Bioremediation of Plastics Through the Use of Mealworms

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Abstract:

Plastic waste poses a serious problem to the environment, as well as human and animal health. Microplastics can damage tissue and release toxic chemicals, and plastics in the ocean can kill animals, who accidentally consume or become entangled in plastic waste (Andrady, 2011; Vethaak & Leslie, n.d.). Of the 6.3 billion tons of plastic produced since 1950, only 9% has been recycled and 12% has been incinerated, with the remainder ending up in landfills or the ocean (Oa & Oa, n.d.). Given that plastic production is expected to quadruple by 2050, an effective and environmentally conscious way to handle our plastic waste is needed (Oa & Oa, n.d.). Bioremediation is a promising solution that involves using organisms, such as bacteria and fungi, to degrade plastic. While bacterial bioremediation typically involves special bioreactors, mealworms are able to degrade polystyrene plastic at room temperature due to their microbiome. However, mealworms cannot survive on plastic alone and require an additional food source, such as corn meal, oat bran, or wheat bran, which can alter their microbiome and the hydrolytic enzymes they produce (Przemieniecki et al., 2020). Our study intends to investigate how corn meal, oat bran, and wheat bran affect the mealworm's microbiome and ability to degrade plastic. Determining the rate of degradation for each food source and the cost will enable small-scale applications of mealworm bioremediation systems, such as in households, neighborhoods, and businesses. Having a better understanding of bioremediation applications will also allow for further research determining the public's willingness to use bioremediation technology.

Section I:

In our world, plastics are becoming a growing threat to ourselves and the environment we live in. As useful as they are, plastic's resistance to break down has led to its buildup in not only our landfills, but our oceans, our forests, lakes and rivers, and nearly every other ecosystem on the planet. As such, a new way of plastic disposal is necessary to manage the growing production of plastics in our world. But unlike conventional methods of plastic disposal, our method does not produce very harmful waste that requires expensive removal. Our method is to use a specific type of mealworms to dispose of plastic waste passively. **By using mealworms that naturally metabolize plastic, we will be able to dispose of plastic in an easy and effective way without pumping harmful chemicals into the atmosphere. With the resources to conduct this research, we will take steps closer to implementing small-scale plastic disposal stations in every household on the planet, making plastic waste an issue of the past.**

With the grant to conduct this research, we will tackle the following research questions:

Can mealworms effectively digest plastic waste?	How harmful are mealworms and their waste products to humans?
At what scale should we implement these plastic disposal stations? Big or small?	How expensive would this be to safely implement?
How will we contain and store mealworms? Will we need to care for them or just feed them plastic?	Which plastics are best for mealworms to degrade? (PP, PET)

There has been research conducted in this area already which will assist us in tackling these questions. But with the resources to conduct our own research, we will be able to tackle these questions first hand and effectively determine if mealworms are an effective solution to plastic degradation.

Section II:

The mass production of plastic began around 60 years ago, as plastic is a cheap and durable material that can serve many different purposes. Over the past few decades, the issue of plastic waste has started to pose a more grave serious problem to our planet and its habitants. Every year, over 300 millions tons of plastic are produced and consumed globally, and only 9% of this total gets recycled. The rest enters the environment, where it will subsequently cause harm to wildlife and disrupt ecosystems.

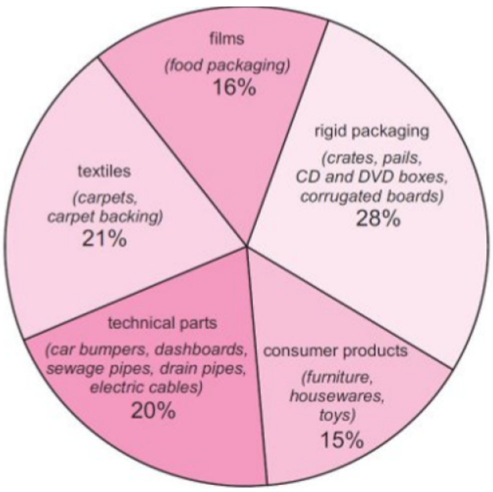
Plastic waste disproportionately pollutes the marine ecosystems that exist in our oceans. For example, plastics often break into smaller pieces known as microplastics. These microplastics can affect marine organisms by blocking their gastrointestinal tracts. Some organisms confuse plastics for food, and because of this, they don't get sufficient nutrients and subsequently starve to death. Microplastics also release toxic compounds like polychlorinated biphenyls (PCBs) in high concentrations. Although plastics can break down into smaller pieces, they still never fully degrade. Because of its highly durable nature, this plastic will remain in the environment undegraded for up to 1,000 years, harming wildlife and the environment, and also the humans that depend on it.


This is a image of the Pacific garbage patch, much of this waste is plastic.

Currently, the only widely-used alternative to landfills, which harm the environment and marine animals, is incineration. Currently, 12% of plastic is incinerated (Oa & Oa, n.d.). Incinerating plastic can release pollutants, carbon dioxide, and toxic chemicals that can have serious effects on human health, including: lesions, eye irritation, organ damage, birth defects, cancer, chronic bronchitis, and more (Ágnes & Rajmund, 2016; Oa & Oa, n.d.). To illustrate, incineration of polyvinyl chloride is illegal or restricted in some countries, as it produces many toxic chemicals, including phosgene, which was used as a chemical weapon in World War II (Ágnes & Rajmund, 2016). Clearly, a solution is needed, and even if biodegradable and recycled plastics become more widespread, there is ultimately a large amount of plastic waste in the environment that must be handled.

One of the most popular and durable plastic materials is polypropylene, used for packaging. Propylene plastic is also found in other everyday items, such as textiles that make up clothes, carpets, mats, and rugs, as well as in furniture, toys, and luggage. Every year, approximately 5 billion pounds of propylene are produced in the United States alone. Along with being one of the most popular plastics, it is unfortunately also one of the least recycled plastics. Less than 1% of all propylene ever produced is recycled. Most of the remaining polypropylene ends up in the landfills, where it will negatively impact the environment in many different ways.

Polypropylene also has damaging effects on humans. Although it is significantly safer than many



This pie chart shows the many uses of polypropylene


These widely used food containers can be made from polypropylene

other plastics currently being produced, it certainly has risks associated with it. In 2019, researchers found that polypropylene disrupted androgen hormones and caused a stress response in human cells. Although more research needs to be done to investigate the toxicity of polypropylene plastic, it is important to remain cautious of the plastics we produce. Therefore, it is imperative to find a solution to degrade polypropylene to mitigate its destructive effects on the environment.

One potential solution to the plastic waste problem is bioremediation, which involves using certain organisms to degrade plastic. Bacteria has been investigated as a potential solution, but many types of bacteria require a bioreactor. *Ideonella sakaiensis* has been identified as a species that can subsist solely on polyethylene terephthalate, but it is unable to grow on and degrade highly crystalline plastic, which limits its potential usage (Wallace et al., 2020).

Given the limitations of bacteria and other microorganisms, larger species are promising. For example, the microbiome of earthworms has been found to degrade


Image of a mealworm (*Tenebrio molitor*)

low-density polyethylene (Huerta Lwanga et al., 2018). Mealworms (*Tenebrio molitor*) are able to degrade polypropylene by chewing plastic, which is then broken down in their gut by their microbiome (Billen et al., 2020). Mealworms can survive at room temperature, which is advantageous because they do not require an additional heat source.

However, mealworms require an additional food source as they cannot survive on plastic alone (Billen et al., 2020). It is currently unknown how the food source affects the mealworm's ability to degrade plastic, as most short-term studies do not provide an additional food source, instead allowing some worms to starve (Billen et al., 2020). Research has shown that a mealworm's diet affects its microbiome in various ways and alters the hydrolytic enzymes produced, which may alter its plastic-degrading efficiency (Przemieniecki et al., 2020). Given that the microbiome plays an important role in plastic degradation, understanding which food source is best for plastic degradation would help make this process more efficient.

No studies have investigated potential applications of mealworm bioremediation, and limited research has been done into the cost of bioremediation using mealworms. One study identified the cost of mealworm bioremediation as approximately $360 USD per ton of plastic waste (Billen et al., 2020). Although the study determined that this price would be economically unsustainable from a business perspective, households may be willing to pay a certain amount to have their plastic waste handled in an environmentally conscious way, given that plastic makes up 10% of all household waste (Oa & Oa, n.d.). It's also possible that businesses or towns may be interested in a small-scale bioremediation system, rather than an industrial application. However, it has not yet been determined how much it would cost to initially set up and to maintain mealworm bioremediation technology on a small scale. Calculating the initial and yearly costs is an essential part of evaluating and implementing this solution.

The general public's willingness to use bioremediation technology is also unknown. It's likely that people would be concerned about microbial bioremediation, so mealworms may be a more accessible way to introduce people to the concept of bioremediation. Filling this gap in the knowledge is essential for ensuring that mealworms are a feasible and realistic way to address our planet's plastic problem.

Section III:

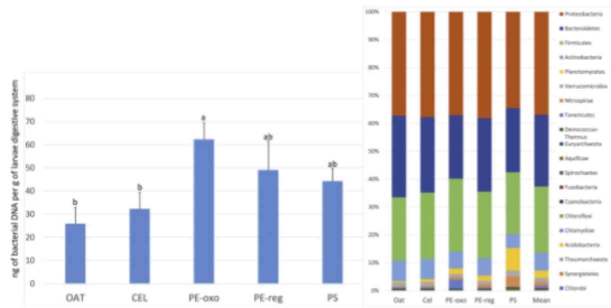
In this project we aim to compare different substrates and how they influence the mealworms microbiome and effect the rate of degradation of polypropylene (PP) plastic. In this set of experiments we are testing the different feeds for meal worms which include oat, wheat, and corn substrates. We will analyze how much the mealworms degrade while also looking at contents in their microbiomes.

The mealworms can be bought from a pet store or online. A pack of 50 mealworms can be bought for about 10 dollars online. For this project 100 worms per bin would be sufficient. Regular store bought bins would be used to house the worms. The bins would have to have lids with holes poked through them. Then the different feeds will be added to each bin. The feeds will be at a thickness about 2 inches from the bottom of the bins and each bin will have the same amount of feed added (*How to Care for Mealworms*, n.d.).

What mealworm bins would look like. They would be stored with cover on the top and polypropylene plastic in the bin.

One bin will have oat feed, another bin will have cornmeal feed, and one bin will have wheat substrate, the last bin will be one with just plastic as a control group. In total there will be four bins each with 100 worms. Along with the feed added we will add the same weight and size pieces of polypropylene plastic. We will mix the plastic around the bin. Once a week a few carrot slices will be added to the bins to bring moisture into the habitat. After two days, the carrot slice will be removed to prevent substrate spoilage. Every week the weight of the plastic pieces will be measured and qualitatively observed and 5 randomly selected mealworms will be removed and the microbiome analyzed. The microbiome can be analyzed through sequencing the V3-V4 region of the 16S rRNA genes found in the microbiome and analyzing the genes though the MetaGenome Rapid Annotation Subsystems Technology (MG-RAST). This analyzing of the genome will follow similar steps as the study *Changes in the gut microbiome and enzymatic profile of Tenebrio molitor larvae biodegrading cellulose, polyethylene and polystyrene waste* (Przemieniecki et al., 2020). This is because they also researched the changes in a mealworms microbiome and used these sequencing techniques. At the end of the study period which would be about 2 months the final samples will be taken and the rest of the worms microbiomes analyzed. For longer studies and application there would need to be more bins and a procedure to separate the matured organism from the worms.

There are 2 essential pieces of data that our experiment will generate: the first data piece is the amount of the plastic that these worms can degrade with the different substrates, which will be measured by the weekly weigh and qualitative observations. If we are successful then hopefully one bin will have less plastic at the end of the experiment than the others which would be seen both quantitatively and qualitatively. We can further use statistical analysis such as IBM SPSS Statistics (version 25) to help look at the consumption of polypropylene and microbiome sequencing (Yang et al., 2021). The results from sequencing the microbiome are also very important data that we are collecting. This connects the microbiome to how efficiently the plastic can be broken down. It can allow further research about what types of bacteria in the microbiome contribute to the degradation of this plastic.



This is what the microbiome analysis data would look like.
These images are from the study *Changes in the gut microbiome and enzymatic profile of Tenebrio molitor larvae biodegrading cellulose, polyethylene and polystyrene waste*

If this experiment is successful and efficient in degrading plastics we could look into a cost benefit analysis into if the model of using mealworms will be beneficial to use practically on a smaller scale such as a neighborhood bin where polypropylene scraps can be thrown into and degraded. From the paper *Biodegradation of polypropylene by yellow mealworms (Tenebrio molitor) and superworms (Zophobas atratus) via gut-microbe-dependent depolymerization* we estimate that the 100 mealworms per bin will degrade about 20mg of plastic per week (Yang et al., 2021). With the data we collect from this experiment we can see if people are willing to be involved with this type of small scale degradation of plastics. Another step that could be taken is rather than a small scale application a larger application of mealworms being used to degrade this type of plastic for a business or institution.

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