.0 ± 91.7	1061.4 ± 102.8
uPVC	1477.5 ± 195.1	1518.2 ± 110.5	1568.3 ± 208.7
PA66	875.4 ± 108.8	775.4 ± 77.6	728.5 ± 110.9
SBR	5.9 ± 0.2	7.0 ± 0.2	7.4 ± 1.4

corresponding to, respectively, CH_3, CH_2 and aromatic ring vibrations increased and enlarged after AD55 compared to bAD and AD35. Also, considering AD55, the peak's intensity increased at 1080, 2850 and 2920 cm^{-1} related to CH_3 and new peaks appeared at 960 and 1740 cm^{-1} related to C═C and 1650 cm^{-1} related to carbonyl group (C═O). For uPVC after AD55, bands at 1600 and 1630 cm^{-1} related to C═C appeared. This could be reason for the change in colour it experienced at the higher process temperature (Malakhova et al., 2023). Also, after AD35 and AD55, the intensity of peaks related to CH_3 at 2850 and 2920 cm^{-1} increased, being AD55 > AD35. For SBR, peaks related to the butadiene part of the molecule at 860 and 950 cm^{-1} (vinyl compound) increased in the order AD55 > AD35 > bAD. Compared to bAD, AD35 and AD55 had peaks at 1600 and 1700 cm^{-1} which could be related to C═O and C═C. The intensity of peaks related to CH_3 at 2850 and 2920 cm^{-1} were higher after AD55.

As presented previously, it was noticed that PS and uPVC after AD55 had their surface covered with significant amount of sewage sludge residue and the SDS washing was not able to remove this residue entirely. Therefore, it was not clear whether the peak's enlargement, increase or appearance observed on FTIR spectra of both polymers would be due to the formation of degradation products or to the presence of the residue. On the attempt to investigate this matter, samples of PS and uPVC after AD55 washed with SDS were also manually cleaned with isopropanol and Δm, Ra, FTIR and SEM analyses were repeated. The new FTIR spectra showed that the changes observed on PS and uPVC after AD55 were likely originated from the sewage sludge residue. Peaks related to typical digestate components are found in 2925 to 2950 cm^{-1} (fats and lipids), 1500 to 1600 cm^{-1} (proteins and amino acids), 1000 to 1100 cm^{-1} region (polysaccharides or polysaccharide-like substances) (Yeneneh et al., 2016). The cleaning with isopropanol exposed the surface of the polymers (Fig. 5) and it was evident that they did not experience important changes regarding functional groups or deterioration due to degradation when comparing to bAD, as seen in Fig. 6. The Δm values were not significantly affected by this procedure, and values stayed within the deviation. For uPVC, Ra decreased to similar value as bAD (indicated in Fig. 3b) and for PS, Ra change stayed within the deviation.

Results for strength (σ) and deformation modulus (E) are presented in Table 1. Considering the deviation of the results, the changes in strength experienced by the samples when comparing bAD, AD35 and AD55 could be considered insignificant. For polyolefins (LLDPE, HDPE, PP), it was noticed a decrease in modulus (correlated to the stiffness of the material) after AD, being $E_{AD35} > E_{AD55}$. An increase in modulus of PS was noticed after AD55 but not after AD35, indicating embrittlement of PS at the former test condition. Compared to bAD, PET experienced some increase in modulus, but it seemed that the increase in temperature was not influential on this property. On the other hand, PA66 experienced a decrease in modulus becoming more ductile, being $E_{AD35} > E_{AD55}$. Increased modulus of SBR is a clear indicator of its embrittlement after AD, being $E_{AD35} < E_{AD55}$. Also, uPVC experienced similar trend. Some results for modulus also exhibit large deviation between parallel samples.

The thermal properties of the polymers (bAD, AD35 and AD55) are presented in Table S3. Regarding the thermal stability, it was not significantly affected neither by AD35 nor AD55. The decrease in thermal stability was smaller than 1.5 % for all polymers. Similarly to Alassali et al. (2018), it was observed a slight decrease in crystallinity (indicated by K values in Table S3) of LLDPE and HDPE after AD35 and AD55, being $K_{AD35} < K_{AD55}$. In the case of PET, K did not change after AD, but post-crystallization enthalpy on the second heating cycle increased in the bAD < AD35 < AD55 order (Fig. S12). No significant changes on T_g were noticed for PS, uPVC or SBR.

4. Discussion

4.1. Degradation mechanisms of polymers during anaerobic digestion

For the elimination of the MPs from the sewage sludge through AD, the polymers would need to undergo degradation processes that leads to successive breakage of their molecular chains and, ultimately, to depolymerization into monomers or mineralization into carbon dioxide and methane (Mohanan et al., 2020). In AD, it could be considered that water, reaction products of AD, stirring, microorganisms and enzymes would be the possible causes concurrently involved in the polymer's degradation due to hydrolysis, chemical attack, oxidation, and mechanical stresses (Quecholac-Piña et al., 2020). Thermal, photolytic, photo-oxidative, and thermo-oxidative mechanisms could be disregarded since it is an environment without molecular oxygen (O_2) or ozone, without UV exposure, since it is a closed reactor, and the temperature is considered too low to initiate thermal degradation (Quecholac-Piña et al., 2020).

Even though it is an anaerobic environment, degradation by oxidation can occur due to the presence of enzymes and ions (Zhang et al., 2022; Alassali et al., 2018). For example, metallic ions, already in ppm levels, could accelerate the formation of free radicals that initiate oxidation (Ehrenstein and Pongratz, 2013). Iron, cobalt, copper, chromium, and manganese are some of the metal ions with this catalytic effect and they have been reported to be present in sewage sludge (Qasem et al., 2021). Degradation of SBR by free radicals is quick and significant due to the alkene groups on the structure (Singh and Sharma, 2008; Lessa Belone et al., 2021). So, metal ions (mainly copper and manganese) promote what is called ‘rubber poison’ in SBR (Ehrenstein and Pongratz, 2013), which was manifested in this study by the visible cracking, increase in Ra and embrittlement after AD35 and AD55. In addition, according to the literature, the reactions occurring in AD produces VFAs (Botheju and Bakke, 2011) and SBR has limited resistance to, for example, diluted acetic acid at the AD temperatures (Ehrenstein and Pongratz, 2013). This could also reflect on the damages this study observed on it. Except of SBR, at the AD temperatures, the other studied polymers are resistant to degradation by organic acids (Ehrenstein and Pongratz, 2013).

Hydrolysis degradation occurs due to the contact with the hydrogen cations (H^+) and hydroxyl anions (OH^-) of the water that cause bond cleavage in polymers that have water sensitive groups in their structure. The increase in temperature accelerates the process (Singh and Sharma, 2008). PET is relatively stable under poor oxygen conditions and hydrolysis is the decisive degradation mechanism, causing chain cleavage (Ehrenstein and Pongratz, 2013). In this study, the main evidence of PET degradation by hydrolysis was the increase in the post-crystallization enthalpy, which indicates shortening of the chains in the amorphous region increasing their mobility and possibility for organization (Dilara Hatinoglu and Dilek Sanin, 2022). The chain cleavage could also be noticed by the increase in modulus, which can indicate some embrittlement of PET. Although being prone to water absorption, according to Gonçalves et al. (2007), hydrolysis is not a main mechanism for PA66's degradation in water at pH $\approx$ 7 and at 25 to 90 °C. However, water can have a plasticizing effect on it and the reduction in strength and modulus that was observed could be indicators of this phenomena, which causes increase in chain mobility (Lessa Belone et al., 2021). Some sign of degradation could also be noticed by the decrease of FTIR peaks related to the amorphous region of PA66. Nonetheless, the rate of PA66's degradation is considered to be slow in the absence of oxygen (Gonçalves et al., 2007).

The initiation points for degradation of polymers with only carbon backbone in the main chain, such as LLDPE, HDPE and PP, are very scarce in AD conditions, since they lack hydrolysable and reactive groups on their structure (Zhang et al., 2022). However, the formation of OH, C=O, C=C, CH_3 and CH_2 groups shown by the increase on their peak's intensity on FTIR results in this study could be regarded as the early stages of their degradation (Alassali et al., 2018). For example, chain scission results in the three last groups listed. The initiation of degradation for these polymers could originate from thermal and thermo-oxidative degradation reactions during processing, additives, or impurities. In the case of impurities, also for LLDPE, HDPE and PP, the metallic ions effect mentioned previously is known in promoting oxidation (Ehrenstein and Pongratz, 2013).

Mechanical stress and wear are imposed to the polymer samples due their rotation inside the load. Erosion caused by direct and random impact of the feedstock particles at different angles and abrasion caused by sliding and rolling of the feedstock particles can be considered the wear modes that likely happen on the polymers' surface (Xie et al., 2015). With that, molecular chain breakage may occur producing free radicals that participate on the subsequent reactions associated with polymer degradation (Singh and Sharma, 2008). Evidence of the mechanical degradation on the surface could be noticed, for example, by erosion and scratches of, respectively, PA66 and HDPE surfaces. Moreover, according to Xie et al. (2015), brittle fracture on the surface result in much higher wear. Thus, for the polymers that experienced embrittlement during AD in this study (PS, PET, uPVC, SBR), mechanical stresses could accelerate the possibility of fragmentation.

Polymer's biodegradation requires the formation of biofilm on the surface so the microorganisms can effectively use the polymer as a substrate (Orr et al., 2004; Mohanan et al., 2020). Therefore, as biofilm seemed to be limited in this study, it could be considered that abiotic degradation mechanisms mentioned previously were the major source of any property changes on the polymeric samples. However, abiotic degradation favours biotic degradation (Singh and Sharma, 2008). For example, the formation of OH groups on the polymer's surface after AD could favour biofilm attachment and establishment due to increase of hydrophilicity (Orr et al., 2004). Biodegradation could also have the potential to have a bigger a role on the degradation of polymers by changing other AD parameters, such as HRT, besides temperature. For example, Malakhova et al. (2023) have reported the biogenic influence on PVC degradation after 90 days of AD at same thermophilic temperature as this study. However, higher HRTs are not compatible with the conventional process currently used at larger scale AD plants (Cazaudehore et al., 2022).

4.2. *Potential of anaerobic digestion to degrade polymers*

To summarize, as seen in Fig. 7, some changes were observed on the samples' properties after 40 days of mesophilic and thermophilic AD. In most of the cases, it was noticed that the changes were higher for thermophilic than for mesophilic conditions, as also observed by Cazaudehore et al. (2022) and Quecholac-Piña et al. (2020). Nevertheless, neither AD35 nor AD55 were able to comprehensively deteriorate the properties of the studied polymers. This could be related to the fact that, for the AD's parameters and environment, the degradation mechanisms were limited. Considering the polymer's degradation, the AD occurs at very low temperatures and short time. Moreover, UV radiation and O_2 that are the most important initiators for degradation for polymers (Ehrenstein and Pongratz, 2013), are absent. In addition, the degradation effects observed were mainly restricted to the surface of the samples (Lessa Belone et al., 2021).

4.3. *Considerations about anaerobic digestion of sewage sludge containing microplastics*

Considering the scenario of MPs in the sewage sludge being subjected to AD, it could be assumed that their degradation could be severer than the degradation of the samples in this study. Most likely, the MPs would have already been exposed to conditions that could introduce more hydrophilic and/or reactive sites to their surface. For example, the surrounding conditions before they reach the WWTPs (e.g. sunlight, oxygen, chemicals) and the wastewater and sludge unit treatment processes before the AD process (Mahon et al., 2017; Lessa Belone et al., 2022). In addition, they have higher surface area than the 1 mm thick polymer sheets used in this study. These would make them more susceptible to biofilm development and degradation reactions (Zhang et al., 2022). On the other hand, MPs degradation could be hindered by absorbed contaminants, such as antibiotics, which can, for example, be toxic to microorganisms, thus affecting the biotic degradation (Azizi et al., 2021). Also, MPs could be folded, twisted, aggregated to each other, or floating on the sewage sludge in the reactor (Yagi et al., 2012).

Therefore, based on these factors and on this study, it could be considered that tendency is that AD, at the presented conditions, would not be suitable as a method to eliminate MPs from the sewage sludge. Even though it could lead to some morphological and chemical changes on the MPs surface, their degradability is slow (Tang, 2023). Thus, their destruction would likely not be achievable on a conventional, large scale AD process.

5. Conclusions

The effects of anaerobic digestion (AD) of sewage sludge on different polymers (LLDPE, HDPE, PP, PS, PET, uPVC, PA66 and SBR) were assessed in mesophilic (35 °C) and thermophilic (55 °C) conditions. Polymer sheets of 1 mm thickness were used, so in terms of diffusion, the samples were considered corresponding to microplastics. The analysis of the degradation of the studied polymers could provide an understanding on the potential of AD to eliminate microplastics from the sewage sludge. This study suggested that AD at mesophilic or thermophilic temperatures would likely have no capacity of microplastics destruction. Within typical durations of AD treatment of sewage sludge, considering solely temperature as the parameter to be changed, microplastics' degradation could be favoured by performing AD at higher temperatures. However, this possibility is not feasible to be implemented at commercial scale anaerobic digesters, because of higher energy demand for heating, more difficult stabilization, and less microbial cultures suitable for higher temperatures. Therefore, based on this study, it is believed that AD cannot yet be considered as a possible remediation of microplastics before sewage sludge is added to soil.

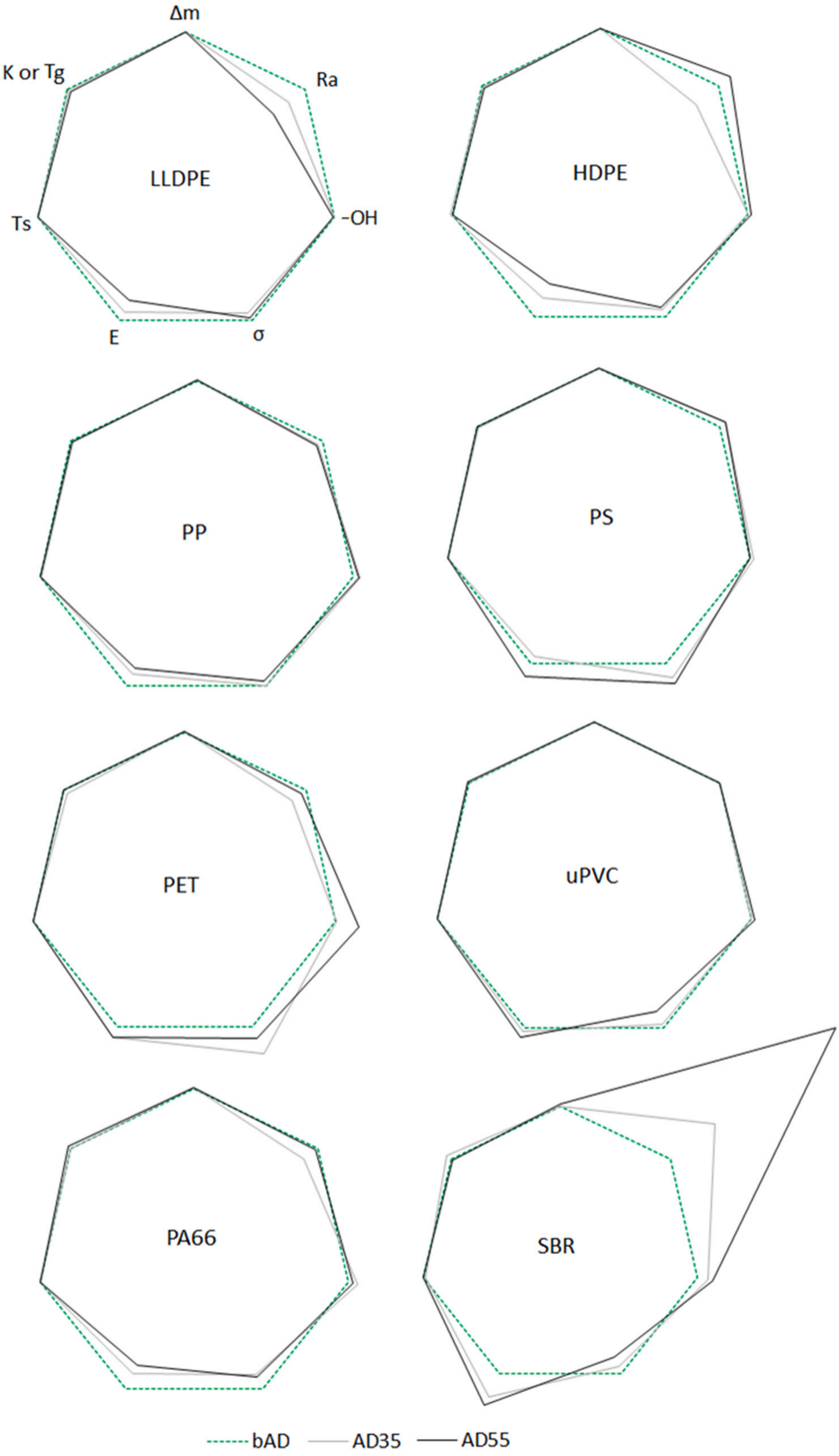

Fig. 7. The property changes the polymeric samples experienced after AD35 and AD55 comparing to bAD. Changes that happened after AD35 and AD55 are expressed in percentages compared to bAD.

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.

Data availability

Data will be made available on request.

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Author statement

Effects of mesophilic and thermophilic anaerobic digestion of sewage sludge on different polymers: perspectives on the potential of the treatment to degrade microplastics.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.scitotenv.2023.168014.

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