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Microbial Degradation of PET Plastic Sustainably Yielding Commercially Viable Products

Atharv Kapoor[1], Cheshte Varshney[2]

[1] Department of Biological Sciences, Sri Venkateswara College, University of Delhi

[2] Department of Biological Sciences, Sri Venkateswara College, University of Delhi

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Abstract

Plastics are extensively used due to their versatility, durability, and low cost. PET stands for Polyethylene terephthalate. PET plastic is widely used all over the world and has many applications ranging from water bottles to fabrics like polyester and many things in between. But its unrestrained use in every field is resulting in heaps and piles of non-biodegradable materials causing damage to the environment and causing pollution. The idea being proposed is to degrade the PET plastic biologically using different bacteria. The bacteria used in this process are Ideonella sakaiensis, Acetobacterium woodii, Pelotomaculum and Methanospirillum hungatei. PET plastic is degraded, yielding Terephthalic Acid (TPA) and Ethylene Glycol (EG) by the action of the bacterium I. sakaiensis. Degradation of EG by A. woodii results in the formation of acetate and ethanol. TPA is degraded by the action of the coculture of Pelotomaculum and M. hungatei thereby yielding methane and acetate. All these products formed have significant commercial uses in various industries. The complete process that is to be carried out can help in achieving sustainability by fulfilling various Sustainable Development Goals set by the United Nations.

Keywords: Polyethylene terephthalate, Microbial degradation, Sustainable development

INTRODUCTION

Plastic- a synthetic material has become an inseparable part of everyone's life. This all has happened because of the need of a material that is versatile and has a wide range of properties and can be used for almost every purpose.

Various properties of plastic like their durability, versatility, plasticity and also their lightweight makes plastic to be a material that is extensively used. The low manufacturing cost of plastic makes them a readily used material. Hence this makes plastic a boon for mankind. But plastic is also a bane for the nature including the animals and the mankind. It can cause severe repercussions if not removed from the cycle of use and disuse. Also, the amount of plastic in circulation cannot be completely removed but it can be reduced and that too can happen only to a certain minimum level. This is so because the rate at which plastic is being manufactured is much more than it being recycled. Most of the plastic waste generated is mostly dumped or incinerated and only a small percentage of it is recycled.

Different types of plastic have been discovered till date since the discovery of plastic in 1907. Out of all Polyethene Terephthalate (PET) is one of the most commonly used plastic type.

Polyethene Terephthalate (PET) consists of polymerised units of the monomer ethylene terephthalate, with repeating units. Ethylene terephthalate (monomer) is manufactured by combining Terephthalic Acid (TPA) and Ethylene Glycol (EG). This reaction is a simple esterification reaction, in which there is a loss of water (dehydration reaction). The reaction is repeated multiple times to form a long chain polymer known as PET. It is a hard, stiff, and strong and a dimensionally stable material that absorbs very little water. PET plastic is also a good gas barrier and chemical resistance properties.

PET plastic is practically visible everywhere around in many forms due to its versatility. The polymer due to its ductility is used in forming threads commonly called as polyester, which has application in the textile industry for manufacturing wearables to carpets and many more things in between. The synthetic resin due to its stiffness and flexibility to be moulded into anything is used as single use plastic bottles with applications in varied range of products varying from cosmetic containers to microwave proof containers and other single use containers used in many different industries. Also, PET plastic due to its excellent chemical resistance and gas barrier properties, does not interact with many chemicals and hence it is approved by the U.S. Food and Drug Administration (FDA) for use in the food industry and is used for packaging all kinds of food products and beverages. Apart from all these major applications PET plastics are used in various industries for various application and is also often fused with other chemical materials to make products suitable for specific requirements.

PET plastic is generally disposed of in three different ways. First, it may be disposed of in landfills unsegregated. Second, PET plastic may be recycled to produce new PET products. Third, PET plastic may be chemically incinerated. The vast majority of PET plastic is dumped into landfills from households and there is no formal way of segregation. Only a few percent of PET plastic are recycled and made into new products.

The raw materials required to manufacture PET are obtained from crude oil. The reservoir of crude oil is depleted at an all-time high rate and PET production is only adding to the increase in rate of the very limited reservoir of crude oil.

Large majority of PET plastic is dumped into the oceans, and it degrades (not completely) into its microplastic form which is lethal to the ocean and the aquatic life and from there on its biomagnification can affect human lives in a drastically negative way. *(Issac, Kandasubramanian,2021)*.

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The microplastic form of the PET plastic leaches a chemical compound that mimics human estrogen and this compound especially in very low concentrations is very harmful to mammals, especially juvenile and infants. *(Yang, Yaniger, Jordan, Klein, Bittner, 2011)*

The productions of PET plastic use antimony trioxide as a catalyst, which is a compound that is classified as possibly carcinogenic and hence it makes PET production less safe. PET which is often used as a microwave proof container when exposed to microwave or when it is used for boiling, this causes the significant increase in the antimony levels often above the contamination limits prescribed by U.S. Environmental Protection Agency (US EPA) and this poses as a potential health hazard of PET plastic. *(Cheng, Shi, Adams, Ma, 2010)*

The idea being proposed is that the degradation of PET plastic can be done using biological agents primarily bacteria which can not only degrade PET plastic but also yield commercially viable products as a by-product thereby making the project if not completely self-sustainable, very cost-effective.

To degrade PET, the pipeline which was created is:

- Bacteria *Ideonella sakaiensis* to degrade PET plastic into ethylene glycol (EG) and terephthalic acid (TPA).
- Degradation of ethylene glycol into acetate by *Acetobacterium woodii*
- TPA into acetate and methane by bacterial *Pelotomaculum* and *Methanospirillum hungatei* that exist in an obligatory coculture for *Pelotomaculum* to work.

METHODOLOGY

Procurement of Raw material

PET plastic can be procured on a large scale from the wide range of sources available, like scrap vendors, waste/defective products from PET manufacturing plants, separated solid products from sewage treatment plants, etc.

Information About Bacteria and Their Culture

All the information about all the bacteria was obtained from **BacDive server** along with their culture medium requirements and the conditions which they live in.

Ideonella sakaiensis (**BacDive ID:** 140803) is an aerobic bacterium with optimum temperature range of 30-37°C and an optimum pH range of 7-7.5.

Acetobacterium woodii (**BacDive ID:** 5402) is an anaerobic bacterium with optimum temperature of 30°C. *Methanospirillum hungatei* (**BacDive ID:** 7132) is an anaerobic bacterium with an optimum temperature of 37°C. *Pelotomaculum* (**BacDive ID:** 11836) is an anaerobe with optimum temperature of 37°C and an optimum pH of 6.9 in the culture media. *Methanosarcina* (**BacDive ID:** 7086) is an anaerobe with optimum growth temperature of 37°C.

Working of Each Bacterium

Working of *I. sakaiensis*

PET is degraded to TPA and EG by the action of *I. sakaiensis* **(Figure 1 Part I)**. The aerobic bacteria attach to the PET surface by the help of its appendages. The bacteria then release the extracellular PETase that breaks the ester bonds in polymer by hydrolyzing them. As a result of which MHET (Mono-2-hydroxyethyl terephthalate) is obtained as the major product. MHET so obtained is then hydrolyzed by the intracellular MHETase to give EG and TPA. *(Taniguchi, Yoshida, Hiraga, Miyamoto, Kimura, Oda, 2019)*

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Working of *A. woodii*

The bacterium *A. woodii* degrades EG to ethanol and acetate **(Figure 1 Part II)**. It carries out the degradation reaction in two phases. In the first phase, conversion of ethylene glycol to acetate and ethanol takes place through the PduC subunit of the 1, 2-propanediol dehydratase PduCDE and CoA-dependent aldehyde dehydrogenase (PduP) or alcohol dehydrogenase. Then in the second phase, the ethanol so produced is converted into acetate and carbon dioxide is used for this conversion. This conversion takes place through the Wood-Ljungdahl pathway (WLP) via bifunctional acetaldehyde/ethanol dehydrogenase (AdhE). *(Trifunović, Schuchmann, Müller, 2016)*

Working of The Coculture

Terephthalic Acid was converted into methane and acetate with traces of benzoate **(Figure 1 Part III)**. This conversion was carried out by a coculture of *Pelotomaculum* and *Methanospirillum hungatei. Pelotomaculum* alone cannot degrade terephthalate as this degradation reaction is not energetically achievable, thus indicating that it is a syntrophic bacterium that depends strictly on the presence of hydrogen-consuming partner organisms (*Methanospirillum hungatei*) to grow on terephthalate and benzoate. *(Qiu, Sekiguchi, Imachi, Kamagata, Tseng, Cheng, Ohashi, Harada, 2004).*

ALTERNATE PATHWAY FOR THE ACETATE PRODUCED

The acetate so produced in point 2 and 3 can be converted to methane by using anaerobic bacteria known as acetotrophic methanogens belonging to the genera *Methanosarcina*. *(Ferry, 2020)* **(Figure 1 Part IV)**

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Small Scale Model

As seen in *Figure 2,* in order to degrade PET on a small scale, PET plastic is shredded into pieces which are then added to the culture of *Ideonella sakaiensis*, grown in minimal medium in a laminar hood. After completion of the degradation the process, culture is centrifuged at 10,000 rpm for 10 minutes and thereby yielding the products i.e., ethylene glycol and terephthalic acid. Separation of TPA & EG using distillation in which EG separate out first due to its boiling point being less than that of TPA. (Boiling point of Ethylene glycol: 197°C and Boiling point of TPA: 300°C). TPA so formed is further degraded by a coculture of *Pelotomaculum* & *Methanospirillum hungatei* in a sealed vacuum chamber. Resulting in formation of methane which escapes out to a separate collecting tank and acetate is formed. Acetate is separated as supernatant by centrifuging the culture at 10,000 rpm for 10 minutes (*Figure 3).* EG so formed from PET degradation is further degraded by *Acetobacterium wodii* in a sealed vacuum chamber. This results in the formation of ethanol and acetate, which are separated from culture by centrifugation at 10,000 rpm for 10 minutes. Ethanol and acetate separation using distillation (Boiling point of Ethanol: 78°C and Boiling point of Acetate: 118°C). According to the demand for methane, acetate can be converted to methane using bacteria *Methanosarcina,* and methane is recovered in a similar fashion as described in the case of coculture.

Large Scale Model

The process is like the one as described in the small-scale model except for a few changes, as seen in *Figure 4*. First, use of aerobic and anaerobic fermenters in place of petri dish in laminar hood or sealed tube respectively; Scaling up of the quantities of reactants at various stages of microbial degradation; Increase in the size of bacterial colony; As a result, quantity of the products retrieved is also increased.

Safe Disposal of Bacteria

After completion of each reaction periods, different species of bacteria involved are autoclaved at a temperature of 121° C for at least 30 minutes by using saturated steam under at least 15 psi of pressure. This is done before decomposing bacteria directly into the environment. Not all the bacteria are to be autoclaved, a small proportion of the culture is to be retained as inoculum for next batch of culturing process and the remaining portion of culture will be disposed of by autoclaving.

RESULTS

There are many traditional methods for disposing of PET plastic which are harmful for us and most importantly they are harmful to nature as well. So opposed to this primitive method of degradation (or disposal), we proposed a much environment friendly and even low risk method of PET degradation. This novel method will not harm nature and will also be beneficial for mankind as it can generate many commercially viable products, many of which can be a good replacement to the conventionally used products.

Commercial Uses of the By-products

The microbial degradation of PET plastic produces some by-products which are of great commercial significance. Terephthalic acid so produced can be used carrier in paint, raw material for certain drugs and Used as a filler in some military smoke grenades.

Ethylene glycol, a good dehydrating agent, can be used as an antifreeze and coolant for cars engine. It is the primary substance to produce polyester fibers and acts as a catalyst in the making of fiberglass.

Acetate is a key material for the manufacturing of cellulose acetate, used in photography as a film base. Acetate can also be used in the production of ethyl acetate and butyl acetate which are the solvents for resins, paints, adhesives, and lacquers. It is also used to manufacture acetic acid commonly known as vinegar.

Ethanol, by-product produced in the reaction carried out by the coculture can possibly be used as a component of gasoline and gasohol. It can also be used in medical wipes and also in manufacturing of hand sanitizers which have become one of the things to be compulsory used in the times of the COVID-19 pandemic.

Methane, another by-product, when combined with other gases, becomes an excellent biofuel which can then be utilized for heating, power generation and transportation. It can also be used in hydrogen production and thereby for production of ammonia and fertilizers.

Methane can also be potentially used in manufacturing of valuable chemicals like methanol, chloroform, carbon tetrachloride and nitromethane.

Cost effectiveness and Efficiency

The project that is being proposed has the potential to be highly cost effective. The commercial value of all the by-products that will be produced will aid in substantially reducing the cost of the project and in a few years, it will be a **profit generating project.** Additionally, as per the demand of the product, process can be regulated in a way that **only the required product is produced.** Since the raw material (PET plastic) is freely available therefore, so there will be no or minimal additional cost for it. The bacteria used in this process of degradation must be acquired once at a low cost and then all the bacteria can be grown in-vitro to carry out next cycle of reactions. Also, the cost of chemicals, regulating pollution and other logistics expenses are nullified in this process.

Degradation of PET plastic into EG and TPA by *Ideonella sakaiensis* takes 6 weeks for complete degradation. Similarly, to completely degrade EG into ethanol and acetate by *Acetobacterium wodii* takes only 2 hours for complete degradation. Also, to degrade TPA to methane and acetate, the coculture (*Pelotomaculum & Methanospirillum hungatei)* takes about 15 days. This time is taken by each bacterium in each step is to carry out complete degradation of the products, but the process can be stopped in between and altered with, to produce the byproduct of choice as per the requirement.

The microbial degradation of PET plastic is a process that has multiple benefits like high efficiency, cost effectiveness, commercially useful byproducts, etc. Along with these benefits this process is also highly environment friendly and helps in achieving sustainable development in a major way.

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DISCUSSION

The by-products produced by the microbial degradation of PET plastic not only yields economically viable products but also great alternatives to the products or chemicals that are conventionally used. Like the use of ethylene glycol for making polyester fibers reduces the dependency on coal and petroleum to minimal levels. Use of ethanol as a part of gasoline and gasohol is replacing diesel and petrol as fuels for vehicles because when ethanol/ gasoline-gasohol combustion releases carbon dioxide and water which are not harmful except carbon which when increased can cause greenhouse effect, but it is much less harmful than other pollutants like oxides of Sulphur and nitrogen, etc. Also use of methane in combination with other gases can be used as biogas, as a fuel for heating purposes will be beneficial because it has a very high calorific value and will provide a good amount of heat also will act as an alternative fuel source to the currently used petroleum products. The biogas will also reduce the amount of pollution and dependence on conventional materials like wood, kerosene, LPG and PNG for fuel purposes. These uses of the by-products generated by biodegradation of PET plastic makes the **project** an environment friendly one.

The forementioned proposal provides a way to degrade plastic in a more efficient and friendly way. Plastic is one of the biggest problems of our time and if its degradation process is implemented well, so this will not only clean the environment and reduce the waste on the planet but also will help in achieving and fulfilling the idea of/behind sustainability. The scientific degradation will be easily carried out in a responsible way and keeping the environment in mind. Also, this process of degradation will also help us achieve and fulfil a wide range of sustainable developmental goals and even their individual sustainable developmental targets, as proposed by the United Nations (UN) which provides a blueprint to achieve a better and more sustainable future for all.

The forementioned proposal will deal with one of the biggest problems of our time i.e., Plastic. This in turn will help us achieve the sustainable development goal (SDG) 12 i.e., Responsible Consumption and Production. Biodegradation / Degradation of PET will help in responsible regulation of plastic waste. Also, Degrading PET using bacteria in fermenters would not release any gases or bacteria directly in the environment, preventing pollution. Additionally, the commercially useful products produced by the degradation process as by products will be produced responsibly. Using methane will reduce use of our dependency on petroleum products and bring about sustainability.

Target 12.5 of sustainable development goal 12 says that efforts should be made to substantially reduce waste production. The project aids in achieving that goal. The biological degradation of PET will potentially reduce its accumulation in landfills. This plastic waste would be utilized in forming various commercially useful products. This project if implemented well would potentially reduce the amount of plastic waste in the environment.

The sustainable development goal states that “Build resilient infrastructure, promote inclusive and sustainable industrialization and foster innovation.” If the project takes place on a large-scale, then the infrastructure build-up can be constructed in a resilient way keeping sustainability in mind. The industrial setup can promote inclusiveness and sustainable industrialization. Finally, the project is one of its own kind projects and is very innovative, which promotes the sustainable development goal in a positive way.

Since the project promotes sustainable infrastructure, promotes inclusive and sustainable industrialization due to which its falls in line with following and promoting target 9.1 and 9.2 of the sustainable development goal 9. Target 9.1 says develop quality, reliable, sustainable, and resilient infrastructure. And target 9.2 says Promote inclusive and sustainable industrialization. Target 9.3 says that enhance scientific research, upgrade the technological

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capabilities of industrial sectors. The project enhances and uses biotechnological research and creates job opportunities related to the same.

Our project is fulfilling the SDG 8 i.e., Decent Work and Economic Growth. This project requires a team of skilled scientists and technicians for monitoring and maintaining the culture and reactions carried by them. If setup in rural areas, the project could provide employment to the people living in that area in various fields like maintenance of the plant or if someone is skilled and experienced enough then he or she can be included in the team of scientists and technicians. The by-products formed will require transportation facilities for transporting them to other sites or industries who will require these by-products as reactants for manufacturing various other commercial products. Hence the proposed project will provide decent work to the needful and as a result of which economic growth of the area concerned will increase due to the manifold profit growth resulting from the production of these profit-oriented by-products of microbial degradation of PET plastic.

The project can surely generate profits for its owners and large-scale industries. But this may provide good business opportunities to small and medium scale enterprises. In the times of the pandemic COVID-19, hand sanitizers are of much use and these are being manufactured by not only big scale enterprises but also by small and medium scale enterprises. So, the ethanol so produced can be given at a very low cost or can be donated also to these small and medium scale enterprises, thereby reducing their cost of manufacturing. These types of policies will promote entrepreneurship among the population people. Hence through such approach we can also fulfil/achieve the target 8.3 of SDG 8 which is, "Promote development-oriented policies that support productive activities, decent job creation, entrepreneurship, creativity and innovation, and encourage the formalization and growth of micro-, small- and medium-sized enterprises, including through access to financial services".

This project will aid in fulfilling the target 1.b under SDG 1, i.e., Create sound policy frameworks at the national, regional and international levels, based on pro-poor and gender-sensitive development strategies, to support accelerated investment in poverty eradication actions. This is done by employing the people of rural areas (where the plant is/can be set) in doing variety of jobs ranging from maintenance of the plant to scientists.

The methane so produced does not get escaped into the atmosphere but gets supplied directly to the biogas plant through the underground pipe. So, by this the methane so produced will not cause the greenhouse effect and thereby the global warming. Through this we can fulfil SDG 13 which is Climate Action.

CONCLUSION

The proposed project aimed to degrade PET plastic using different bacteria at different steps of the process, produced different types of by-products. The by-products so formed are of great commercial significance across various industries. These by-products and their process of manufacturing using different bacteria helps to make the process sustainable.

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FIGURES

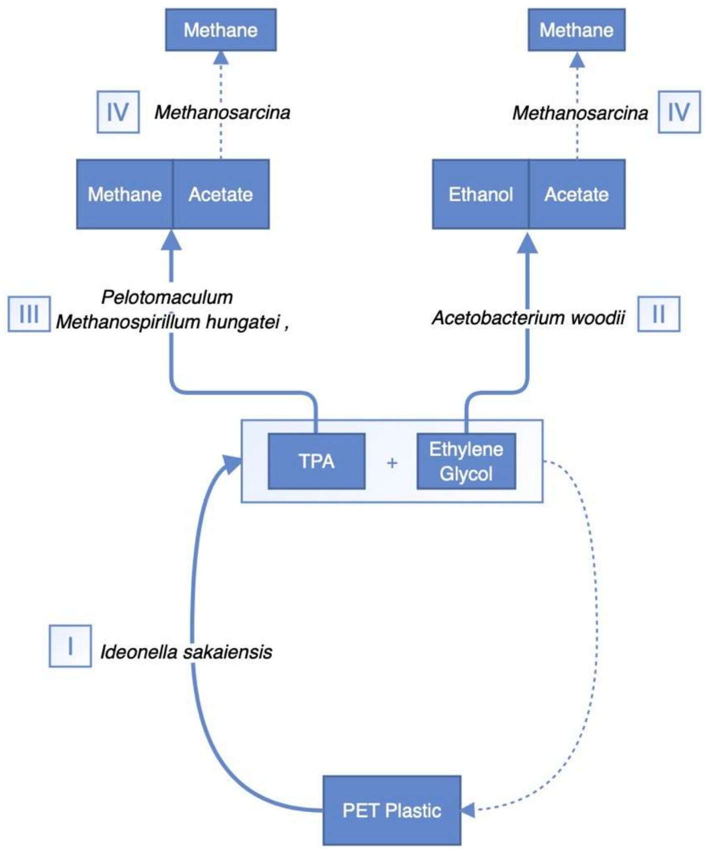

Figure 1: General outline of complete microbial degradation of PET plastic.

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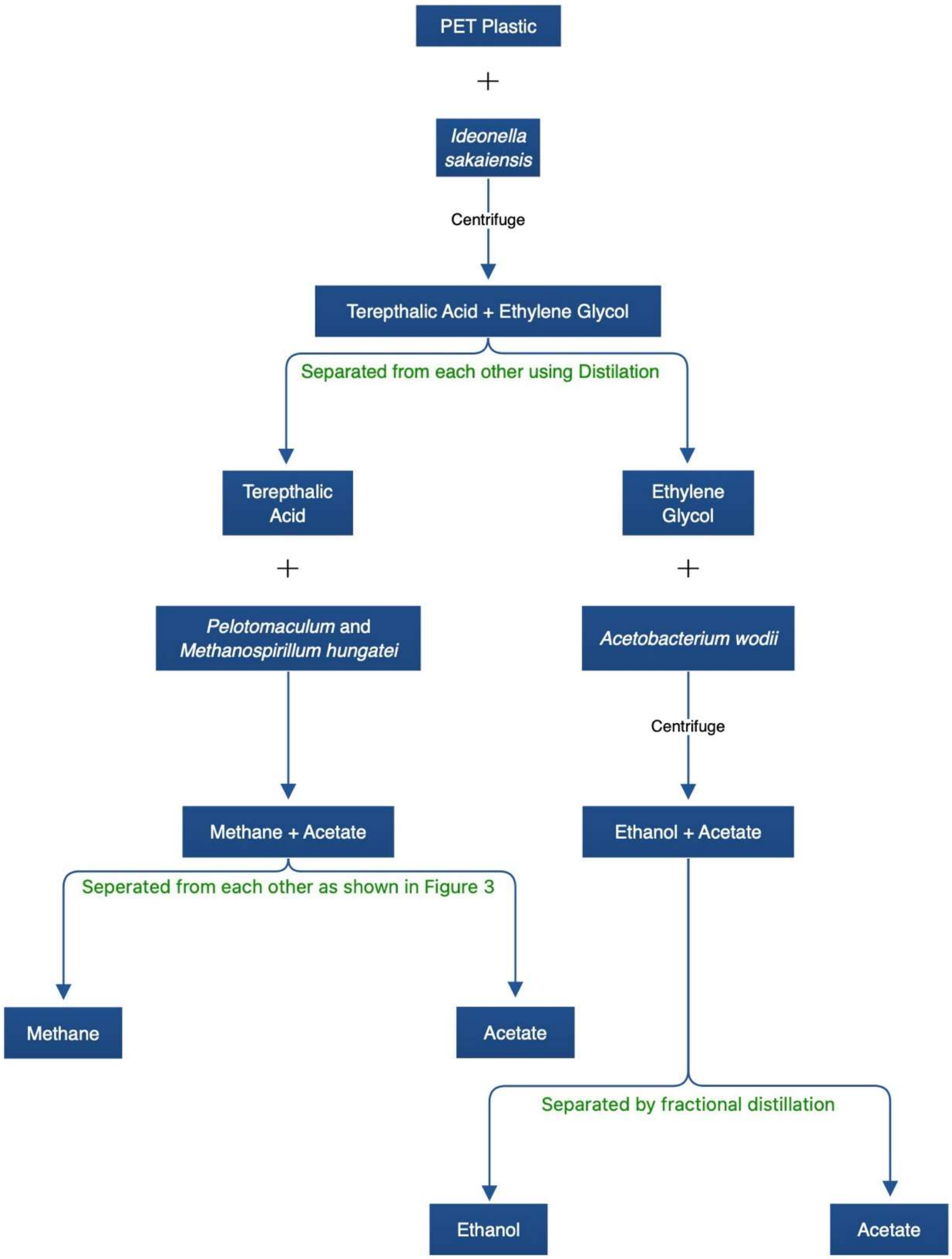

Figure 2: Small scale model of degradation of PET plastic

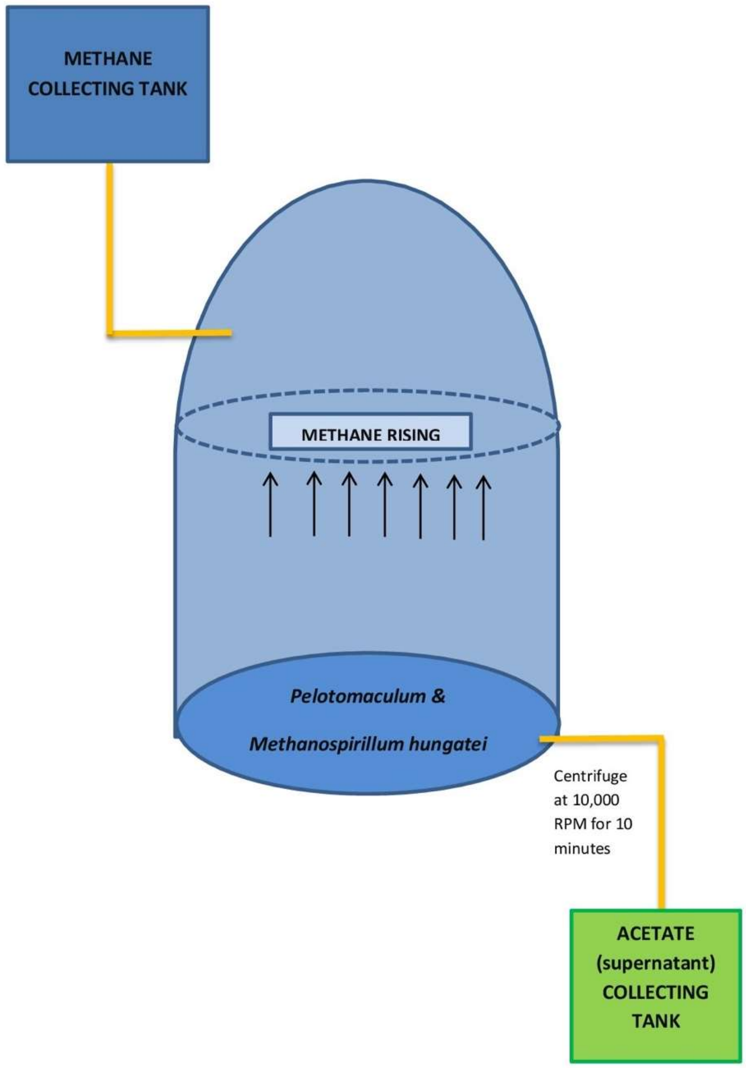

Figure 3: Separation of the products formed by TPA degradation.

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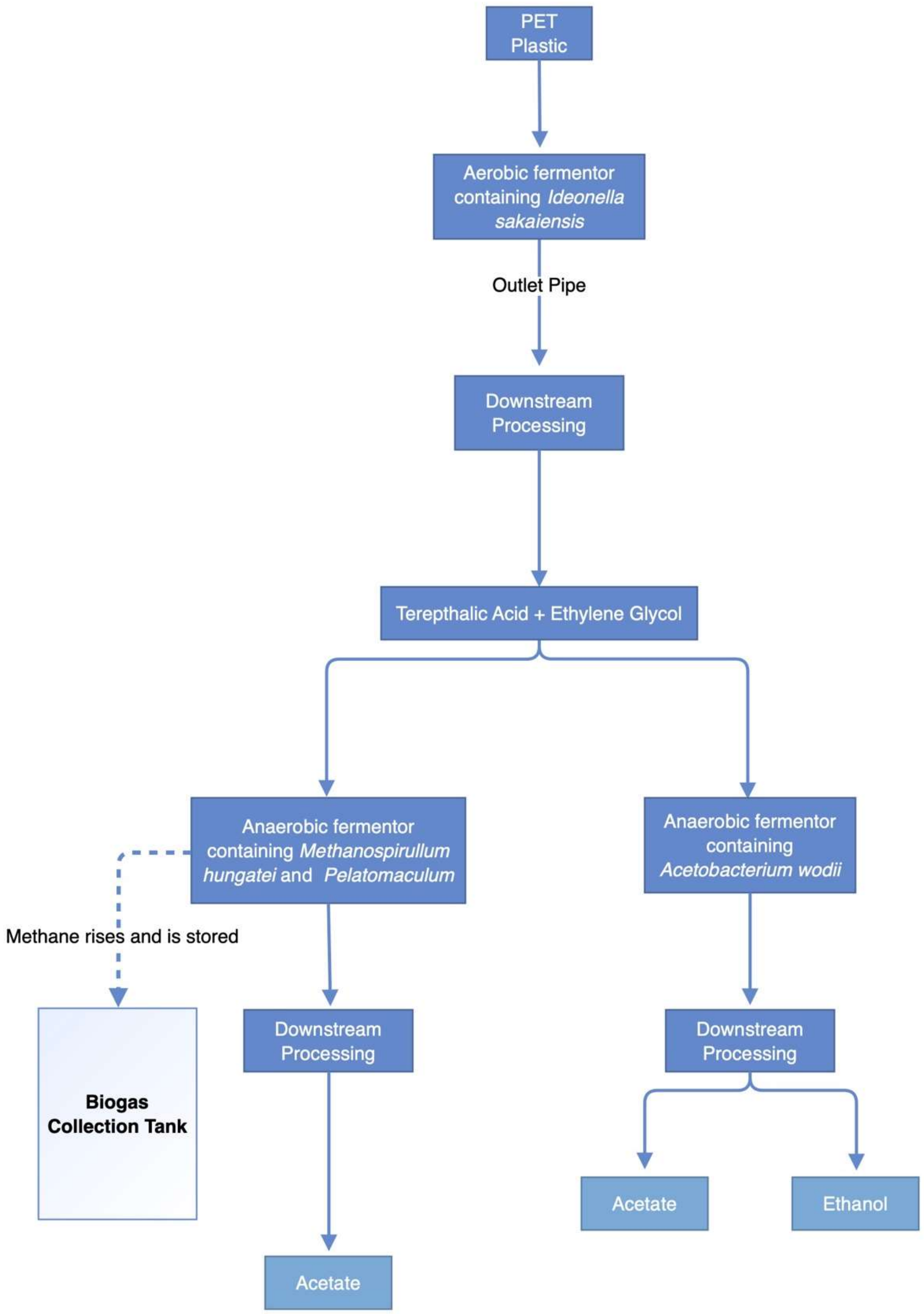

Figure 4: Large scale model of degradation of PET plastic