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Evaluation of the anaerobic degradation of food waste collection bags made of paper or bioplastic

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Abstract:	The amount of compostable bioplastics collected with the food waste is constantly growing, particularly due to the bags used for collection. According to the Italian legislation, compostable bioplastics must be accepted by all biological treatment plants, including aerobic and anaerobic facilities. Anyway, the compostability standard requires only the assessment of the aerobic degradability, while it is generally not required to test the behaviour under anaerobic conditions. This aspect is evaluated in the paper, where the anaerobic degradability of bioplastic bags used for the food waste collection is assessed. First, Biochemical Methane Potential (BMP) tests were performed on four commercial types of bioplastic bags, including those designed only for the collection of food waste and the shoppers, that can be reused for the same purpose. Subsequently, an innovative approach for this kind of substrate was applied, subjecting two bags to semi-continuous co-digestion tests together with the food waste. Both tests were performed by comparing the behaviour of bioplastic bags with that of an alternative collection paper bag. Finally, tests to evaluate the influence of physical phenomena on the degradation of bioplastics were performed to better understand the results of biological tests. BMP tests indicated a good degradability (>71%) of bioplastic bags, while semi-continuous tests showed a much lower degradability (<27%), confirmed by the observation of the undigested bag pieces. On the contrary, the paper bag presents interesting characteristics, because its degradability in the semi-continuous tests (82%) resulted even higher than that observed in the BMP tests (74%). These results highlight an important difference between the bags mono-digestion by means of BMP tests and the semi-continuous co-digestion tests with food waste, which better simulate the full-scale operational conditions.

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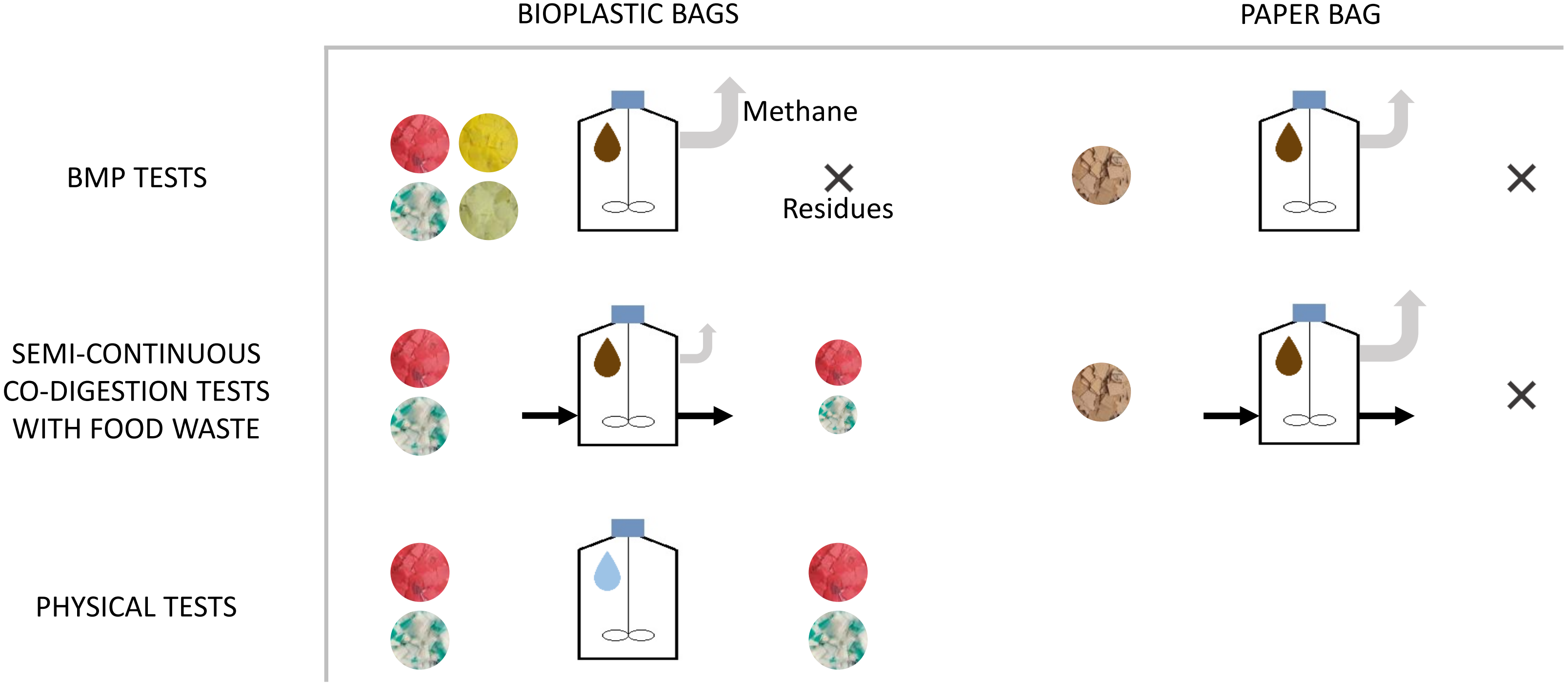
Graphical Abstracts



Highlights

- Bioplastic and paper bags were tested with BMP and semi-continuous digestion tests
- Paper bags show a good degradability in both the tests (>74%)
- The BMP tests on bioplastic bags indicate a good degradability (>71%)
- Semi-continuous digestion of bioplastics with food waste shows a low degradability
- The degradability of bioplastics is influenced by physical factors

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Abstract

The amount of compostable bioplastics collected with the food waste is constantly growing, particularly due to the bags used for collection. According to the Italian legislation, compostable bioplastics must be accepted by all biological treatment plants, including aerobic and anaerobic facilities. Anyway, the compostability standard requires only the assessment of the aerobic degradability, while it is generally not required to test the behaviour under anaerobic conditions. This aspect is evaluated in the paper, where the anaerobic degradability of bioplastic bags used for the food waste collection is assessed. First, Biochemical Methane Potential (BMP) tests were performed on four commercial types of bioplastic bags, including those designed only for the collection of food waste and the shoppers, that can be reused for the same purpose. Subsequently, an innovative approach for this kind of substrate was applied, subjecting two bags to semi-continuous co-digestion tests together with the food waste. Both tests were performed by comparing the behaviour of bioplastic bags with that of an alternative collection paper bag. Finally, tests to evaluate the influence of physical phenomena on the degradation of bioplastics were performed to better understand the results of biological tests. BMP tests indicated a good degradability (>71%) of bioplastic bags, while semi-continuous tests showed a much lower degradability (<27%), confirmed by the observation of the undigested bag pieces. On the contrary, the paper bag presents interesting characteristics, because its degradability in the semi-continuous tests (82%) resulted even higher than that observed in the BMP tests (74%). These results highlight an important difference between the bags mono-digestion by means of BMP tests and the semi-continuous co-digestion tests with food waste, which better

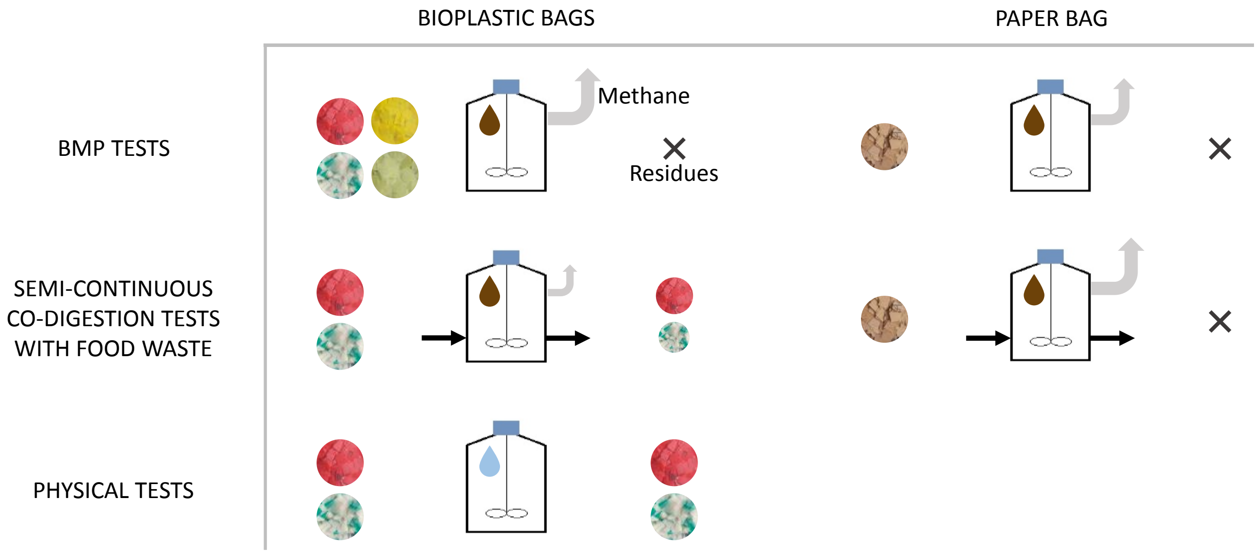
Abbreviations: BDB1, bioplastic dedicated bag 1; BDB2, bioplastic dedicated bag 2; BSB1, bioplastic shopper bag 1; BSB2, bioplastic shopper bag 2; PB, paper bag.

simulate the full-scale operational conditions.

Keywords

food waste; collection bag; paper; bioplastic; anaerobic digestion

Graphical abstract



1 Introduction

The global production of bioplastics is rapidly increasing, expected to approach 3 million tonnes in the year 2025. Currently, 58% of the bioplastic production is related to biodegradable polymers of biogenic or fossil origin (European Bioplastics, 2020). The Italian market of biodegradable and compostable bioplastics was originally driven by bags used for food waste collection and for containing the overall purchase and loose foods like fruit and vegetables at grocery stores. In recent years there was a robust growth of the sector of other packages as well as rigid items made of bioplastics, such as disposable tableware. Accordingly, the amount of compostable bioplastic found in the food waste has increased from 1.5% in the period 2016/2017 to 3.9% in 2019/2020 (CIC-COREPLA, 2020). Such numbers are expected to affect the operation of food waste processing facilities that were not originally designed for the handling of compostable bioplastics (Utilitalia, 2020).

Regarding food waste management in Italy, a significant expansion of anaerobic digestion plants was observed in recent years: about 2.9 million tonnes of food wastes were sent to anaerobic digestion or integrated anaerobic/aerobic plants in the year 2019, compared to only 1.7 million tonnes sent to composting (ISPRA, 2020). In anaerobic digestion facilities, bioplastics are typically removed before the digestion process, because they cannot be separated from conventional plastics (whose amount entwined with food waste is still relevant) and because their management can cause operational problems in the biological reactors (Dolci et al., 2021).

Anyway, according to the Italian legislation, compostable bioplastics must be accepted by biological plants (both aerobic and anaerobic), provided that they fulfil the technical standards UNI EN 13432:2002 for packaging and UNI EN 14995:2007 for other materials (UNI EN, 2002; UNI EN, 2007). In compliance with such standards, only aerobic degradability tests must be performed, while it is generally not necessary to test the biodegradability under anaerobic conditions.

This aspect is examined in the present study, where the anaerobic degradability of bioplastic bags employed for the food waste collection is assessed at the laboratory scale. In Italy, most of the food waste is set out for collection in bioplastic bags. The bags specifically manufactured for separate collection of food wastes (dedicated bags) were tested. Moreover, the bags used for the overall shop at the supermarkets (shopper bags) that are often re-used in the home for food waste collection were examined. Both types are made with the Mater-Bi® polymer, a compostable bioplastic according to the UNI EN 13432:2002 standard, whose composition is 70% polybutylene adipate terephthalate, 20% starch, and 10% additives (Elfehri Borchani et al., 2015).

The behaviour of organic substrates under anaerobic degradation conditions is typically investigated by means of biochemical methane potential (BMP) tests. The literature delivers some studies focused on the evaluation of the anaerobic degradability of Mater-Bi® films (Battista et al., 2021; Dolci et al., 2021; Calabrò et al., 2020; Vasmara and Marchetti, 2016), Mater-Bi® products (Cazaudehore et al., 2019), or films made of similar starch-based blends (Zhang et al., 2018; El-Mashad et al., 2012) by means of BMP tests.

It is common knowledge that the temperature affects the kinetics and degradation rates of the anaerobic digestion processes. On the contrary, the ultimate methane yield generally appears not to be significantly influenced by the temperature (Angelidaki and Sanders, 2004). This is confirmed by the literature findings, showing similar ultimate methane yields for BMP tests performed under mesophilic and thermophilic conditions on various substrates. Golkowska and Greger (2013) found similar total methane yields for maize (-3% for thermophilic conditions) and cellulose (+3%). Moreover, BMP tests performed on food waste and sewage sludge (Gu et al., 2020) did not show statistically significant differences on methane yields between mesophilic and thermophilic conditions. Examining sewage sludge, Mirmasoumi et al. (2018) observed differences lower than 16%, in favour of thermophilic tests.

Conversely, the degradability of the Mater-Bi® polymer is likely to be significantly influenced by the test temperature. Thermophilic BMP tests on Mater-Bi® bags (Dolci et al., 2021) showed relevant differences compared to mesophilic conditions (+48% and +520% for the two examined bags). These different yields are confirmed by Cazaudehore et al. (2019) and Vasmara and Marchetti (2016), with increases of 260% and 240%, respectively. Accordingly, while mesophilic conditions are less effective on the degradation of this polymer, thermophilic conditions seem to enhance degradation. Therefore, this study is focused on a deeper investigation of Mater-Bi® bags anaerobic degradability under thermophilic conditions.

In the first part of the activity, BMP tests were performed on four commercial types of Mater-Bi® bags from different producers. As previously stated, several studies assessed the anaerobic degradability of Mater-Bi® films, and bioplastics in general, through this kind of test. Anyway, BMP tests allow for the evaluation of the maximum amount of methane achievable from a substrate in an optimised lab-scale batch system, under conditions different from those found in full-scale digesters. Moreover, the anaerobic degradability of bioplastics is typically evaluated with mono-digestion tests, while in the waste management systems bioplastics are co-digested with food waste.

To overcome these limitations, in addition to BMP tests, the novelty of this study is the evaluation of the anaerobic degradability of the bags with semi-continuous co-digestion tests together with food waste, to better simulate actual operating conditions in full-scale digesters. This was carried out on two out the four examined bioplastic bags. To the authors' knowledge, similar tests were performed only by Zhang et al. (2018), where the anaerobic degradability of different compostable bioplastics was assessed by performing semi-continuous co-digestion tests with food waste under mesophilic conditions.

Both BMP and semi-continuous tests were performed comparing the behaviour of bioplastic bags with that of a recycled paper bag specifically designed for the food waste collection.

Regarding the behaviour of paper food waste collection bags under anaerobic conditions, previous BMP tests showed a good anaerobic degradability both under mesophilic and thermophilic conditions (Dolci et al., 2021). Moreover, unlike bioplastic bags, the paper bags do not need to be removed at the pre-treatment stage in full-scale anaerobic digestion plants, which means that they actually enter the biological process.

2 Material and methods

Four bioplastic bags were selected for the tests, including two bags specifically designed for the food waste collection (bioplastic dedicated bag 1 - BDB1 and bioplastic dedicated bag 2 - BDB2), and two shopper bags, reusable for collecting the food waste (bioplastic shopper bag 1 - BSB1 and bioplastic shopper bag 2 - BSB2). Moreover, the paper bag specifically designed for the food waste collection (PB) was tested.

The experimental plan first included BMP tests, performed on all five bags, and on a synthetic food waste prepared for the subsequent semi-continuous tests. Then, semi-continuous co-digestion tests on bags and food waste were performed on two out the four bioplastic bags and on the paper bag (co-digestions of food waste - BSB1, food waste - BDB1, and food waste - PB). Finally, in addition to the previous tests combining biological and physical effects, batch tests with only water (so-called physical tests) were performed on BSB1 and BDB1 to evaluate the physical effects on bioplastic degradation.

2.1 Substrates characterisation and preparation

Before the tests, substrates (bags and food waste) were characterised and prepared.

2.1.1 Food waste collection bags

Mater-Bi® bags are manufactured by extruding Mater-Bi® granules; in this process, different dyes and inks (not further specified) are typically added. Moreover, shopper bags are thicker than dedicated bags, for the purpose of accomplishing their primary function. Such differences can affect their bioaccessibility and anaerobic degradability. Accordingly, bags manufactured by different companies with different thickness (two shoppers and two dedicated bags) and colours (red, yellow, not coloured with a green lace for its closing, and green) were selected.

The tested paper bag, made of recycled fibres, is provided with a separate cartonboard bottom to be inserted inside the main bag before the use (22% of the total weight of the bag - wet basis).

For all the tests, bags were manually cut in square pieces with 1 cm side (Figures S1 and S2 of the Supplementary Material - SM).

Wet weight, total solids (TS), volatile solids (VS), chemical oxygen demand (COD), and total Kjeldahl nitrogen (TKN) were measured for all the bags. Moreover, their thermal properties were analysed by means of the differential scanning calorimetry (DSC). Finally, since BMP and semi-continuous tests were performed in wet conditions (and physical tests in water), the behaviour of bioplastic pieces in terms of water sorption was investigated with water swelling tests.

This section describes the characterisation and preparation of substrates and subsequently the setting of BMP, semi-continuous, and physical tests.

2.1.2 Synthetic food waste

The composition of the synthetic food waste (Table 1) was defined based on 90 composition analysis of the organic fraction delivered to three composting plants and to one waste-to-energy facility. During its preparation, water was added to allow for the components shredding. Inert materials (e.g. bones and shells), typically removed in the pre-treatment section of full-scale facilities, were not added to avoid their accumulation in the reactors of the semi-continuous tests. Once prepared, the food waste needed for all the trials was mixed to ensure constant properties and then stored at -18°C. The synthetic food waste (Figure S3 of SM) was characterised in terms of TS, VS, COD, TKN, ammonium (NH_4^+), pH, total alkalinity (TA), and volatile fatty acids (VFA).

Table 1 Synthetic food waste composition without the water added to allow for the components shredding.

Component		Amount (weight %, wet basis)
Fruit and vegetables	edible portion	42
	apples, potatoes, and carrots skin	14
	citrus peel	8
	other non-edible parts	16
Bread and cereals		8
Meat and fish		6
Pasta		3
Wet paper		2
Dairy products and eggs		1

2.2 Inoculum characterisation

A digestate from a wet thermophilic plant processing food waste and sludge from a wastewater treatment plant was used as inoculum in the BMP and semi-continuous tests.

Before the tests, the inoculum was preincubated for 5 days at tests temperature (50°C) and then characterised for TS, VS, COD, and TKN. NH_4^+, pH, TA, and VFA were also verified as suggested by Holliger et al. (2016) for BMP tests.

2.3 BMP tests

2.3.1 Tests settings

The four bioplastic bags, the paper bag, and the food waste were tested in triplicate in 600 mL stirred bottles (80% working volume). Tests were carried out at thermophilic conditions (50 ± 0.5°C). A 2 inoculum to substrate ratio (VS based) was considered. In addition to inoculum and substrate, a mineral medium containing macro and micro-nutrients (OECD, 2006) was dosed in each bottle together with tap water, added to reach the working volume. Three bottles serving as blanks were set up without substrate. The pH was measured in all the bottles before the experiments, resulting in the range 8.0 - 8.5. Before the tests, N_2 was flushed in the headspace to obtain anaerobic conditions (Holliger et al., 2016; VDI, 2016). A volumetric device (Automatic Methane Potential Test System II - AMPTS, Bioprocess Control®) was employed to measure the methane produced during tests.

For each substrate, the final BMP was defined as the mean of triplicates when the daily net production of each of the last 3 days resulted lower than 1% of the corresponding cumulative net production (Koch et al., 2019), and, in any case, with a minimum test duration of 21 days (UNI/TS, 2018).

Despite this, tests on bioplastic bags were continued for at least 40 days because BMP tests curves with stepped trends were observed in previous studies on starch-based biopolymers (Dolci et al., 2021; Cho et al., 2011; Mohee et al., 2008; Russo et al., 2009). At the end of each test, the pH was verified resulting in the range 7.5 - 8.0 for all the bottles. Finally, the digestate of each bottle was sieved at 2, 0.50, and 0.25 mm to retain possible residues.

2.3.2 Kinetic analysis

A kinetic analysis of the BMP tests results of each substrate was then performed to evaluate, at first, the time to achieve 50% and 90% of the final BMP. Subsequently, for each substrate, different models were fitted to the data and then compared by means of efficiency criteria (Section 2.1 of SM). Finally, the selected model for each substrate allowed to compute its anaerobic degradability in an ideal digester, modelled as a continuous stirred-tank reactor (CSTR), as a function of its hydraulic retention time (HRT). These estimates were compared with the results of co-digestion semi-continuous tests (Section 4), to point out possible effects of biomass acclimation or synergistic effects given by the co-digestion of bags and food waste.

2.4 Semi-continuous tests

2.4.1 Tests settings

The tests of food waste and bags co-digestion were performed in 2.4 L stirred reactors, with a 2 L working volume, under thermophilic conditions (50 ± 0.5°C). 1.6 L of digestate were dosed as inoculum in each bottle at the beginning of the tests. Subsequently, semi-continuous conditions were obtained by removing part of digestate and adding the new substrates and water twice a week, to maintain a HRT of 21 days and an organic loading rate (OLR) of 2.20 kgCOD/(m^3×d).

The HRT, comparable to that of the plant providing the inoculum (20 days), was selected according to literature findings on lab-scale tests performed under thermophilic conditions for the wet anaerobic digestion of food waste: 14.9 - 22.2 days (Hartmann and Ahring, 2005), 10 - 20 days (Micolucci et al., 2018), 30 days (Xiao et al., 2018). Moreover, guidelines for the design and the management of wet thermophilic anaerobic digestion of food waste indicate typical HRTs in the range 15 - 18 days for full-scale reactors (CITEC, 2007).

Regarding the OLR, for the wet thermophilic anaerobic digestion of food waste, typical values at the full-scale are in the range 2 - 4 kgVS/(m^3×d) (CITEC, 2007). However, literature suggests for lab-scale tests lower OLRs to guarantee the stability of the process. Ghanimeh et al. (2018) found stable conditions between 0.5 and 2 kgVS/(m^3×d), while Shi et al. (2018) observed an unstable process at 3 kgVS/(m^3×d). Accordingly, the selected OLR is cautiously low, in the range 1.66 - 1.72 kgVS/(m^3×d), corresponding to 2.20 kgCOD/(m^3×d), for the tested substrates (food waste and food waste co-digested with bags).

Tests were performed in four reactors; in the first period (phase 1, 20 feed cycles corresponding to more than three HRTs) all the bottles were fed with only food waste to reach inoculum acclimation and stationary conditions. In the second period (phase 2, 19 feed cycles), three reactors were fed also with bioplastics (11.5% of the OLR on COD basis), the fourth serving as blank (Table 2). The selected proportion corresponds to about 1 kg of food waste inserted into a collection bag.

The selection of the bioplastic bags to be tested (BSB1 and BDB1) was based on BMP tests results (Section 3.3), in which all the bags were characterised by a good anaerobic degradability (higher than 70%). Conservatively, to assess the effectiveness of semi-continuous co-digestion tests on bioplastics degradation in the worst conditions, only the shopper and the dedicated bag with the lowest anaerobic degradability achievable in an ideal CSTR digester with a HRT of 21 days were tested.

Table 2 Fed substrates and corresponding OLR in the two phases of the semi-continuous tests.

Reactor	**Fed substrates**	
	Phase 1 (days 1 - 73)	**Phase 2 (days 74 - 147)**
1	Food waste 2.20 kgCOD/(m^3×d)	Food waste 1.95 kgCOD/(m^3×d) BSB1 0.25 kgCOD/(m^3×d)
2	Food waste 2.20 kgCOD/(m^3×d)	Food waste 1.95 kgCOD/(m^3×d) BDB1 0.25 kgCOD/(m^3×d)
3	Food waste 2.20 kgCOD/(m^3×d)	Food waste 1.95 kgCOD/(m^3×d) PB 0.25 kgCOD/(m^3×d)
4	Food waste 2.20 kgCOD/(m^3×d)	Food waste 2.20 kgCOD/(m^3×d)

2.4.2 Tests operation

Reactors were opened twice a week to enable a representative sampling of the digestate (the removal of undigested bioplastic pieces was not achievable through common discharge operation of semi-continuous reactors). After the completion of discharge, substrates and water were fed to the reactors, and N_2 was flushed in the headspace to restore anaerobic conditions.

During the tests, the produced methane was measured with the AMPTS device. The methane contained in the headspace of reactors lost during each system discharge was accounted for, according to the headspace volume and the biogas composition, regularly measured.

Analyses of pH, TA, TS, VS, VFA, NH_4^+ and periodically COD and TKN were performed on the extracted digestates. In phase 2, extracted digestates were sieved (2 mm) to recover undigested pieces of bags. All the residual bioplastic pieces were washed with tap water, dried at 35°C, and weighed (after the weight stabilisation at room temperature) to evaluate their mass losses during the digestion. Moreover, undigested bioplastic pieces with a surface equal to at least ¾ of that of the input were recovered by hand and counted. These operations were also performed for all the content of reactors when dismissed at the end of phase 2.

Instability occurred during the first part of phase 1, leading to a progressive decrease of TA and of the methane production, along with an increase of the digestate VS to TS ratio.

Literature findings indicate high requirements of micro-nutrients (in particular Fe, Zn, Ni, Cu, Co, Mo, Mn, and W) for the anaerobic digestion of food waste, especially under thermophilic conditions, due to the lack of bioavailable nutrients contained in this substrate (Romero-Güiza et al., 2016; Yirong et al., 2015; Qiang et al., 2013; Takashima et al., 2011). Accordingly, the minimum concentration of each nutrient necessary to maintain the digestion stability was calculated. Three mineral mediums containing macro and micro-nutrients were prepared and periodically dosed in the reactors to keep the computed minimum concentrations constant. The mediums compositions and dosages are reported in Section 3 of SM. Besides, when required, $NaHCO_3$ was dosed to maintain the TA of the systems higher than 3 $gCaCO_3/L$.

2.4.3 Statistical analysis

Statistical tests were performed to compare the reactors in terms of methane productions per unit of mass of fed COD. First, reactors were compared in the last part of phase 1 to verify the absence of statistically significant differences before the phase 2. Moreover, the differences in the phase 2 were evaluated. The software SPSS v.26 was used to support the analysis.

For each comparison, the normality of the specific methane production distribution of each reactor was numerically verified by applying the Shapiro-Wilk test (more reliable for small samples size) with a 0.05 significance level. Since the normality of distributions was not always satisfied, the non-parametric Kruskal-Wallis test with a 0.05 significance level was applied for the evaluation of differences among reactors. Then, to compare each pair of reactors, post hoc pairwise comparisons were performed using the Mann-Whitney U test, with a $0.05/3 = 0.017$ significance level, adjusted on the total number of tests according to the Bonferroni correction.

2.5 Physical tests

In addition to the effect of biological activity, the degradation of bioplastics could be affected by physical phenomena (e.g. heat effect on bags, hydrophobicity or hydrophilicity of bags). Accordingly, additional tests for the evaluation of physical effects on bioplastic bags were set up. Batch tests were performed under thermophilic conditions (50 ± 0.5°C) in two sterilised reactors (2.4 L) filled with deionised water (2 L) and bioplastic pieces (BSB1 and BDB1). During the tests, reactors were maintained stirred and under anaerobic conditions for 5 weeks.

After 21 days, all the bioplastic pieces were extracted, dried at 35°C, weighed to evaluate their weight losses and re-inserted in the reactors. This operation was repeated at the end of the tests. Moreover, water samples were periodically extracted to measure the COD released from bioplastics.

2.6 Characterisation of tests residues

Residual bioplastic pieces from semi-continuous and physical tests were analysed by means of the DSC to compare the thermal properties of the examined bioplastics before and after the tests.

2.7 Analytical methods

TS and VS were determined in duplicate according to Standard Methods 2540 (Rice et al., 2017). COD and TKN were determined according to Standard Methods 5220 (Rice et al., 2017) and ISO 5663 (ISO, 1984), respectively. The pH was directly measured by means of a portable multiprobe meter (Hach-Lange HQ40D). VFA concentrations were determined according to Standard Methods 5560 (Rice et al., 2017), using a gas chromatograph (Dani Master GC) coupled with a flame ionisation detector. TA was measured by automatically titration with H_2SO_4 up to pH 4.3 (Hach Lange BIOGAS Titration Manager). NH_4^+ was measured using spectrophotometric test kits (Hach-Lange) on 0.45 μm filtered samples. The biogas composition was determined using a gas chromatograph (DANI Master GC Analyser equipped with two columns HayeSep Q and Molesieve 5A).

The water swelling tests were performed by conditioning the bags in a vacuum oven (50°C) for one hour and then immersing them into distilled water at constant temperature. Then, they were periodically recovered, dabbed with damp blotting paper to remove the water in excess and weighed. The swelling of bags was calculated as the percentage increase in weight compared to the conditioned weight, when the plateau weight was reached. Due to the subjectivity of the procedure, the test was repeated seven times for each bag by different operators.

The DSC was performed in three runs (from 25°C to 200°C, from 200°C to -50°C, and from -50°C to 200°C at 20°C/min) with a DSC 823e-Mettler Toledo.

3 Results

3.1 Substrates characterisation

Table 3 reports the results of the substrates characterisation.

Table 3 Results of the substrates characterisation.

Parameter	**BSB1**	**BSB2**	**BDB1**	**BDB2**	**PB**		**Synthetic food waste***
					main bag	**bottom**	
Weight (g/bag)	14.3	13.1	10.3	7.08	17.4	4.78	-
TS (g/kg)	994 ± 1	983 ± 1	980 ± 2	981 ± 2	948 ± 1	952 ± 1	104 ± 1
VS (g/kg)	799 ± 1	863 ± 1	977 ± 2	975 ± 1	861 ± 3	811 ± 2	99.7 ± 0.5
COD (g/kg)	1,364 ± 5	1,423 ± 116	1,644 ± 56	1,577 ± 16	1,041 ± 71	925 ± 99	129 ± 1
TKN (gN/kg)	1.25 ± 0.20	1.13	0.810 ± 0.047	0.758	1.94 ± 0.14	2.13 ± 0.10	2.49 ± 0.02
NH_4^+ (mgN/L)	-	-	-	-	-	-	116 ± 44
pH	-	-	-	-	-	-	4.21 ± 0.01
TA ($mgCaCO_3/L$)	-	-	-	-	-	-	103 ± 23
VFA ($mgCH_3COOH/L$)	-	-	-	-	-	-	3,611 ± 356

* Including the water added to allow for the components shredding. Without the water, the synthetic food waste had the following characteristics: 95.9% VS/TS, 19.3% TS, 4.61 gN/kg TKN.

The DSC performed on bioplastic bags showed the PBAT melting and crystallisation peaks (100 - 125°C and 50 - 100°C) and the starch melting peak (150 - 175°C) in all the bioplastic bags (Figure 1). These results confirm the findings of Elfehri Borchani et al. (2015) and the DSC tests on PBAT/starch blends reported in Seligra et al. (2016) and Garalde et al. (2019).

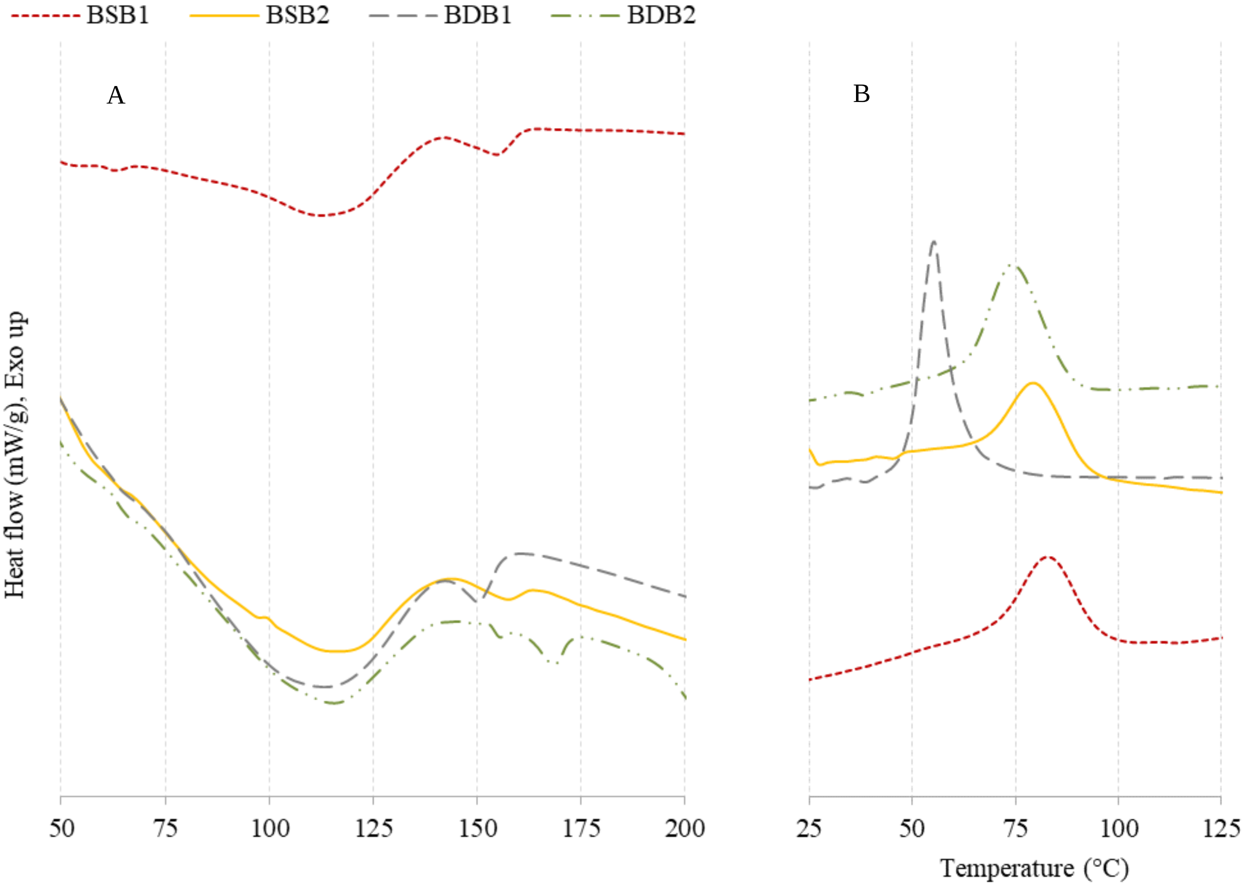


Figure 1 DSC thermograms of melting (A) and crystallisation (B) of tested bioplastic bags.

Examining the results of the water swelling tests, the conditioning stage indicated a different humidity sorption from the environment. In detail, a lower weight loss was observed for BSB1 (-0.3%), compared to -0.8% of the other shopper (BSB2) and to the losses of the dedicated bags (-1.8% for both BDB1 and BDB2). At the end of the tests, BSB1 showed a water sorption significantly lower compared to the other bags (BSB2 +80%; BDB1 +260%; BDB2 +170%). Moreover, for BSB2 and especially for BDB1 and BDB2, most of the weight increase was related to the absorbed water; on the contrary, for BSB1, part of the weight increase was associated to water drops adherent to the surface after the dabbing operation (Figure S4 of SM).

3.2 Inoculum characterisation

The digestate used as inoculum in the BMP and semi-continuous tests showed the following characteristics: TS = 23.6 ± 0.2 g/kg, VS = 13.4 ± 0.2 g/kg, COD = 17.7 ± 0.1 g/kg, TKN = 2.10 ± 0.11 gN/kg, NH_4^+ = 1.22 ± 0.05 gN/L, pH = 8.00 ± 0.08, TA = 7.93 ± 0.37 $gCaCO_3/L$, VFA = 83 ± 22 $mgCH_3COOH/L$.

3.3 BMP tests

Table 4 reports, for each substrate, the main results of BMP tests, including the anaerobic degradability, computed assuming a theoretical methane production of 330 $NmLCH_4/gCOD$ (6% of COD used for cell growth). The BMP tests curves are represented in Figure 2.

Table 4 BMP tests results: final BMP mean ± standard deviation; coefficient of variation (CV); anaerobic degradability on COD basis; test duration; time to get the 50% ($t_{50\%}$) and the 90% ($t_{90\%}$) of the final BMP.

Substrate	**Final BMP** ($NmLCH_4/gVS$)	($NmLCH_4/gCOD$)	**CV (%)**	**Anaerobic degradability (%)**	**Test duration (days)**	$t_{50\%}$ **(days)**	$t_{90\%}$ **(days)**
BSB1	473 ± 15	277 ± 9	3.2	84	42	22	35
BSB2	472 ± 26	286 ± 16	5.6	87	56	6	42
BDB1	392 ± 33	233 ± 19	8.3	71	42	8	35
BDB2	497 ± 24	307 ± 15	4.8	93	45	7	37
PB	290 ± 19	243 ± 16	6.6	74	23	3	6
Food waste	418 ± 7	323 ± 5	1.6	98	21	2	6

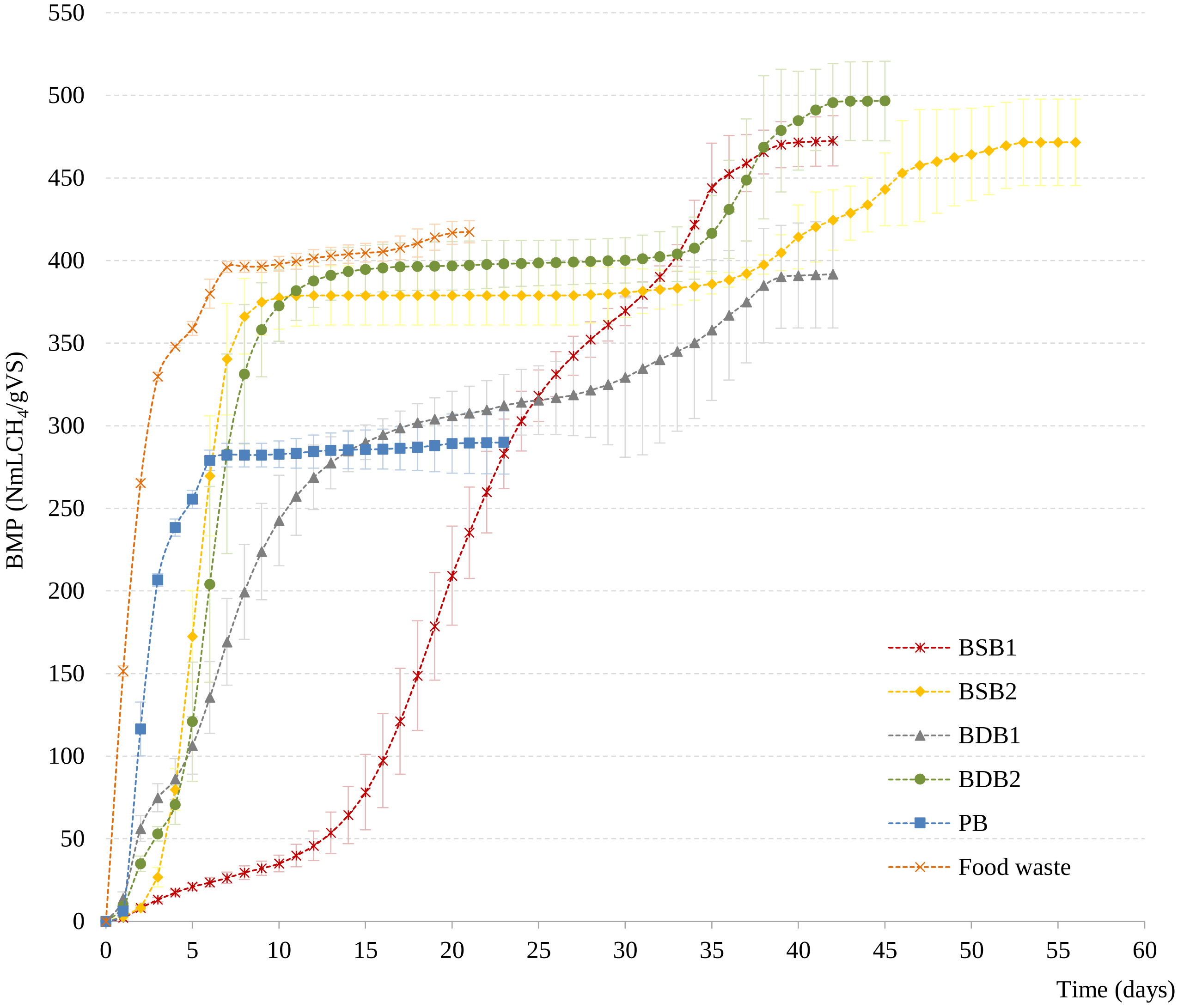


Figure 2 Cumulative net specific methane production (at T = 0°C and P = 1 atm) of BMP tests as a function of time (mean and standard deviation of triplicates).

Comparing the bags, the final BMP values of bioplastics are between 35% and 71% higher than that of paper, while the anaerobic degradability of the two materials is more similar.

As regards the digestate sieving at the end of tests, no residues of substrates bigger than 0.25 mm were detected, except for BDB1, for which non negligible amounts of material were held by the 2, 0.5, and 0.25 mm sieves (Figure S5 of SM). Accordingly, the final BMP of BDB1 is between 17% and 21% lower than that of the other bioplastic bags. The residues are mainly constituted by the bioplastic lace integrated in the bag to be used for its closing, distinguishable by the different colour.

As regards the food waste, which turned out to be almost completely degradable (98%), the final BMP resulted comparable to findings of previous studies on the organic fraction of municipal solid waste: 430 NmLCH$_4$/gVS (Chuenchart et al., 2020), 410 - 460 NmLCH$_4$/gVS (Li et al., 2016), 300 - 570 NmLCH$_4$/gVS (Davidsson et al., 2007).

3.3.1 Kinetic analysis

The degradation of paper is much faster than that of bioplastics: 90% of the final BMP in reached in 6 days, in comparison with at least 35 days needed for bioplastics.

In detail, three out of the four bioplastic bags (BSB2, BDB1, and BDB2) reached 50% of the final BMP in less than 8 days and the subsequent slowing down is due to the stepped trend, typically observed for starch-based biopolymers (Dolci et al., 2021; Cho et al., 2011; Mohee et al., 2008; Russo et al., 2009). On the contrary, the kinetic behaviour is significantly different for BSB1, with only 6% of the final BMP reached in the first 8 days; while 50% was reached in 22 days, suggesting that the initial hydrolytic phase is slower than that of the other bioplastics.

According to the kinetic analysis reported in Section 2.1 of SM, PB data are fitted with the Gompertz model, accounting for the initial lag phase required by enzymes to hydrolyse complex carbohydrates, such as cellulose (Li et al., 2019). The same model can be used to describe BSB1 data. As regards the other bioplastic bags, the pronounced stepped trend is accurately fitted with a “double Gompertz” model.

Assuming 6% of COD used for cell growth, the models allowed to compute degradation efficiencies of 56% (BSB1), 69% (BSB2), 44% (BDB1), 68% (BDB2), and 70% (PB) achievable in an ideal CSTR digester with a HRT corresponding to that selected for the semi-continuous tests (21 days).

Similarly to what observed for PB, the food waste degradation is sensibly fast: 2 and 6 days are enough to reach 50% and 90% of the final BMP, respectively. According to the kinetic analysis reported in Section 2.1 of SM, food waste data are fitted with the Cone model, allowing to compute a 86% degradation efficiency in an ideal CSTR digester with a HRT of 21 days.

3.4 Semi-continuous tests

3.4.1 Phase 1

In the second part of phase 1, the dosage of nutrients solutions and $NaHCO_3$ allowed to achieve a similar and stable behaviour for the four reactors. It is worth noting that in full-scale plants digesting food waste nutrients and TA are often supplied by co-substrates (e.g. manure and sewage sludge) (Xu et al., 2018). In detail, in all the reactors the pH stabilised between 7.1 and 7.4, the VFA to TA ratio resulted always below 0.1 $mgCH_3COOH/mgCaCO_3$, and the ammonium was stable at values lower than 700 mgN/L. Moreover, the average specific methane production of the four reactors resulted in the range 294 - 304 $NmLCH_4/gCOD$ (380 - 393 $NmLCH_4/gVS$) with a volumetric methane content in the biogas always higher than 65%. These parameters are typical of a stable food waste anaerobic digestion process (Cecchi et al., 2005). The observed methane productions correspond to a good anaerobic degradability of food waste, between 89% and 92%, consistent with the estimates from

BMP tests for an ideal CSTR digester (86%). Regarding the extracted digestate, TS were observed in the range 0.8% - 1.2%, with a VS to TS ratio always lower than 65%.

Moreover, the behaviour of the four reactors in terms of specific methane productions distributions was verified to be statistically the same in the phase 1. In detail, the Kruskal-Wallis test showed the following results: H = 0.957; asymptotic significance = 0.812; mean rank: reactor 1 = 43.0, reactor 2 = 44.0, reactor 3 = 38.1, reactor 4 = 44.9.

3.4.2 Phase 2

As regards the overall phase 2, characterised by the introduction of bags samples in reactors 1 (BSB1), 2 (BDB1), and 3 (PB), all the parameters were found to be in the ranges indicated for the last part of phase 1 in all the reactors (Tables S8 to S11 of SM).

The average specific methane productions of the reactor 4 fed with only food waste resulted equal to 303 NmLCH_4/gCOD, corresponding to a 92% anaerobic degradability (Table 5), comparable to what observed during the last part of phase 1.

To better evaluate the influence of collection bags on the digestion process, results for phase 2 are reported separately for the first part (days 74 - 113, phase 2A) and the second part (days 114 - 147, phase 2B). The duration of the phase 2A (2 HRT) was enough to achieve more than 99% of the estimated degradation efficiency at the steady state for all the substrates, according to their mass balances.

The first comparison among the bags was made in terms of methane production and anaerobic degradability. In the phase 2A, the substitution of 11.5% of COD with bags led to reductions equal to 7.6%, 7.7%, and 2.8% in reactors 1, 2, and 3, respectively, compared to the reactor 4.

Differences among reactors in terms of specific methane production distributions in the 10 feed cycles of the phase 2A were then statistically tested. In detail, the Kruskal-Wallis test showed the following results: H = 15.4; asymptotic significance = 1.49E-3; mean rank: reactor 1 = 11.9, reactor 2 = 11.9, reactor 3 = 22.3, reactor 4 = 27.9. Accordingly, the methane production resulted statistically different in at least one pair of reactors. Subsequently, a post hoc procedure was carried out for the pairwise multiple comparison. In detail, the Mann-Whitney U test was performed (the Bonferroni correction was applied reducing the significance level to 0.05/3 = 0.017 according to the total number of tests). Table S12 of SM reports the results of pairwise tests. Differences in methane productions resulted statistically significant between each reactor fed with the bioplastic bags and the reactor fed with only food waste. Moreover, the asymptotic significances of tests between each reactor fed with the bioplastic bags and the reactor fed with the paper bag resulted slightly above the significance level calculated according to the Bonferroni correction.

During the phase 2B, reductions equal to 9.9% (reactor 1), 8.0% (reactor 2), and 1.2% (reactor 3) were observed compared to the reactor 4, in terms of methane production. Figure 3 reports the boxplots of the specific methane productions in the 10 feed cycles of the phase 2A (Figure 3A) and in the 9 feed cycles of the phase 2B (Figure 3B), for the four reactors.

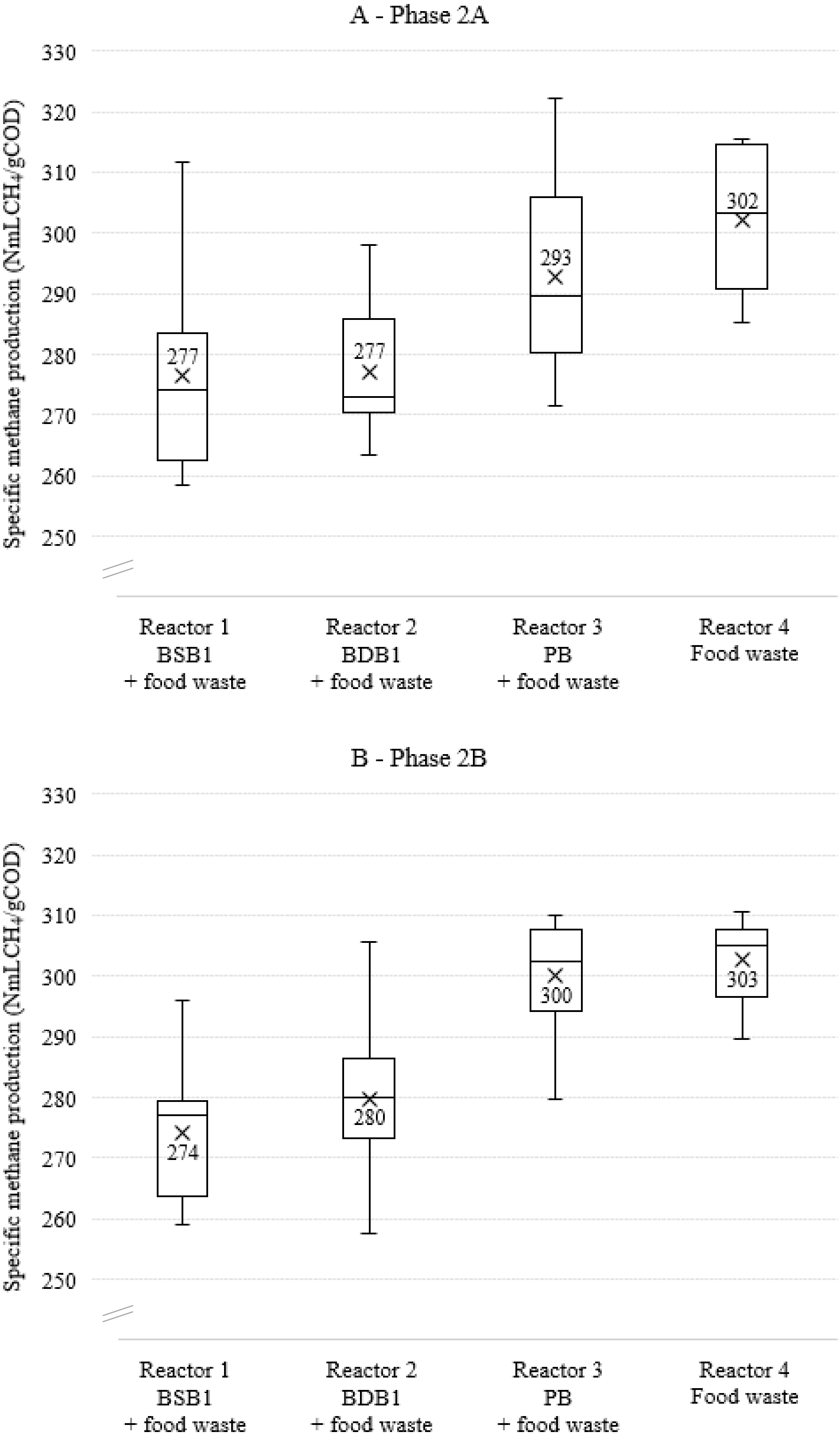


Figure 3 Boxplots of the specific methane productions (at T = 0°C and P = 1 atm) for the four reactors of the semi-continuous tests (A - phase 2A and B - phase 2B). X represent the means of specific methane productions.

In addition to differences in terms of methane production, the transient state during the phase 2A (Figure 3A) is evidenced by interquartile ranges of the boxplots higher than those observable in the phase 2B (Figure 3B).

Differences were then statistically tested with the Kruskal-Wallis test: the methane production resulted statistically different in at least one pair of reactors (H = 20.3; asymptotic significance = 1.50E-4; mean rank: reactor 1 = 9.11, reactor 2 = 12.3, reactor 3 = 25.1, reactor 4 = 27.4).

Results of the post hoc procedure, performed with the Mann-Whitney U test (applying the Bonferroni correction), are shown in Table S12 of SM. Differences in methane productions resulted statistically significant between each reactor fed with the bioplastic bags and the reactor fed with only food waste and between each reactor fed with the bioplastic bags and the reactor fed with the paper bag.

In addition to the comparison in terms of methane production, the physical status of undigested substrates was examined. The digestate sieving during phase 2 showed negligible residues from reactor 4 fed with only food waste. On the contrary, several undigested pieces of BSB1 and BDB1, similar in shape and colour to the fed pieces, were observed (Figures 4 and S18). As regards PB, only small amounts of residues were retained during sieving, in which single pieces were not detectable.

Figure 4 Pieces fed to semi-continuous tests of BSB1 (A), BDB1 (B), and PB (C). Undigested pieces of semi-continuous tests washed and dried of BSB1 (D) and BDB1 (E). Undigested pieces of semi-continuous tests of PB (F).

As regards bioplastic bags residues, the overall mass of undigested pieces resulted equal to 93%

(BSB1) and 69% (BDB1) of the inserted weight. The similarity in shape observed between fed and undigested bioplastics is confirmed by the number of residual pieces with a surface equal to at least ¾ of that of fed substrates, equal to 96% (BSB1) and 98% (BDB1).

3.5 Physical tests

The physical tests indicated that BDB1 is more rapidly and effectively affected by physical agents. In detail, it showed a 16% weight loss after 21 days, with no further changes until the end of the test. Moreover, after the same time, the pieces started to break (Figure S19 of SM). On the contrary, only 4% of the original weight was lost by BSB1 in 21 days, reaching 7% at the end of the test.

Accordingly, an immediate release of the COD of the substrate in water was observed for BDB1 (7% after one week, with no further changes during the test). For BSB1, no release was observed in 21 days (<1%), while a 2% release was observed at the end of the test.

3.6 Characterisation of tests residues

Figure 5 shows the results of the DSC performed to compare the characteristics of bioplastics before and after the tests.

The comparison for BSB1 did not show significant differences among the pieces before the tests, after the semi-continuous test, and after the physical test. Only a small shift of the melting and crystallisation peaks temperatures was observed for pieces after both the tests, compared to pieces before the tests. This result agrees with the small and comparable weight losses observed in the semi-continuous (7%) and physical tests (7%).

As regards BDB1, more significant differences were observed. In detail, the DSC on pieces after the physical test showed the variation in shape and the shift of melting and crystallisation peaks. This behaviour is further pronounced when analysing the DSC on pieces after the semi-continuous test. Such results are in accordance with the weight losses measured in the semi-continuous test (31%), higher than those observed in the physical test (16%), in turn more relevant compared to weight losses of BSB1 in both the tests (7%).

DSC results confirm a higher effectiveness of biological tests on BDB1 degradation, in addition to the non-negligible physical effects.

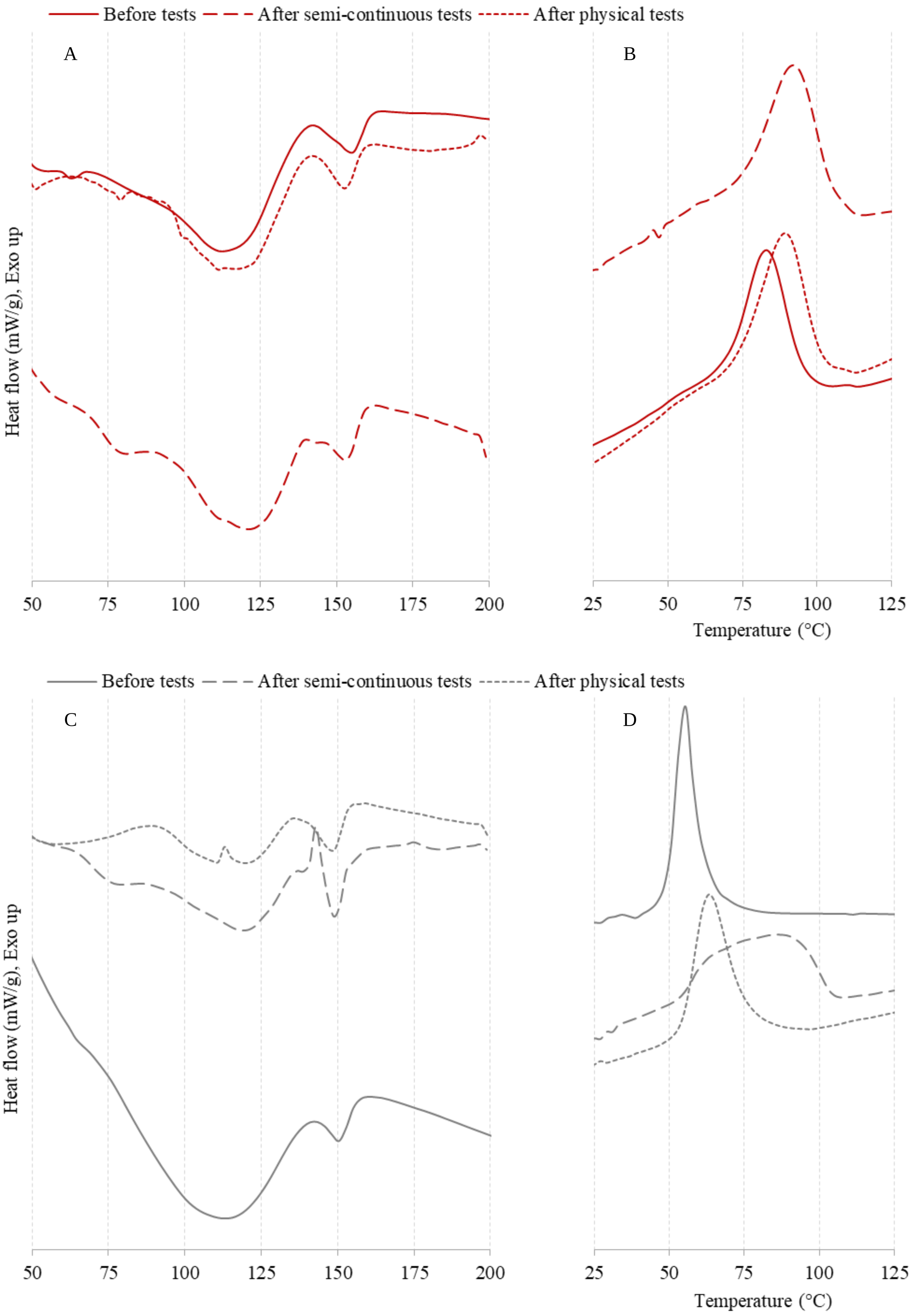


Figure 5 DSC thermograms of BSB1 (A - melting and B - crystallisation) and BDB1 (C - melting and D - crystallisation) before the tests, after the physical tests, and after the semi-continuous tests.

4 Discussion

The results of the BMP tests indicate a good degradability of the four bioplastic bags (71% - 93%), corresponding to final BMPs in the range 392 - 497 $NmLCH_4/gVS$, significantly higher than the literature findings on the Mater-Bi® polymer tested under thermophilic conditions. In detail, results of the present study are 49% to 89% higher than the best results obtained by Dolci et al. (2021) (between 127 and 263 $NmLCH_4/gVS$, with a corresponding anaerobic degradability in the range 22% - 57%). Moreover, an increase between +59% and +101% is observed compared to the result of Cazaudehore et al. (2019) (247 $NmLCH_4/gVS$), and at least +111% compared to the findings of Calabrò et al. (2020) (up to 186 $NmLCH_4/gVS$). Even higher increases (at least +247%) are observed in comparison with outcomes of less recent studies such as Vasmara and Marchetti (2016) (113 $NmLCH_4/gVS$). Concerning PB, the final BMP of 290 $NmLCH_4/gVS$ and the 74% degradability confirm its high compatibility with the anaerobic process observed in Dolci et al. (2021) (262 $NmLCH_4/gVS$; 64% degradability).

As regards the semi-continuous co-digestion conditions, the degradability of bioplastic bags is significantly lower than the estimates from BMP tests, in particular for BSB1 (Table 5). Since the beginning of phase 2 (phase 2A), the anaerobic degradability of BSB1 is 26 percentage points lower than the estimate; the result of the phase 2B shows a further worsening (-18 percentage points). On the contrary, similar results in phases 2A and 2B are observed for BDB1 (15 and 17 percentage points lower than the estimate, respectively).

Table 5 Anaerobic degradability on COD basis of each substrate tested in both BMP tests and semi-continuous tests, assuming 6% used for growth. To compute the degradability of bags in semi-continuous tests, the food waste contribution to methane production was subtracted according to the results obtained in the reactor 4 fed with only food waste.

Substrate	**Anaerobic degradability (%)**			
	BMP tests		**Semi-continuous tests**	
	Final BMP	**Estimate for an ideal CSTR (HRT of 21 d)**	**Phase 2A (days 74 - 113)**	**Phase 2B (days 114 - 147)**
BSB1	84	56	30	12
BDB1	71	44	29	27
PB	74	70	69	82
Food waste	98	86	92	92

On the contrary, at the beginning of phase 2, the result for PB is in line with the estimate. In the phase 2B, a relevant increase is observed (+13 percentage points), leading to an anaerobic degradability 12 percentage points higher than the estimate for an ideal CSTR digester and even 8 percentage points higher than the ultimate anaerobic degradability measured with BMP tests. These results suggest some effect of biomass acclimation or synergistic effects given by the co-digestion of PB and food waste.

Overall, the semi-continuous tests outcomes indicate a pronounced difference between paper and bioplastic bags at the steady state, confirming the importance to support BMP tests with analyses simulating the actual operating conditions.

In addition to the differences between paper and bioplastics, the tests indicate a different behaviour of the examined bioplastic bags. Although BSB1 shows a higher final BMP, its kinetic behaviour in BMP tests is characterised by a lag phase lasting up to more than 10 days. In the same period, BDB1 reaches more than 60% of the final BMP. In addition, there are significant differences in the anaerobic degradability in semi-continuous tests (Table 5), confirmed by the different weight losses of the tested substrates (31% for BDB1 and 7% for BSB1). Differently from what observed in terms of degradability and weight losses, the number of residual pieces with a surface equal to at least ¾ of that of fed substrate is similar for both bioplastic bags (>95%), suggesting that the observed degradation of BDB1 is not related to a material breaking.

Comparing the results to literature findings, BDB1 allows for a significantly higher weight loss than those observed in Zhang et al. (2018) for two starch-based bioplastics (7.9% and 2.1%), while BSB1 shows lower differences. For both BDB1 and BSB1, the residual number of pieces is similar to what observed in Zhang et al. (2018) for the two starch-based bioplastics (97% and 96%). Globally, the results of Zhang et al. (2018) indicate a little or no evidence of degradation of the two tested starch-based bioplastics[1]. Anyway, it is worth noting that the study by Zhang et al. (2018) was performed under mesophilic conditions, significantly less effective for the anaerobic degradation of this type of bioplastics (Section 1).

The differences observed between BSB1 and BDB1 in both the BMP and in the semi-continuous tests are confirmed by the physical tests, suggesting an influence of physical agents on the degradation in the biological tests and pointing out that the magnitude of the effect of physical agents depends on the bag type. This thesis is also supported by the results of water swelling tests, showing a significantly higher water sorption for BDB1 compared to BSB1 (+170%).

The different results of all the tests performed on the two types of bioplastic bags reveal that their specific features, such as the thickness or the inks and dyes added to the Mater-Bi® polymer, significantly affect the behaviour when subjected to physical and biological agents.

5 Conclusions

BMP tests and semi-continuous co-digestion tests together with food waste were performed on

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[1] Evaluations in Zhang et al. (2018) are based on the bioplastics weight loss during digestion and on the ratio between undigested and fed pieces. The methane production is not measured.

bioplastic and paper bags for the collection of food waste to evaluate their behaviour under anaerobic thermophilic conditions.

For bioplastic bags, the BMP tests indicated a good degradability (>71%); on the contrary, co-digestion semi-continuous tests did not show comparable results, being the anaerobic degradability lower than 27% with undigested bag pieces still detectable in the effluent digestate. Moreover, a different behaviour was observed in semi-continuous tests for two different types of bioplastic bags, i.e. BSB1 and BDB1. This suggests to extend the evaluation to other commercial types of bioplastic bags, e.g. BSB2 and BDB2 for which, despite showing BMP tests results similar to BSB1 and BDB1, significant differences could be observed in semi-continuous tests.

According to the physical, DSC, and water sorption tests, the difference observed for the bioplastic bags can be at least partially explained by the effect of physical factors on bags.

On the contrary, the paper bag presents interesting characteristics, because its anaerobic degradability in the co-digestion semi-continuous tests (82%) resulted higher than that observed in the BMP tests (74%). These results indicate a good compatibility with the anaerobic digestion process, leading to some potential energy benefits because of the additional generation of biogas. By assuming an average content of 2 kg of food waste (Dolci et al., 2021), paper bags allow for an 8% increase in the methane production per mass unit of food waste in addition to that obtained from the sole food waste. On the contrary, for the tested bioplastic bags, this increase is lower than 2%.

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CRediT authorship contribution statement

Giovanni Dolci: Conceptualisation, Formal Analysis, Investigation, Methodology, Writing - Original Draft, Writing - Review & Editing. **Valeria Venturelli**: Conceptualisation, Formal Analysis, Investigation, Methodology, Writing - Original Draft, Writing - Review & Editing. **Arianna Catenacci**: Conceptualisation, Formal Analysis, Methodology, Writing - Review & Editing. **Riccardo Ciapponi:** Formal Analysis, Investigation. **Francesca Malpei**: Resources, Supervision, Writing - Review & Editing. **Stefano Ettore Romano Turri**: Resources, Supervision. **Mario Grosso**: Resources, Supervision, Writing - Review & Editing.

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