PLASTIC PARTICLE CONTAMINATION IN SUBSTRATE AND FRASS OF BLACK SOLDIER FLY LARVAE CULTIVATION IN COMMUNITY SOLID WASTE RECYCLING CENTERS

KONTAMINASI PARTIKEL PLASTIK PADA SUBSTRAT DAN FRASS BUDIDAYA LARVA LALAT TENTARA HITAM DI KOMUNITAS SENTRA DAUR ULANG SAMPAH PADAT

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Abstract

Bioconversion of solid waste (SW) by black soldier fly (BSF) is a promising sustainable SW treatment approach. However, insufficient segregation may lead to plastic contamination in BSF larvae (BSFL) substrates, compromising the quality of BSFL-derived products and potentially introducing plastic particles into the food chain. This study aimed to investigate plastic abundance in SW from different sources and determine abundance of plastic particles in BSFL substrates and frass. The research was conducted at the Jambangan Recycling Center (JRC) and the Wonorejo Composting Center (WCC) in Surabaya, Indonesia. The results showed that 74% of SW at JRC was suitable for BSFL substrate, while only 36.62% of SW can be used in WCC. The amount of plastic abundance in the BSFL substrate in JRC and WCC were 2.28% and 7.63%, respectively. At JRC, 12 macroplastic (MaP), 9 mesoplastic (MeP), and 334 microplastic (MP) particles were detected per kilogram dry-weight (DW) of BSFL substrate, whereas 250 MP particles were found in frass. The insufficient SW segregation at the WCC has led to an abundance of plastic particles per kilogram DW of BSFL substrate (i.e., 40 MaP, 35 MeP, and 734 MP particles) and frass (484 MP particles). Plastic transparent films were mostly found in both substrates and frass. The size of MaPs typically ranged from 2.5 to 10 cm, while MPs predominantly fell within the 1 µm to 1 mm range. These findings highlight the importance of source segregation to reduce plastic contamination and enhance the efficiency and safety of BSF bioconversion process.

Keywords: *bioconversion, black soldier fly, food waste, microplastics, SW segregation.*

Abstrak

Biokonversi sampah padat (SP) oleh larva lalat tentara hitam (BSF) merupakan pendekatan pengolahan SP berkelanjutan yang menjanjikan. Namun, pemilahan sampah yang tidak memadai dapat menyebabkan kontaminasi plastik dalam substrat larva BSF (BSFL), yang dapat menurunkan kualitas produk turunan BSFL serta berpotensi memasukkan partikel plastik ke dalam rantai makanan. Penelitian ini bertujuan untuk mengkaji kelimpahan plastik dalam SP dari berbagai sumber serta menentukan kelimpahan partikel plastik dalam substrat dan frass BSFL. Penelitian dilakukan di Sentra Daur Ulang Jambangan (JRC) dan Pusat Pengomposan Wonorejo (WCC) di Surabaya, Indonesia. Hasil penelitian menunjukkan bahwa 74% SP di JRC layak digunakan sebagai substrat BSFL, sedangkan di WCC hanya 36,62% SP yang dapat dimanfaatkan. Kandungan plastik dalam substrat BSFL di JRC dan WCC masing-masing sebesar 2,28% dan 7,63%. Di JRC, terdeteksi 12 partikel makroplastik (MaP), 9 partikel mesoplastik (MeP), dan 334 partikel mikroplastik (MP) per kilogram bobot kering (BK) substrat BSFL, sedangkan frass mengandung 250 partikel MP. Ketidakoptimalan pemilahan SP di WCC menghasilkan jumlah partikel plastik yang lebih tinggi per kilogram BK substrat BSFL (yaitu 40 MaP, 35 MeP, dan 734 MP) serta frass (484 partikel MP). Film plastik transparan paling sering ditemukan baik pada substrat maupun frass. Ukuran MaP umumnya berkisar antara 2,5 hingga 10 cm, sedangkan MP sebagian besar berada pada rentang ukuran 1 µm hingga 1 mm. Temuan ini menekankan pentingnya pemilahan sampah dari sumbernya untuk mengurangi kontaminasi plastik dan meningkatkan efisiensi serta keamanan proses biokonversi oleh BSF.

Keywords: biokonversi, lalat tentara hitam, sampah makanan, mikroplastik, pemilahan sampah.

1. INTRODUCTION

Solid waste (SW) generation is strongly correlated with population growth and economic development. In 2016, 2.01 billion tons of waste generated globally, with an average of 0.74 kilograms per capita per day (Kaza et al., 2018). By 2020, this figure reached 2.1 billion tons per year and is projected to increase by 56%, at approximately 3.8 billion tons by 2050 (United Nations Environment Programme, 2024). Between 2000 and 2019, Indonesia's food loss and waste ranged from 115 to 184 kilograms per capita per year (Ministry of National Development Planning of the Republic of Indonesia, 2021). This upward trend has persisted. In 2024, food waste (FW) was still the largest proportion of the country's total SW. In that year, 39.36% of the total 33.79 million tons of SW was FW, followed by plastic waste at 19.64% (Ministry of Environment and Forestry of the Republic of Indonesia, 2024). Given the increasing volume of FW, sustainable and efficient SW management strategies are urgently needed.

Recently, black soldier flies (BSF) (*Hermetia illucens*) have gained attention as effective bioconversion agents for managing organic waste. Through BSF bioconversion, circular economy can be implemented, as the larvae can transform a various organic material into protein-rich biomass and frass. This frass can be used as compost (Indri et al., 2021; Shao et al., 2023; Siddiqui et al., 2024). From one ton of household FW, approximately 201 kg of dry matter of BSF frass can be produced (Lalander et al., 2020). BSF frass is rich in essential macronutrients, including carbon (35.8%), nitrogen (2.2%), phosphorus (0.5%), and potassium (0.7%) (Kawasaki et al., 2020). Additionally, its organic composition and active compounds, enhance soil health by stimulating beneficial microbes and strengthening plant resilience against pathogens (Gebremikael et al., 2020).

Proper segregation of SW is needed to produce high-quality BSF-derived products. According to Farahdiba et al. (2023), effective separation and pre-treatment can improve SW utilization efficiency and the recovery value significantly. Segregation serves as a critical initial step to ensure that the SW is free from hazardous substances and inorganic materials. In practice, although the BSF substrate is required to be organic and biodegradable (Dortmans et al., 2017), plastic contamination may still be unavoidable. As a result of environmental exposure to sunlight, air, heat, and moisture, plastic waste can degrade into smaller particles such as microplastics (MPs) and nanoplastics (NPs) (Bajt, 2021).

Previous studies reported that plastic contamination in the BSF substrate had a negligible impact on larval survival rates (Beale et al., 2022; Dam et al., 2024; Romano & Fischer, 2021). While the presence of plastics did not significantly affect larval development, it might induce oxidative stress in BSF. The stress was shown by elevated levels of homocysteine—a byproduct of glutathione metabolism—and 4-aminobenzoic acid. This phenomenon occured along with the downregulation of pyrimidine metabolism and upregulation of purine metabolism (Beale et al., 2022). De Filippis et al. (2023) also demonstrated that exposure to plastics could alter the BSF gut microbiome at the species and strain levels. Consequently, genes associated with polymer chains degradation were highly enriched in the metagenomic profiles.

The introduction of various 6 types plastic particles into BSF rearing substrates was done by Beale et al. (2022). The study concluded that MPs in insect rearing substrates generally had minimal effects on larval weight. However, exposure to PLA-MPs led to a 28.7% reduction in BSF larval weight relative to the supplemented control (Piersanti et al., 2024). BSFL, depending on factors such as particle concentration, mouthpart size, and developmental stage, may ingest microplastics (Lievens et al., 2023). However, BSFL could effectively excreting these particles prior to pupation stage (Heussler et al., 2024). Nevertheless, further investigation is necessary to determine the extent to which BSF larvae ingest plastic particles within the rearing substrate.

This study was conducted in two SW recycling centers, Jambangan Recycling Center (JRC) and Wonorejo Composting Center (WCC) in Surabaya. The JRC processes approximately 831 kg of commingled SW per day, of which 302 kg are FW. This SW was collected from various sources, including households, catering services, traditional markets, and community associations. In contrast, the WCC received 7.65 tons of FW daily from various sources. Including the Keputran traditional market, the Sutorejo recycling center, and several culinary centers (Zahra et al., 2024). Although BSF bioconversion was applied to treat only 14.26% and 0.22% of FW at the JRC and the WCC, respectively, the method was able to reduce 84% and 54% of the initial substrate. Approximately 0.03–1% of the FW was transformed into larvae biomass, whereas 0.09–4.97% was converted into BSF larvae frass (Zahra et al., 2024).

The objective of this study was to investigate the amount plastic waste component in the SW prior to be used as BSFL substrate in the two recycling centers. The abundance, size, color, shape, and type of plastic particles in the BSF substrates and frass was also determined. In addition, this study provides an understanding of strategic plastic waste management approaches. The approaches aim to reduce plastic contamination during bioconversion processes and ensure the quality of BSF-derived products.

2. MATERIALS AND METHODS

2.1 Sampling locations

Solid waste samples used as BSFL substrates were collected from the JRC (7°19'2.74"S, 112°43'0.32"E) and the WCC (7°18'42.34"S, 112°47'23.93"E) in Surabaya, Indonesia (**Figure 1**) for composition analysis. In addition, the BSF frass samples for plastic contamination analysis were also collected from the BSF culture unit in both recycling centers.

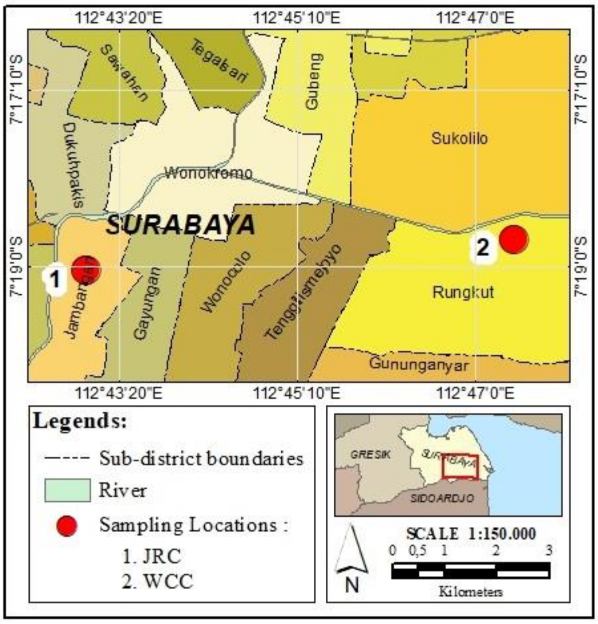

Figure 1. Sampling locations

2.2 Sampling method

The SW samples which were used as BSFL substrate were analyzed according to ASTM D5321-92 method for composition analysis over 8 consecutive days. The SW composition was then categorized into: i) biodegradable organic waste (garden and agricultural waste, fruits, vegetables, and food waste), ii) non-biodegradable organic waste (bones, spines, and eggshells), iii) plastics (PETE/PET, PVC, HDPE, PP, PS), and iv) other materials (cloth, paper, metal, glass, etc.). Composite sampling was conducted at each sampling locations to collect BSF substrate and frass samples over three feeding periods. Not least than 1 kg of substrate and frass samples were collected during this study. Residue of the BSF rearing process was sieved for compost production. The frass samples were collected from the BSF residue after sieving process. The samples were then kept in glass containers and homogenized to ensure reliable and representative results.

2.3 Sampling preparation and identification

The plastic particles in the BSF substrate and frass samples collected from the JRC and the WCC were examined and categorized based on their size. The size categories were macroplastics (MaPs, >2.5 cm), mesoplastics (MePs, 5 mm–2.5 cm), and microplastics (MPs, 1 µm–5 mm) (Lippiat et al., 2013). Afterwards, sample was extracted following the modified National Oceanic and Atmospheric Administration (NOAA) protocol (Masura et al., 2015).

MaP particles (>2.5 cm) were manually collected and quantified. A representative 400 g sample was oven-dried at 90^{o}C for 24 h. To facilitate particle disaggregation, 400 mL of sodium hexametaphosphate solution was added, followed by rapid agitation for 1 h and sieving in mesh sizes of 5 mm (No. 4), 0.5 mm (No. 35), and 0.074 mm (No. 200). To isolate microplastic particles, a 5 M NaCl solution was added to the sample to allow the particles flotation over a 24-h period. Next, a wet peroxide oxidation (WPO) process was performed by adding 10 mL of 0.04 M Fe(II) solution and 10 mL of 30% H_2O_2 to the sample. Next, 75°C heating and 30 minutes stirring was conducted to the sample. Subsequently, sample underwent density separation and filtration, in which the final filtrate was passed through a Whatman GF/C glass microfiber filter (1.2 µm pore size). The filter papers were stored in petri dishes and placed in a desiccator to remain dry and prevent contamination before further analysis.

MeP and MP particles were identified using optical microscopy with a Dino-Lite Edge AM7915 digital microscope and DinoCapture 3.0 software. Based on their physical attributes, including type, shape, size, and color, plastic particles were characterized and classified. Moreover, the abundance of each plastic category present in the BSF substrate and frass were analyzed quantitatively. Plastic abundance was expressed as the number of particles per kilogram of BSF substrate and frass (particles/kg).

3. RESULT AND DISCUSSION

3.1 BSF substrate composition

The composition of the BSF rearing substrate at the JRC and the WCC is illustrated in **Figure 2**. At JRC, the BSF substrate comprised food waste (34.93%), vegetables (34.03%), fruits (27.49%), and garden and agricultural waste (0.49%). Other components consisted of plastic waste (2.28%), cloth (0.17%), and paper (0.05%). In contrast, the BSF substrate at WCC consisted of garden and agricultural waste (86.41%), fruits (4.05%), vegetables (0.33%), and food waste (0.04%). Plastic waste was detected in the sample at approximately 7.63%., Meanwhile, whereas the presence of other components, such as metal, cloth, paper, and miscellaneous items, were also identified at 0.05%, 0.86%, 0.18%, and 0.46%, respectively. Overall, approximately 74.19% of substrate at JRC was biodegradable by BSF larvae, compared to only 36.61% at WCC. A detailed classification of BSF substrate is presented in **Table 1.**

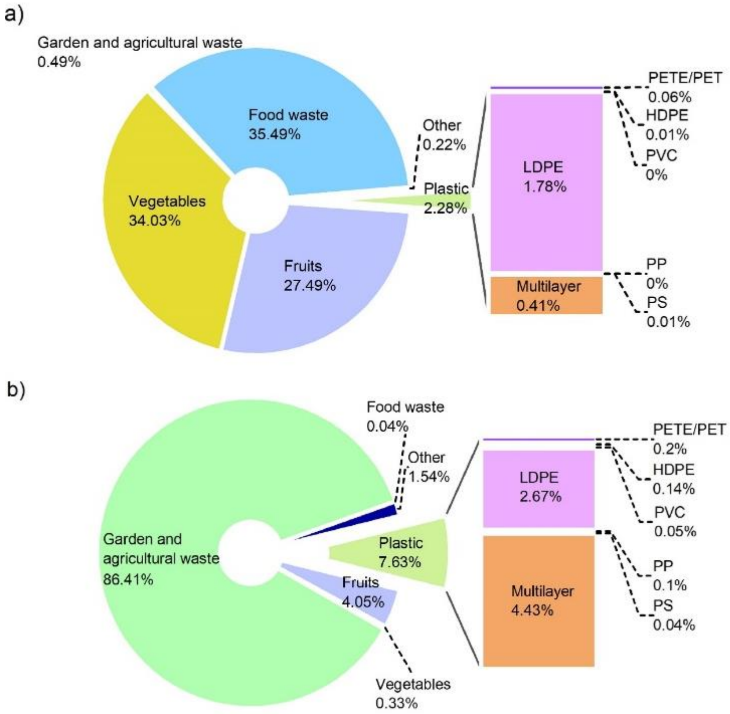

Figure 2. BSFL substrate composition at: a) JRC, b) WCC

Table 1. Classification of BSF substrate

Substrates	Percentage (%)	
	JRC	WCC
Garden and agricultural waste		
- Fiber	0.49	25.92
- Dry leaves	-	8.64
- Plant stems	-	17.28
- Food waste and other residues*	-	34.56
Fruits		
- Fruit peels	11.00	1.42
- Fruit seeds	2.75	0.61
- Rotten fruit*	13.74	1.62
- Fruit residues*	-	0.40
Vegetables		
- Vegetable waste*	25.52	0.03
- Corn cobs	6.81	0.30
- Corn husks	1.70	-
Food waste		
- Rice*	22.70	-
- Meat*	5.24	-
- Tempeh*	1.75	-
- Cooked vegetables*	3.49	-
- Noodle*	1.75	-
- Bones and spines	0.01	0.04
- Eggshells	0.55	-
Plastics	2.28	7.63

Substrates	Percentage (%)	
	JRC	WCC
Other materials	0.22	1.54

*digestible substrate by BSFL

The JRC has consistently implemented the 3R in SW management since 2016. It treated organic SW from households, catering services, traditional markets, and community associations through composting and BSF bioconversion (Zahra et al., 2024). The community awareness of SW handling has provided the availability of sorted substrate for the BSFL (Fadilla & Kriswibowo, 2022). Comparative data showed that the digestible substrate availability in JRC (74%) was higher than that in WCC (36.62%). Additionally, lower plastic contamination in JRC (2.28%), was lower than that in WCC (7.63%). This underscored the effectiveness of Jambangan community-driven SW management in producing cleaner, nutrient-rich substrates for BSFL rearing in JRC.

A detailed comparison of BSFL substrate composition (**Table 1**) revealed that the JRC utilized a more diverse and nutrient-rich substrate profile. The existence of biodegradable organic waste (e.g., rotten fruit 13.74%, vegetable waste 25.52%, rice 22.70%, and minimal lignocellulosic content (e.g., fiber 0.49%) was dominating. In contrast, the WCC relied heavily on garden and agricultural waste, including fiber (25.92%), plant stems (17.28%), and food waste along with other residues (34.56%). These wastes were less digestible for BSFL due to their high cellulose and lignin content.

The nutritional profile of substrates plays a critical role in BSFL development. Nutritious substrates with a minimum 10–15% of protein and10–60% carbohydrate can have a positive influence on the survival, growth, and development of BSFL. Medium-to-high C/N ratio, high volatile solid, and low cellulose, hemicelluloses, and lignin contents can also support the BSFL growth (Barragan-Fonseca et al., 2019; Beesigamukama et al., 2021; Lalander et al., 2019; Mehta et al., 2015). Thus, food waste, including fruit and vegetable waste, can be a potential substrate for BSFL. BSF reared on food waste has 87.2% survival rate, compared to 90.7% for those reared on fruits and vegetables. However, the bioconversion efficiency for both substrates was relatively low, i.e., 13.9% for food waste and 4.1% for fruits and vegetables waste (Lalander et al., 2019). Notably, a mixture of waste tends to improve larval survival rates, pupal weight, and protein content compared to a single substrate composition. This improvement is likely due to a more balance and diverse nutrient composition in the mixed substrate (Mishra & Suthar, 2023; Rehman et al., 2017).

Moreover, the presence of provitamin A-rich substrates (e.g., carrots and pumpkins) in household SW streams enables BSFL to bioaccumulate α- and β-carotene. This contributes to micronutrient recycling within the food chain (Borel et al., 2021). The substrate reliance on high-fiber agricultural waste in WCC poses challenges for BSFL digestion due to limited lignin-degrading enzymatic capacity (Rehman et al., 2025). To address these limitations, microbial fermentation has been shown to enhance substrate digestibility by accelerating lignin degradation (Yu et al., 2024). A previous study (Isibika et al., 2021) reported that BSF reared on banana and orange peels has a bioconversion efficiency of only 2.30–6.80%. Mixing these wastes with other nutrient-rich materials, such as fish waste, has been found to improve BSF bioconversion efficiency by up to 12.3%.

3.2 Plastic waste composition

The composition of plastic waste in FW used as BSFL substrates at JRC and WCC reflected significant differences in upstream SW segregation practices. Plastic waste at the SW used as BSF substrate in JRC comprised five types of polymers. The largest proportion (78.12%) was LDPE, followed by multilayer plastics (18.09%), PETE/PET (2.85%), HDPE (0.49%), and PS (0.45%). This limited diversity indicated a cleaner, more controlled SW stream, as JRC primarily received waste from households and communities with established segregation practices (Fadilla & Kriswibowo, 2022). In contrast, the BSF substrate in WCC contained diverse polymer types, such as PETE/PET, HDPE, PVC, LDPE, PP, PS, and multilayer plastics (**Figure 2**). Within this mixed waste stream, multilayer plastic was the most dominant type (58.11%), primarily originating from food packaging and sachets. LDPE was the second most prevalent polymer (34.99%), mainly derived from plastic bags and similar packaging materials. PETE/PET was present at 2.68%, typically from mineral water bottles. Other plastic types comprised HDPE (1.83%), PP (1.27%), PVC (0.67%), and PS (0.46%). This plastic polymers came from food containers, straws, water pipes, electrical cables, styrofoam, egg trays, and plastic cutlery. WCC's diverse plastic waste was due to SW received from the Sutorejo Recycling Center, where insufficient pre-sorting led to mixed shredding.

3.3 Plastic abundance in BSF substrate and frass

The plastic abundance values in BSF substrate and frass samples at the two locations are compared in **Figure 3**. As observed, substrate samples in JRC had a lower MaPs (12 particles/kg) component than that of in WCC (40 particles/kg). Due to the high MaPs abundance, the abundance values of MePs (35 particles/kg) and MPs (734 particles/kg) in the BSF substrate in WCC exceeded those in JRC. Specifically, JRC had lower values, with 9 MePs particles/kg and 334 MP particles/kg. The presence of MP in the substrates was particularly concerning, as these particles fall within the ingestible size range for BSFL. Previous studies reported that only MP could be ingested by BSFL (Beale et al., 2022; Heussler et al., 2024; Lievens et al., 2022; 2023). The likelihood of ingestion was influenced by particle concentration, larval mouthpart size, and developmental stage (Lievens et al., 2023). Furthermore, while plastic particles could be excreted prior to pupation stage (Heussler et al., 2024), their temporary presence in the larval gut may still disrupt biological functions (Beale et al., 2022).

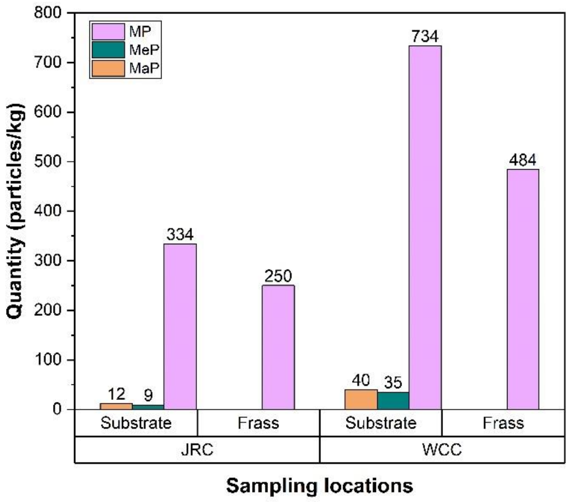

Figure 3. Plastic abundance in the BSF substrate and frass

The MPs abundance in BSF frass in WCC was higher (484 particles/kg) than that in JRC (250 particles/kg), as seen in **Figure 3**. In contrast, MaPs and MePs were absent in the analyzed frass samples, likely due to the mesh sieving process (± 0.5 mm). This process may have facilitated the separation of MaPs and MePs while retaining certain MPs within the sieve. Additionally, MPs' small size enabled ingestion by BSF during digestion, affecting their final distribution in frass fractions. Effective source segregation ensures feed safety and enhances the quality of end-products (Gold et al., 2021). The presence of plastic waste in organic substrates is not only hinder processing efficiency process but also contributes to increase the operational costs. Bottausci et al. (2024) highlighted that plastic contamination adversely affects composting processes, necessitating additional SW management measures. Compost must meet agricultural standards, including proper nutrient content, particle size, and the absence of plastics, metals, and other inorganic contaminants (Huynh et al., 2023). Thereby, the implementation of at source segregation could significantly reduce plastic contamination in the compost products.

3.3.1 Macroplastic distribution

The macroplastic (MaP) distribution at both locations revealed notable variations across four key parameters. These MaPs typically came from food packaging, plastic pieces, toiletries, plastic bags, raffia rope, and others. Mechanical processes, such as shredding, have contributed to the fragmentation of plastics into smaller particle sizes. At the JRC, MaPs within the 2.5-10 cm size category had the highest portion (accounted for 74.29%). Meanwhile, MaPs measuring 10–20 cm and 20–30 cm constituted 22.93% and 2.78%, respectively. Plastic films in the 2.5–10 cm size range, as well as transparent plastic and LDPE, were also more prevalent in the BSFL substrate. Specifically, their respective shares were 68.95%, 46.65%, and 89.53%.

In addition to these dominant characteristics, other MaP features also contributed to the overall distribution profile. Although less abundant, plastic filaments were consistently present in 2.5-10 cm size (2.78%). Plastic films in the 10-20 cm and 20-30 cm ranges accounted for 22.93% and 2.78%, respectively. Similarly, while transparent MaPs were most prevalent, colored MaPs such as white (5.13%), red (41.95%), black (3.70%), and orange (2.56%) were also identified. Furthermore, polymer types such as PP (5.34%), PS (2.56%), and multilayer plastics (2.56%) were found in smaller proportions compared to LDPE.

To further substantiate these observations, comparative data from the WCC were examined. As analyzed, approximately 74.17% of the identified MaPs measured between 2.5–10 cm in size, followed by 20.57% in the 10–20 cm range and 5.26% in the 20–30 cm range. Plastic films (2.5–10 cm), mainly from food packaging and plastic bags, constituted 42.77% of the total MaPs found in BSFL substrate. Among the various colors and type of plastics, transparent films and LDPE were the most prevalent, accounting for 42.07% and 43.17%, respectively. These findings align with the abundance of LDPE illustrated in **Figure 2**. Salmah et al. (2013) noted LDPE's properties- fluidity, flexibility, transparency, and a glossy surface-make it ideal for food packaging, sheets, and films.

In addition to these dominant characteristics, other MaP types were also consistently observed. Plastic filaments were present in the 2.5-10 cm (2.74%), 10-20 cm (3.17%), and 20-30 cm (1.89%) size ranges. Plastic films in the 10-20 cm and 20-30 cm ranges accounted for 14.43% and 1.89%, respectively. Plastic fragments contributed 24.21% (2.5-10 cm), 2.97% (10-20 cm), and 1.48% (20-30 cm), while plastic foam was found in the 2.5-20 cm (4.50%). Colored MaPs such as red (5.31%), white (17.42%), green (6.16%), yellow (8.05%), blue (5.96%), black (6.59%), grey (0.63%), orange (3.77%), brown (1.48%) and pink (2.56%) were also identified, indicating a broader diversity of plastic sources compared to JRC. Furthermore, polymer types such as PVC (11.67%), HDPE (10.64%), PP (14.99%), PS (5.08%), and multilayer plastics (14.46%) were found in smaller proportions compared to LDPE. Nevertheless, no MaP particles were found in BSF frass at both centers, which confirmed the previous hypothesis (Lievens et al., 2022). Larvae are unlikely to ingest MaPs and MePs, as it exceeded their mandibular brush size (380 μm).

3.3.2 Mesoplastic distribution

The mesoplastics (MePs) at both centers typically were derived from the breakdown of larger plastic items. Approximately, 9 MeP particles/kg DW of substrate were detected at the JRC. Transparent MePs was detected at 58.33%, which was the highest in terms of color, followed by red at 41.67%. Moreover, about 100% of MePs at the JRC appeared in film form. In contrast, 35 MeP particles/kg DW of substrate were found at the WCC. Compared to other color, transparent MePs were the most prevalent, making up 27.78%. Furthermore, colored plastics such as white (22.69%), red (4.17%), green (11.11%), blue (23.15%), orange (3.70%), pink (3.70%), and purple (3.70%) were also identified. In terms of shape, MeP film was the most dominant form, accounting for 53.70% of the total, followed by fragment (36.57%) and filament (9.72%).

3.3.3 Microplastic distribution

Microplastics (MPs), defined as plastic particles ranging from 1 µm to 5 mm, were further classified into small microplastics (SMP; 1 µm–1 mm) and large microplastics (LMP; 1–5 mm). MPs identified in BSF substrates at both centers are illustrated in **Figure 4**. At the JRC, 334 secondary MP particles/kg DW of substrate were observed. In contrast, WCC had a higher concentration of 734 MP particles/kg DW, with 97.29% were identified as secondary MPs and 2.71% as primary MPs. Across both sites, SMP comprised most MPs, accounting for 94.71% at JRC and 79.75% at WCC, whereas LMP constituted 5.29%, and 20.25% respectively.

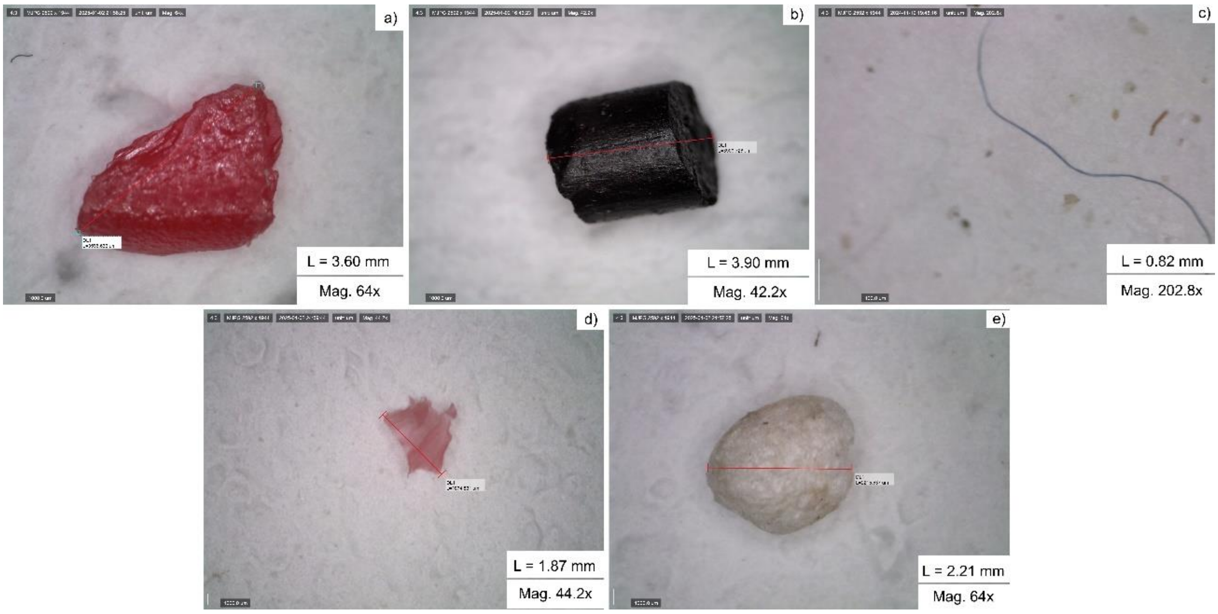

Figure 4. Microplastics found in both locations a) fragment, b) pellet, c) filament, d) film, e) foam

Transparent MPs were the most common identifiable, comprising 45.55% at JRC and 27.55% of MPs at WCC. Colored plastic such as white (8.74%), red (15.01%), green (8.07%), yellow (0.38%), blue (15.56%), black (1.61%), and brown (5.08%) were also identified at JRC. In contrast, WCC showed a broader distribution of colored MPs, such as white (12.60%), red (16.13%), green (6.82%), yellow (1.68%), blue (22.21%), black (8.83%), grey (0.83%), brown (2.93%), pink (0.13%), and purple (0.30%).

Morphological analysis revealed that MPs detected at both centers were mostly in the form of films, fragments, filaments, foam, and pellets. Among these, plastic films were predominant, representing 71.98% at JRC and 66.62% at WCC. Fragment plastics were more prevalent at WCC (15.24%) than at JRC (8.28%), while filaments were found in similar proportions at both locations (11.66% and 11.41%). Foam was more common at JRC (8.08%) compared to WCC (4.02%), and pellets were only detected at WCC

(2.71%). These findings are consistent with the fragmentation behavior of LDPE and other common packaging materials.

Regarding BSFL frass, approximately 250 and 484 MP particles per kilogram DW of frass were identified at JRC and WCC, respectively. Secondary MPs dominated at both centers, accounting for 96.94% at JRC and 100% at WCC. Small microplastic particles (SMP) remained the prevailing size fraction in the frass samples, comprising 91.85% and 95.07% at JRC and WCC, respectively, while large microplastic particles (LMP) contributed 8.15% and 4.93%. These results confirm that BSFL are more likely to ingest and bioaccumulate MPs than MaP and MeP particles, subsequently excreting them before reaching the pupation stage (Heussler et al., 2024; Lievens et al., 2023).

Furthermore, although MP ingestion had no significant impact on larval development or mortality, smaller pupae size was observed (Piersanti et al., 2024), when BSFLs were fed with 20% PVC MP particles. The most concerned issue is the presence of MPs in the frass. When used as compost, these particles may persist in the soil for extended periods. MPs can reduce soil aggregation and lower bulk density, and negatively affecting water and air flow within the soil (Okori et al., 2024). Additionally, they may release adsorbed toxic substances and other chemical contaminants during their lifecycle, posing significant risks to ecosystem health.

4. CONCLUSION

This study demonstrated that SW segregation played a crucial role in reducing plastic contamination in BSF substrates. The amount of biodegradable organic waste that can be digested by BSFL at the JRC and the WCC were 74.19% and 36.61%, respectively. However, insufficient SW segregation has led to the contamination of plastic particles in both BSFL substrate and frass. Approximately 12 MaP, 9 MeP, and 334 MP particles/kg DW of substrate were identified at JRC, while 250 MP particles/kg DW were detected in frass. Conversely, 40 MaP, 35 MeP, and 734 MP particles/kg DW substrate and 484 MP particles/kg DW of frass were found at WCC. Transparent plastic films were the most dominant type at both locations. The abundance of plastics found in the BSFL substrate indicates that SW segregation at both centers was insufficient. Although plastic contamination does not significantly affect BSFL survival, growth, or development, MP particles were still detected in the BSFL frass. These findings highlight the importance of sufficient SW segregation to minimize plastic contamination in both the BSFL substrate and frass. This, in turn, enhancesg the efficiency and safety of the SW bioconversion process.

Acknowledgement

The first author acknowledges the Indonesian Endowment Fund for Education/Lembaga Pengelola Dana Pendidikan (LPDP), Ministry of Finance Indonesia for providing the financial support through a scholarship grant.

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