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# Biodegradation of Single Use Plastic Waste by Insect Larvae: A Comparative Study of Yellow Mealworms and Superworms

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**ABSTRACT**

The global plastic waste crisis, worsened by ineffective recycling and environmental pollution, necessitates exploration of alternative disposal methods. This paper examines the biodegradation capabilities of yellow mealworms, *Tenebrio molitor* and superworms, *Zophabas atratus*, focusing on their consumption of expanded polystyrene (EPS), low-density polyethylene (LDPE), and biodegradable plastics. Plastic waste, mainly composed of non-hydrolyzable plastics like polyethylene and polystyrene, presents challenges due to slow degradation. The research reveals the larvae's preference for EPS, highlighting the importance of species-specific considerations in plastic waste management. Preference for EPS is crucial because it is bulkier and more difficult to dispose compared to other types of plastics. Experimental setups monitored larval consumption, with weight measurements and frass production indicating preferences. Fourier-transform infrared spectroscopy confirms signs of biodegradation in the frass, demonstrating the transformative impact of larval digestion on plastic structures. Despite valuable insights, challenges like maintaining larval nutrition and understanding environmental factors' influence on degradation efficiency require further exploration. Utilizing insect larvae for plastic waste management holds promise for sustainable mitigation, but continued research is crucial for practical implementation.



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## 1. INTRODUCTION

### 1.1 Environmental Implications

Annual plastic waste production exceeding 359 -368 million tons has emerged as a significant global environmental concern, due to mass production, durability, and mismanagement from 2018 to 2019. (Peng et al.,2020, Lou et al.,2021) With only 9-18% of plastic waste being recycled, majority of plastic waste is disposed via incineration, landfilling, or release into the natural environment. Furthermore, improperly managed wastes cause substantial environmental pollution such as accumulation in soil, oceans, and freshwater ecosystems (Luo et al.,2021, Quan et al.,2023). Plastic waste not only affects marine life through issues like ingestion by fish and shrimp, but also poses possible indirect health risks to humans who in turn consumes these animals. This may lead to chronic exposure to plastic consumption that may pose a serious risk to health as plastics lack essential nutrients necessary for growth (Pham et al.,2023).

The reason for this accumulation is the extended life span of plastics. With an extended lifespan of hundreds of years, this brings harm to marine and terrestrial ecosystems and threatens both human and animal health (Pham et al.,2023). Another reason is the low degradation rates of plastics in natural environments, due to their macromolecular structure. Other inherent characteristics of plastic polymers, such as high molecular weight and high crystallinity, also hinders natural degradation. (Wang et al.,2022)

## 1.2 Different Types of Single-Use Plastics

Relentless accumulation of plastic waste is caused mostly by plastic types such as polyethylene (PE), polypropylene (PP), polystyrene (PS), polyvinyl chloride (PVC), polyethylene terephthalate (PET), and polyurethane (PUR). Common products like foam boxes, food containers, and plastic bags, persist in the environment for extended periods, particularly in developing countries. (Pham et al.,2023).

Non-hydrolyzable plastics, including PE and PS, which make up a substantial portion of petroleum-based plastic products, are more resistant to enzymatic depolymerization or degradation, adding to the challenges of plastic waste management. (Peng et al.,2020)

Plastic waste, particularly from polyethylene (PE) and polystyrene (PS), constitutes a significant environmental concern, being major contributors to common plastic waste (Yang et al.,2021). Polystyrene (PS) and polyethylene (PE), which account for a significant portion of plastic waste, exhibit extremely low biodegradation rates in natural environments, necessitating alternative methods for efficient degradation. (Lou et al.,2021)

With the increase in the demand for online shopping and the delivery of the products directly to residential houses, there has been an increase in the amount of plastics used for this industry, particularly Styrofoam, LDPE bubble wraps, and even biodegradable packaging materials. Styrofoam, a widely used form of polystyrene (PS), poses a significant environmental challenge due to its high production volume of nearly 2 million tons per year in Europe, leading to substantial PS waste generation. The lightweight nature of Styrofoam makes it prone to wind dispersion and water pollution, creating environmental issues along shores and waterways, emphasizing the need for effective PS waste management strategies. (Matyja et al., 2020). On the other hand, literature is lacking for the discussion of the increase of LDPE bubble wraps and biodegradable packaging, which necessitates the conduct of this study.

## 1.3 Challenges of Plastic Degradation

Effective waste management for PE and PS is hampered by the slow and limited traditional microbial degradation methods for PE and PS, often requiring weeks, months, or even years for noticeable effects (Yang et al., 2021). Despite extensive research since the early 1970s, the traditional scientific consensus was that rapid PS degradation would necessitate energy intensive photolytic of thermolytic techniques to cleave -C-C- bonds (Yang et al., 2018). However, recent discoveries of biodegradation, which involves biological mechanisms in the guts of insect larvae such as mealworms, has challenged this assumption (Yang et al., 2018). Biodegradation is considered an economically and environmentally friendly alternative for plastic disposal (Quan et al., 2023). Slow traditional methods of plastic biodegradation involve bacteria, fungi, and mixed microbial cultures, prompting research into more promising methods involving larvae from specific beetle and moth families, such as darkling beetles and pest moths (Peng et al., 2020). Insect larvae, including yellow mealworms, superworms, and greater wax worms have been identified as capable of degrading various types of plastics, (Wu et al., 2023).

The plastic-eating capacity of insects, particularly mealworms, has been observed since the 1950s, but early reports lacked investigation into the fate of ingested plastics; recent studies, however, demonstrate the potential for gut microbe-dependent cleavage of long-chain PS molecules in mealworms (Yang et al., 2018). Mealworms, specifically the larvae of *Tenebrio molitor*, have been demonstrated to efficiently degrade and mineralize polystyrene (PS), a commonly used petroleum-based plastic (Yang et al., 2020). This is because *T. molitor*’s digestive tract is highly differentiated and adapted to digesting various types of food (Przemieniecki et al., 2020). While mealworms have been extensively studied for their ability to biodegrade PS, the potential of superworms remains unexplored (Yang et al., 2020). Insect larvae, particularly superworms (*Zophobas atratus*), have demonstrated the ability to ingest and degrade plastic packaging materials, especially PS, providing a promising alternative for plastic waste management and emphasizing the need for understanding specific degradation pathways and impacts on superworms, including the potential effects of microplastics. (Quan et al., 2023) *Zophobas atratus* larvae have shown the capability to biodegrade PS and LDPE via a gut microbial-dependent mechanism, highlighting the potential of these insect larvae in plastic waste management and providing new insights into the adaptability of gut microbiota and internal enzymes for PE and PS biodegradation. (Peng et al., 2022)

### 1.4 Insect Larvae as Potential Solution

Invertebrates, especially insect larvae of beetles, including *Tenebrio molitor* and *Zophobas atratus*, voluntarily masticate, eat, and penetrate persistent plastics, accelerating the biodegradation process through synergistic contributions of gut microbiota and digestive enzymes (Peng et al., 2022). Plastic degrading enzymes are proven to be found in insects (He et al., 2024). It has been validated that enzymes and microbes in insects can degrade plastics through catabolism (Lee et al., 2023). Emphasis is placed on the gut microbial consortium to degrade synthetic plastic wastes (Ali et al., 2021).

Insect larvae, including species like superworms and yellow mealworms, have been discovered to ingest and degrade various types of plastics, including polystyrene (PS), polyethylene (PE), polypropylene (PP), polyurethane (PU), polylactic acid (PLA), and polyvinyl chloride (PVC), providing promising directions for biodegradation. (Wanget al., 2022)

Factors such as differences in plastic consumption rates and efficiencies, biodegradation capacities, functional group changes and depolymerization patterns, exist between yellow mealworms and superworms (Wang et al., 2022). This highlights the significance of understanding species-specific biodegradation preferences and mechanisms. (Wang et al., 2022)

While mealworms and their gut bacteria show promise for PS waste degradation, the effectiveness of this approach is influenced by various factors such as temperature, humidity, photoperiod, oxygen concentration, population density, parental age, food quality, and toxic substances, highlighting the complexity of implementing larval-based recycling technologies (Matyja et al.,2020). This highlights the need for further research regarding this matter.

In addition to this, the frass generated by the insect larvae may be used for various purposes. Studies show bioinsecticides can be derived from the frass of *Tenebrio molitor* (Urrutia et al., 2023). Organic biofertilizers can also be derived from frass derived from insect larvae, which will promote plant growth and enhancing the nutritional content in crops while minimizing wastes (Kee et al., 2023). Aside from frass, the insect biomass itself can be a source of food as it is composed of functional proteins with hepatoprotective, antithrombotic, antioxidant properties (da Costa Rocha et al., 2021). Considering the potential high value products being derived from frass and biomass, the use of insect larvae for biodegradation proves to be advantageous compared to the use of other methods such as microorganisms or enzymes.

Research on the consumption of different plastic types, including expanded polystyrene (EPS), low-density polyethylene (LDPE), and biodegradable plastics polypropylene (PP) were not conducted yet, hence, this study was conducted to address the plastic waste problem of the above types, through the utility of insects. In addition, the comparative capacity of mealworms and superworms are less discussed in literature. This study was conducted to compare the ability of mealworms and superworms to degrade EPS, LDPE, and biodegradable plastics for potential use in plastic biodegradation which can potentially contribute for plastic waste management.

## 2. METHODS

Three types of plastics were used for this experiment: expanded polystyrene (EPS), low-density polyethylene (LDPE), and biodegradable plastics. The plastics were chosen because these types of plastics are the most used in online shopping delivery packaging. EPS is used to cushion fragile items, LDPE is used to add an additional layer of protection, and the outer covering is usually made of biodegradable plastics as indicated. 4 conditions for each insect larvae were done: pure EPS, pure LDPE, pure biodegradable plastic, and mixed EPS, LDPE, and biodegradable plastic. One hundred six-week-old larvae of mealworms and superworms were introduced in each setup in three replicates. The total number of setups is 24. The weight of the plastic samples and larvae were obtained at the start of the experiment. The experiment was done for 21 days following the protocol from Zhu et al. (2021).

### 2.1. Sample Preparation

EPS, LDPE, and biodegradable plastics from online shopping delivery packaging were cut at a length and width of 5 cm x 5 cm. In addition, the EPS was cut into 2 cm thickness for the pure samples, while 1 cm was measured for the mixed samples. Each sample were initially placed at equal distances from each other in the microwavable polypropylene containers to ensure equal access by all larvae. For the pure setups, six pieces each of plastics were placed. For the mixed setups, 2 pieces each of the plastic types were placed.

### 2.2. Larvae Preparation

Larvae were starved 48 hours before the start of the experiment (Zhang et al., 2023, Zhong et al., 2022). One hundred larvae each of mealworms and superworms were placed inside the sealed microwavable polypropylene containers containing the plastic samples (Lou et al., 2021). To ensure air will be introduced inside the plastic containers, holes were punctured on the lids using a soldering iron. The containers were stored on iron shelves at room temperature. Temperature for the experiment was at room temperature of 25 degrees Celsius.

### 2.3. Plastic Consumption

The weight of plastic was measured to determine the collective consumption of plastic by each larvae type. The plastics was weighed by separating the plastic samples and weighed using Ohaus analytical balance (Ohaus Instruments, Shanghai, Co. Ltd). The weight of plastics until two decimal places were obtained. The data was collected at day 3, 6, 9, 13, 17 and 21.

### 2.4. Insect Growth

The growth of the larvae is monitored to determine the effect of the consumption of plastics to the growth of larvae. The plastic samples are first separated from the containers. After weighing of plastic samples, the larvae will be separated from the frass by using mesh screens. Larvae are then weighed using an analytical balance. The weight of larvae until two decimal places were obtained.

### 2.5. Average Consumption

Average consumption is defined as the amount of plastic being consumed by the larvae for each sampling period. This is done to determine the relationship between the amount of plastic consumed by the larvae and the amount of frass being produced. The average consumption is computed by subtracting the current weight of the plastic during the sampling period with the previous weight of the previous sampling period, as shown in the following equation:

$$average\ consumption = previous\ plastic\ weight - current\ plastic\ weight \quad (1)$$

### 2.6. Frass Weight Determination and Fourier Transform Infrared Spectroscopy

Frasses are the solid particles that are produced by the plastic consumption of larvae. The frass is determined by subtracting the weight of the polypropylene container, plastic, and larvae from the total weight of each setup. To do this, the total weight of the setup is first weighed. The plastics and larvae are then weighed separately. Larvae was separated from the rest of the setup with the use of mesh screens. The polypropylene container was measured at the start of the experiment. The Shimadzu FTIR was used in the experiment to examine the frass for signs of biodegradation (Yang et al.,2011 and Zhong et al., 2023). The settings used for FTIR include the following: Intensity mode: %Transmittance, apodization: Happ-Genzel, No. of scans: 45, Resolution, 4 $cm^{-1}$.

## 3. RESULTS AND DISCUSSION

### 3.1 The Average Consumption of Plastics by Yellow Mealworm and Superworm

The biodegradation of expanded polystyrene (EPS) by superworm and mealworm indicates their use in waste management. Both mealworm and superworm consume EPS as their sole food source as indicated by a decreasing trend in the average EPS weight. This decreasing trend reveals positive consumption of EPS by the insect larvae (Fig. 1). The amount of EPS consumed by superworms are clearly greater than yellow mealworm with the EPS weight of 2.70 grams in day 1 to 0.80 grams in day 21, showing an average consumption of 1.90 grams EPS by superworm. The yellow mealworms consumed only a very minute amount of 0.10 grams for the 21-day duration (from 2.70 to 2.60 grams). While yellow mealworms have been extensively studied for their ability to biodegrade plastics, our results suggests that superworms are more effective in biodegrading EPS than yellow mealworm. In a study by Quan et al. (2023), they revealed that superworms can ingest and degrade plastic packaging materials, providing a promising alternative for more efficient plastic waste management. Superworm larvae have shown the capability to biodegrade plastics via a gut microbial-dependent mechanism (Peng et al., 2022).

The same trend was observed in the average consumption of biodegradable plastics by superworms where a decreasing weight of biodegradable plastics was observed, indicating positive consumption in the set-up (Fig. 2). On the other hand, there was no reduction in the weight of the biodegradable plastics fed to mealworm, indicating no preference in this type of plastic. Between superworms and mealworms, superworms were proven to consume biodegradable plastics than mealworm.

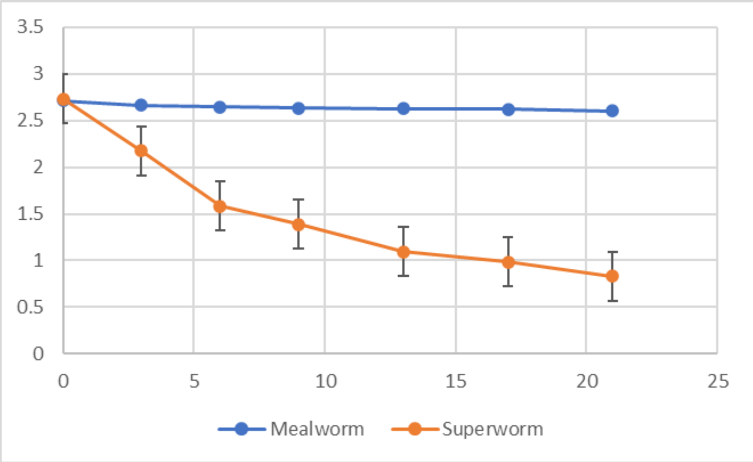


**Fig. 1.** Average weight of expanded polystyrene (EPS) revealing consumption of EPS by mealworms and superworms over 21 days

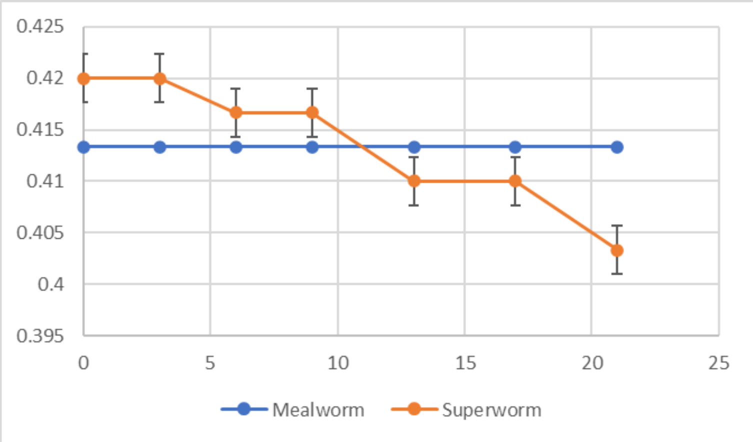


**Fig. 2.** Average weight of the biodegradable plastics indicating consumption of superworm and non-consumption of mealworm over 21 days

While EPS and biodegradable plastics show a decreasing average weight, which indicated positive consumption, the set-up on low-density polyethylene (LDPE), shows increasing trend for superworms to fluctuating LDPE weight in mealworms (Fig. 3). A possible explanation for this is the frass that is excreted by the insect larvae have adhered to the LDPE, hence the increased in weight when measured. While the frass was removed to the best of our ability upon measurement, the adherence of the remaining frass to LDPE was hard to remove may be due to the static electricity that is produced by the rubbing of superworms along the surface of the LDPE. Frass, being small in diameter and light in weight, tend to be attracted more to the LDPE charged with static electricity. Further studies are recommended to verify this observation.

Contrastingly, Pham et. al., (2023) described the consumption of polystyrene (PS) by mealworm to be steadily increasing. Yang et al. (2020) computes the average consumption of superworms as 0.58 mg/d per worm, which is around four times that of the average consumption of mealworms at 0.12 mg/d per worm.

The set-up for mixed feeding were conducted to study the plastic preference of the insect larvae as feed. Considering mixed feed, EPS is the most preferred plastic as indicated by the full consumption of this plastic feed in the set up (Fig. 4). This trend is the same as when EPS is the sole feed for the larvae. When feed with mixed plastics, there was almost no preference for biodegradable plastics and LDPE (Figs. 5 and Fig. 6) as shown by the almost constant trend line with a very minute decrease in biodegradable plastics by superworms, indicating very minor consumption. In Fig. 6, the trend is the same as when LDPE is the sole feed showing slight increase in the weight of the LDPE, though the change is significantly lower compared too when LDPE is the sole feed. While there is a slight decrease in LDPE weight for superworm indicating positive consumption at the start of the experiment, there was an increase in weight towards the end of the feeding period. This can still indicate positive feeding consumption as the increase in LDPE weight might be due to frass adhering the LDPE plastic.

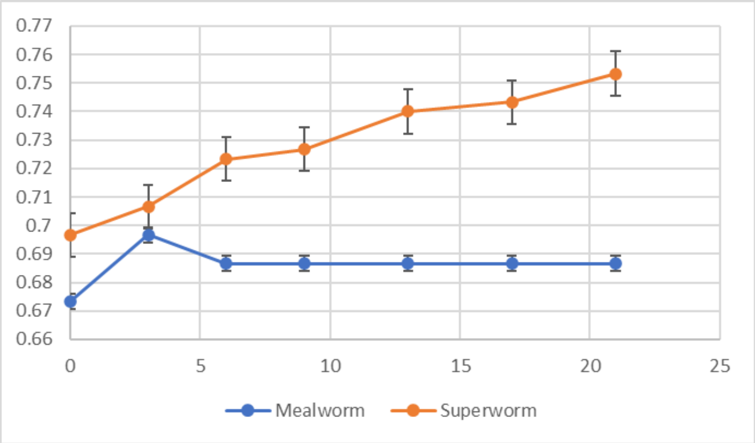


**Fig. 3.** Average LDPE weight of mealworms and superworms from sole LDPE setup

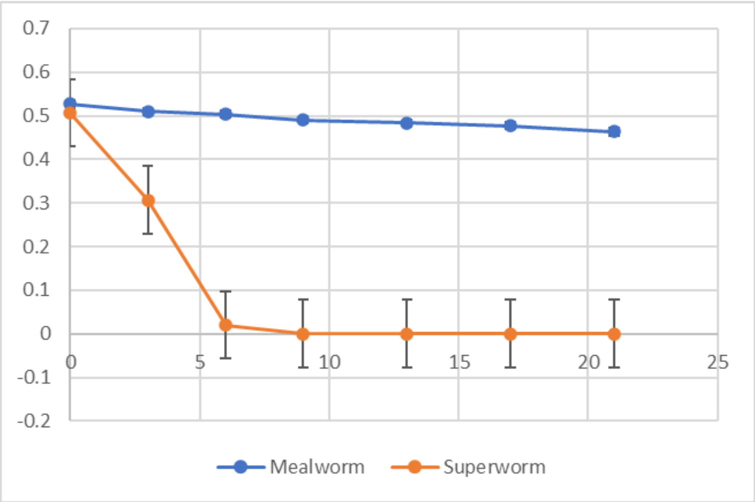


**Fig. 4.** Average EPS weight indicating consumption of mealworms and superworms from mixed plastics setup over 21 days

In mixed feeding set-up, the insects have preference to consume EPS more than LDPE and biodegradable plastics. The less preference to LDPE can be explained by the structure of LDPE. As discussed by Pham (2023), LDPE has more carbon-carbon bonds (i.e. more branching), resulting to a more bio resistant property. Zhong et al. (2022) supports this observation, stating that LDPE is composed of linear chains with branching. A study by Luo et al., (2021) supports this, stating PS consumption has the highest consumption while PE consumption has the lowest consumption.

### 3.2 The Effect of Plastics as Insect Feed on Insect Weight and Frass Production

Fig. 7 shows the average weight of the larvae when fed with plastics. The weight of the larvae has a downward trajectory, regardless of the type of plastics being consumed. This implies that the consumption of plastics has no contribution to the nutrition of the larvae. Particularly, the superworms, despite being able to consume a significant amount of EPS compared to other types of plastics, the trend for the decrease in weight is the same.

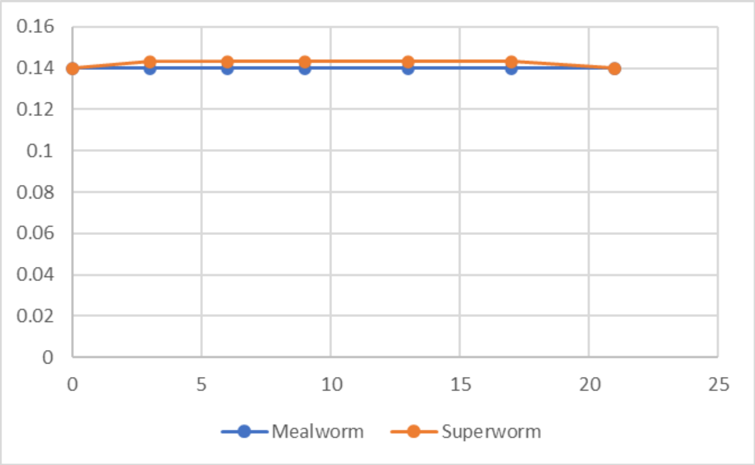


**Fig. 5**. Average biodegradable plastic packaging consumption of mealworms and superworms from mixed plastics setup over 21 days

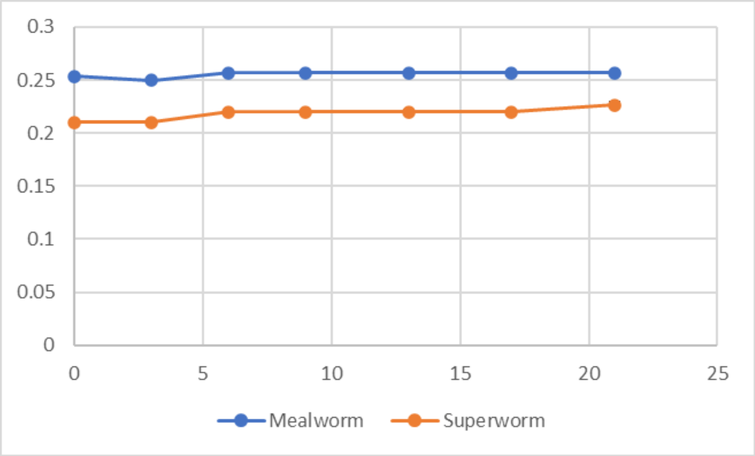


**Fig. 6.** Average LDPE consumption of mealworms and superworms from mixed plastics setup over 21 days

This study conforms with the result of Wang et al. (2022), where a there is a net decrease of weight of superworms with plastics-only diet. Pham et al. (2023) observed that for mealworms, an initial loss of mass was observed for the first 10 days, with gains after 20 days, followed by drastic decrease after 30 days. Furthermore, in a study by Billen et al. (2020), they reported that the sole diet of LDPE is deemed insufficient for growth of the insects.

The decrease in weight suggests that while the worms can feed on the plastics, pure plastic diet cannot sustain mealworms and superworms as also reported in a study by Lou et al., (2021). Peng et al., (2022), who stated that plastics as the sole carbon source lacks sufficient nutrients to support growth and the metamorphosis of the larvae to the next phase of the life cycle, which is the pupa. Decrease in weight implies that insect larvae cannot be used for the degradation of plastics in the long term. It is proposed that fresh worms are to be used depending on when the rate of consumption starts to decrease, as indicated by the slope of the graph.

In Fig. 8, a general decreasing trend was shown for the average frass production. Presence of frass indicates that the larvae have consumed plastics, and the remains were secreted by the larvae. However, the decrease of production of frass indicates the decrease in consumption of the plastic by the insect larvae. This supports the previous finding on average consumption that also showed a decreasing trend. The type of plastic may influence the amount of frass produced, as can be seen by the higher amount of frass produced by superworms that ingested pure EPS and the mixed feed, which also contained EPS. The trend for frass weight of superworms in mixed feed had a steep decline after day 6, which is around the same date when the EPS in the mixed feed was fully consumed.

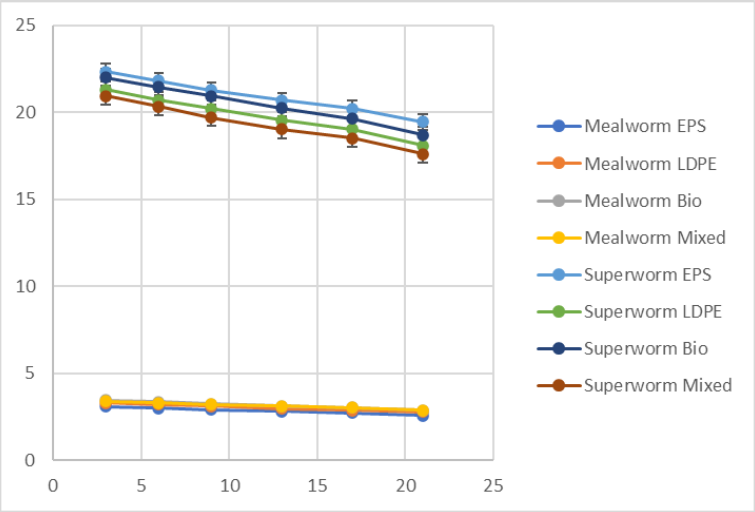


**Fig. 7.** Average insect weight when fed with different plastic types measured over 21 days

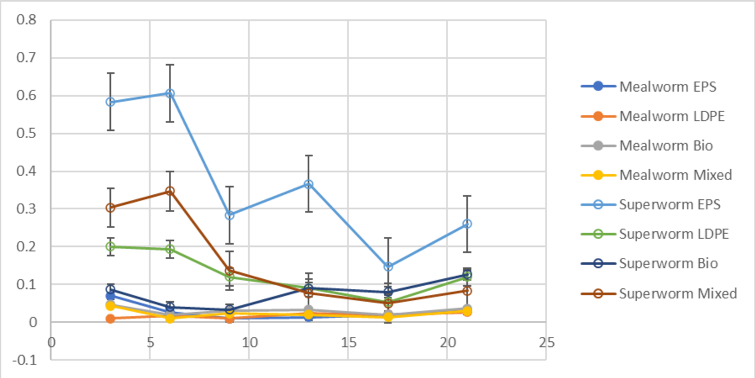


**Fig. 8.** Average frass production of insects when fed with plastics over 21 days

The average consumption vs average frass is shown in the Fig. 9. The setup involving superworms consuming purely EPS gained the most average frass per average consumption. The setup involving superworms consuming mixed plastics also has more average frass per average consumption. Frass is also produced despite no consumption of plastic. This can be attributed to worms that are still secreting wastes from the feed prior to the starvation process. Particles from molted skin covering may also contribute to the amount of frass measured.

### 3.3 The Visual Comparison of the Different Types of Plastics Before and After Feeding to the Insects

Fig. 10 and Fig. 11 shows the consumption of EPS by superworms and mealworms, respectively. In Fig. 10, the consumption by superworm is indicated by the large holes in the samples. In a study by Wang et al. (2022), they noted large holes in PS foams fed to superworms. Yang et al., (2020) observed holes in the samples after 1 hr. Fig. 11 shows the mealworms consuming the EPS, as indicated by the small holes in the EPS samples. As can be seen, not all the samples were consumed by the mealworms as evidenced by the lack of holes by some of the EPS samples. The same observation was noted by Wang et al. (2022), who observed hollows and pits in their samples.

When it comes to the consumption of biodegradable plastic by the superworms, tears were found at the edges of the plastic, which indicates the attempts of the superworms to consume the plastic (Fig. 12). The tears are more prevalent compared to that of the mealworms, which are almost non-existent. In mealworms, there was no sign of attempt to consume the biodegradable plastics and there were no signs of tear at the edges of the plastic, which is the sign of consumption by the larvae.

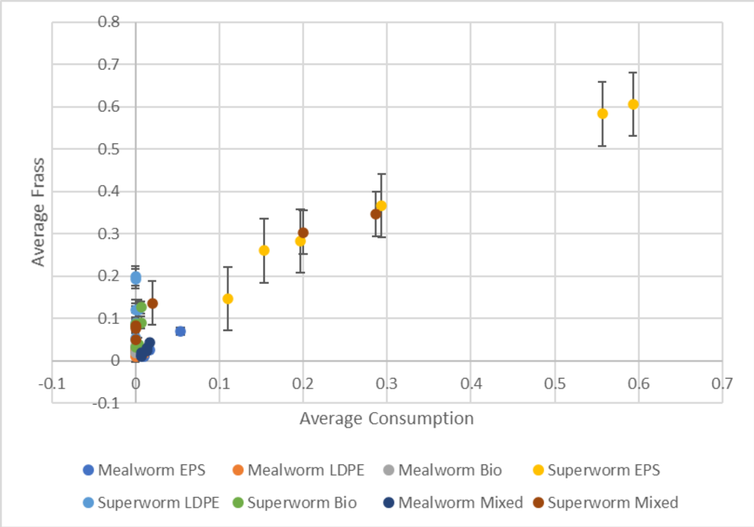


**Fig. 9.** Average consumption versus average frass production of mealworm and superworm fed with plastics

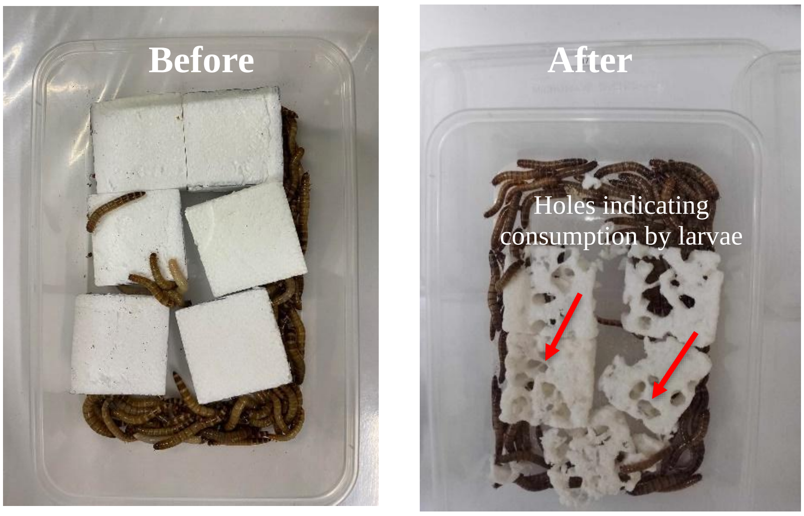


**Fig. 10.** Consumption of pure EPS by superworms after 21 days


**Fig. 11**. Consumption of pure EPS by mealworms after 21 days


**Fig.12.** Consumption of pure biodegradable plastic packaging by superworms

Consumption of LDPE by the superworms are seen in Fig. 13. There were slight tears in the sides of the LDPE, indicating the superworms' attempt to consume the plastic. There are indications of frass in portions of the plastic that are difficult to remove. This confirms the previous observation that static electricity that is generated by the superworms rubbing itself into the plastic sheets as the cause, leading to the increasing trend of weight recorded. The trend of increase in weight in superworms being greater than that of mealworms can be attributed to the fact that superworms generate more frass. Being larger in size, the superworms worms may also generate more static electricity as compared to mealworms.

For mealworms, consumption of LDPE is very minor as seen by minor signs of tear in the edges of the plastic (Fig. 14). This is an indication of the mealworms attempt to consume the sample, particularly for plastic sheets (Pham et al., 2023). There are indications of frass in portions of the plastic that are difficult to remove. This confirms the previous observation that static electricity that is generated by the mealworms rubbing itself into the plastic sheets as the cause, leading to the increasing trend of weight recorded.


**Fig. 13.** Consumption of pure LDPE by superworms

When fed with mixed plastics, there is a clear preference of superworm and mealworm to EPS when given a choice. Fig. 15 shows that EPS was fully consumed by superworms at the end of the 21-day observation period. Mealworms also show preference to EPS as indicated by holes in EPS from mixed-fed set-up (Fig. 16). There were little to no signs of consumption in the LDPE and biodegradable plastics. This further supports the preference of mealworms towards EPS as compared to LDPE and biodegradable plastics. Between superworm and mealworm, superworms consume more plastics than mealworm.

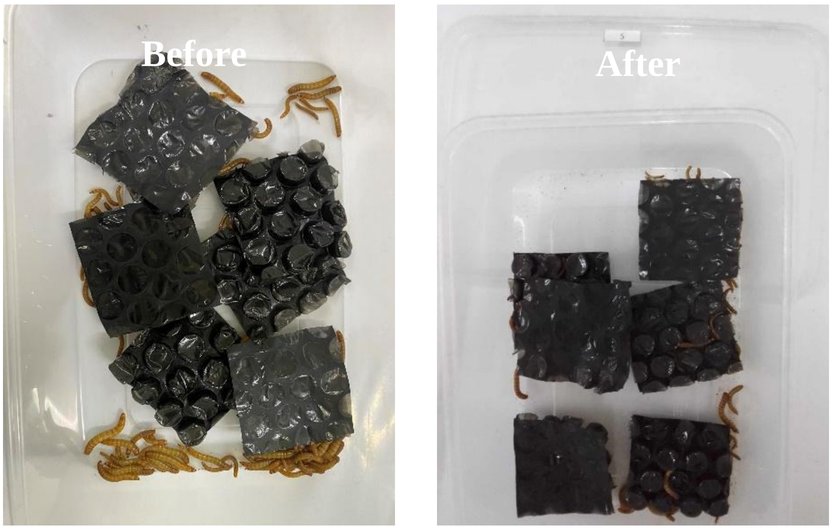


**Fig. 14.** Consumption of pure LDPE by mealworms


**Fig. 15.** Consumption of mixed plastics by superworms

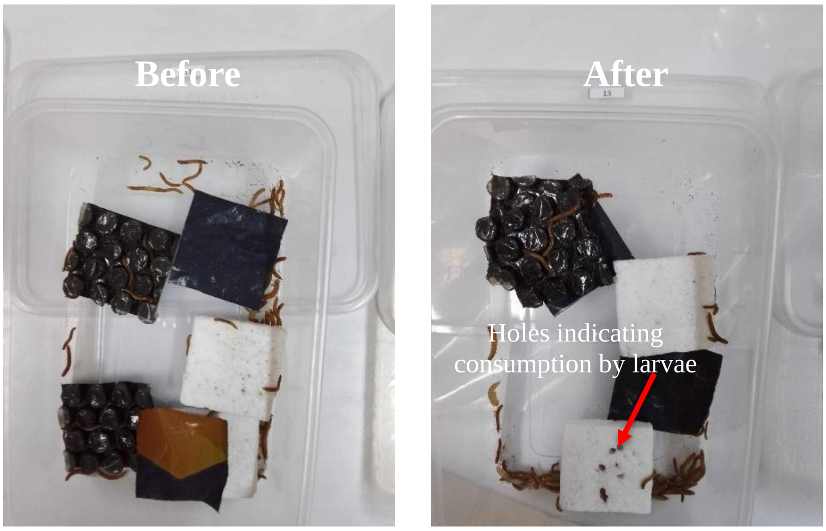


**Fig. 16.** Consumption of mixed plastics by mealworms

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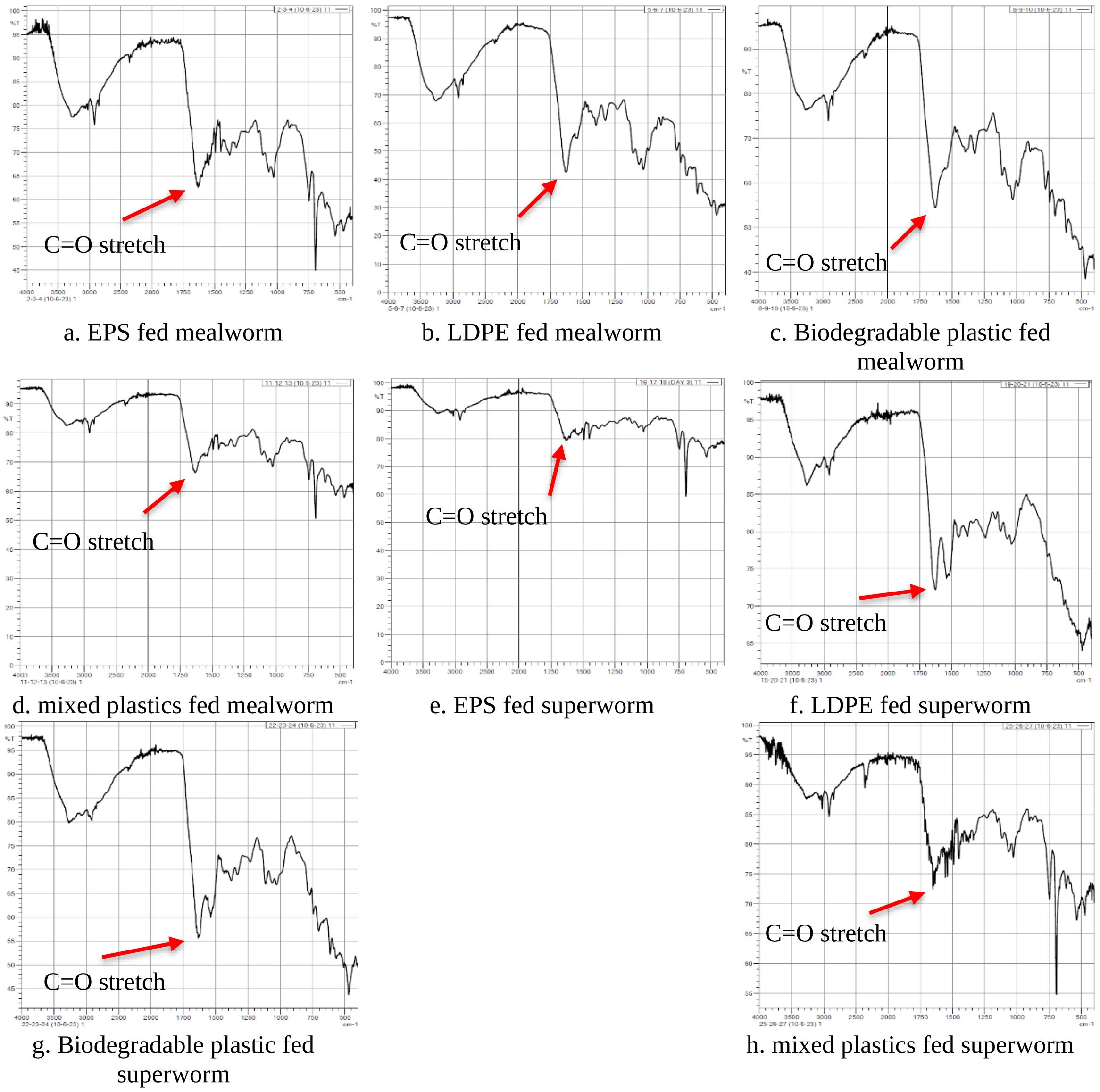
### 3.4 Biodegradation Measurements Using Fourier Transform Infrared Spectroscopy (FTIR)

Fig. 17 shows the functional groups found on the frass from the different setups: a) EPS fed mealworm, b) LDPE fed mealworm, c) Biodegradable plastic fed mealworm, d) mixed plastics fed mealworm, e) EPS fed superworm, f) LDPE fed superworm, g) Biodegradable plastic fed superworm, and h) mixed plastics fed superworm. Peaks in the FTIR were commonly observed in the 3300-2500 $cm^{-1}$ frequency range for all samples, which implies the presence of carboxylic acid groups. All samples also exhibited peaks in the 1650 $cm^{-1}$ range, indicating the presence of C=O stretch in the frass samples. The previous observations indicate the addition of oxygen functional groups into the polymer chains, a preliminary and key step in plastic degradation, as well as oxidation and depolymerization of PS in the guts of mealworms and superworms (Wang et al., 2020). The indications of plastic degradation for mealworms are proven by the presence of the peaks. It can be inferred that the indications of biodegradation despite the lack of trend in the consumption of plastics is proof that the larvae consumed the plastics, but only at minimal amounts.




a. EPS fed mealworm    b. LDPE fed mealworm    c. Biodegradable plastic fed mealworm

d. mixed plastics fed mealworm    e. EPS fed superworm    f. LDPE fed superworm

g. Biodegradable plastic fed superworm    h. mixed plastics fed superworm

**Fig. 17**. Fourier Transform Infrared Spectroscopy (FTIR) results

It can be noted that there are peaks found in the 700 $cm^{-1}$ range, particularly in the samples containing EPS. This indicates groups containing a C=C bond. Since polystyrene consists of a benzene ring, this is an indication of the cleavage of the ring, another evidence of biodegradation. Cleavage of the ring implies that the circular structures are converted into linear structures, which makes it easier for further biodegradation to occur. Additionally, the broad peaks of 2500-3500 $cm^{-1}$ indicates the change of property from hydrophobic to hydrophilic (Yang et al.,2018, Yang et al., 2021). While this is not a sign of biodegradability, it is an indication that the frass is more soluble to water, and thus more environment friendly.

Based on the findings of the study, yellow mealworms and superworms are shown to have potential to biodegrade EPS the most. The insect larvae have shown preference in ingesting EPS compared to LDPE and biodegradable plastics. Further studies on the rate of degradation will bring more value to this finding as it will determine if the process can be upscaled to commercial scale. Study for the long-term effect of plastic ingestion is also a promising direction. Diversifying to other types of single use plastics such as polyurethane and PET will also be beneficial to the environment. A clear difference between the amount of EPS ingested by superworms compared to yellow mealworms is also observed. More studies that would investigate if increasing the quantity of yellow mealworms will lead to a better performance may be done and compared to the amount ingested by superworms. This will further give insight into which insect larvae is better suited to be used for biodegradation. Additional studies on the biochemical mechanisms that enable the insect larvae to biodegrade the plastics will also aid in the deeper understanding of this process. As previously discussed, enzymes and gut microbiota is pointed to be responsible for the process. Isolating the enzyme or microorganism most responsible for biodegradation will also enable the development of other processes and operations such as bioreactors.

## 4. CONCLUSION

Biodegradation of plastivs like expanded polystyrene (EPS) by insect larvae such as yellow mealworm and superworms are proven to be viable, thus providing an environmentally friendly alternative to other processes of recycling plastics. This is proven by the following observations:

1. There is a notable decrease of plastics, especially for EPS.
2. Frass, which is an excrement by the larvae, are produced even towards the end of the observation period. This shows the larvae are still able to ingest the plastic and excrete the product.
3. Signs of biodegradation in the frass via FTIR is an indication that the ingestion of plastic by larvae underwent biodegradation within the gut of the larvae.

Given the same number of yellow mealworm and superworms, there is a clear indication of the preference for EPS compared to LDPE and Biodegradable plastics. The superworm particularly ingested the most quantity, which is from 2.7 grams to below 0.80 gram for the pure feed. The preference for EPS over the other plastics can be further proven by the mixed feed samples, wherein EPS was consumed more, or in the case of superworms, were totally consumed. Preference for EPS is significant because it is bulkier and more difficult to dispose compared to LDPE and biodegradable plastics. However, the study highlights the inadequacy of a pure plastic diet as evidenced by the decreasing larvae weight trend.

FTIR results peaks of functional groups such as carboxylic acids and C=O stretch, which showed indications of the initial biodegradation of plastics and change in properties. These are significant since the complex structures such as rings being converted to C=O implies a simpler structure because of biodegradation, while property change to hydrophilic indicates that the sample will be more soluble to water and thus more environment friendly.

Aside from the current single-use plastics used in the study, other types of single-use plastics such as PET and polyurethane may also be subjected to the same plastic-eating insect larvae. Further studies such as the biochemical mechanisms and the long-term effect of plastics may be further explored to widen the knowledge in this field.

### Author Contribution

GAMG and MSG conceptualized the study. GAMG and MSG conducted experiments for insect larvae. GAMG wrote the original draft of the paper. MSG contributed to the manuscript preparation and finalization. All authors have read and agreed to the published version of the manuscript.

### Conflict of Interest

The authors declare that they have no competing interests.

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