Plastic-degrading microbes

Why is there so much plastic in nature, but dead trees and animals disappear?

Eva C. Sonnenschein

Department of Biotechnology and Biomedicine, Technical University of Denmark, Kgs. Lyngby, Denmark

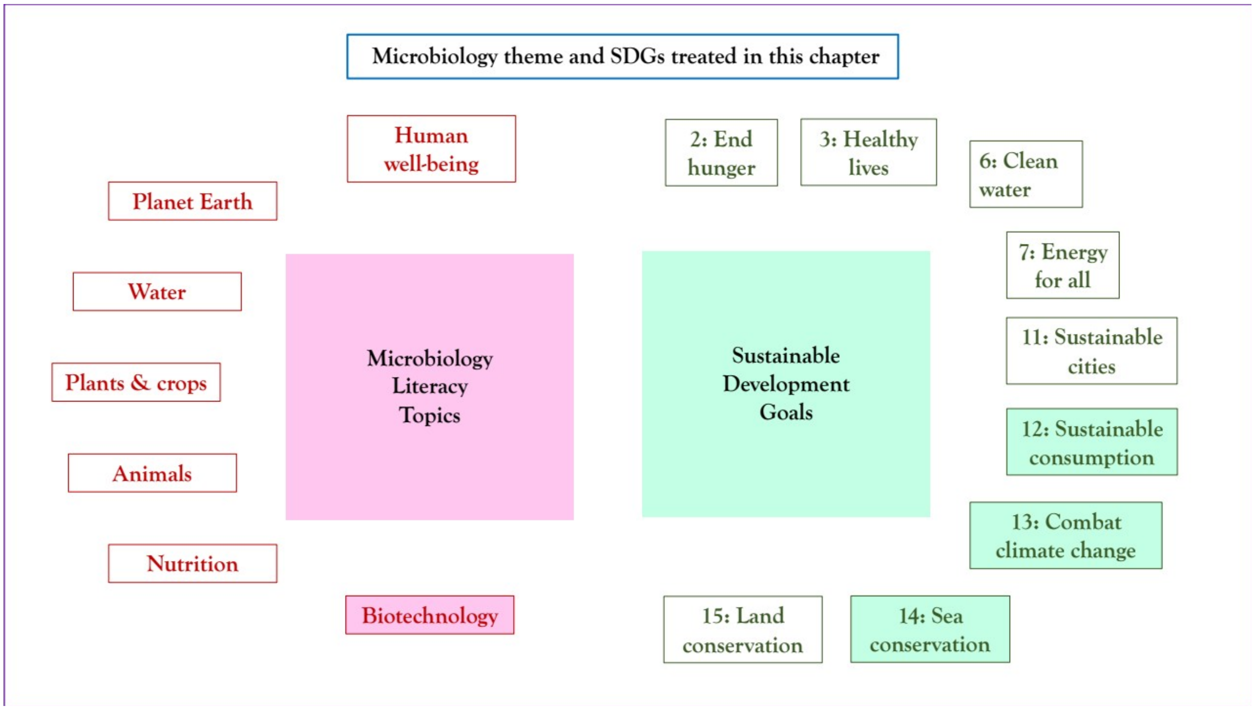
Plastic-degrading enzymes

Storyline

Today, plastic – a group of synthetic materials consisting of different types of largely inert organic polymers – is omnipresent in nature and potentially harmful to wildlife and us, humans. However, many natural polymers such as cellulose (from plants) or chitin (from fungi and crustaceans) can be degraded by microorganisms. Some microbes secrete specialized enzymes to break these molecules down and feed on them – like we digest our food. Many microbial enzymes are utilized in everyday products (such as laundry detergents) or in industrial and medical applications. So, why do microorganisms not break down the plastic in nature? Or do they? Actually, a few plastic-degrading microorganisms have been isolated and characterized in the laboratory. Thus, bacteria have evolved to utilize this novel material as food, giving hope to possibility that we could exploit their enzymes to re-use plastic waste and create a sustainable solution for the plastic problem. Currently, the microorganisms and their enzymes are however not efficient enough to reduce the plastic waste in the environment. If that were the case, it might also cause problems, as we humans are also very dependent on the durability of plastic. Thus, microorganisms harbor a great natural reservoir for biotechnological solutions that could support the Sustainable Development Goals but, as with any new invention, the effect on all stakeholders needs to be carefully evaluated.

The Microbiology and Societal Context

Microbiology: biodiscovery of novel bioactivities; biodegradation; bioremediation; biotechnology; sustainable products; polymer degradation; plastic microbiome. *Societal*: healthy environments; healthy lives; waste management; wastewater treatment; circular economy

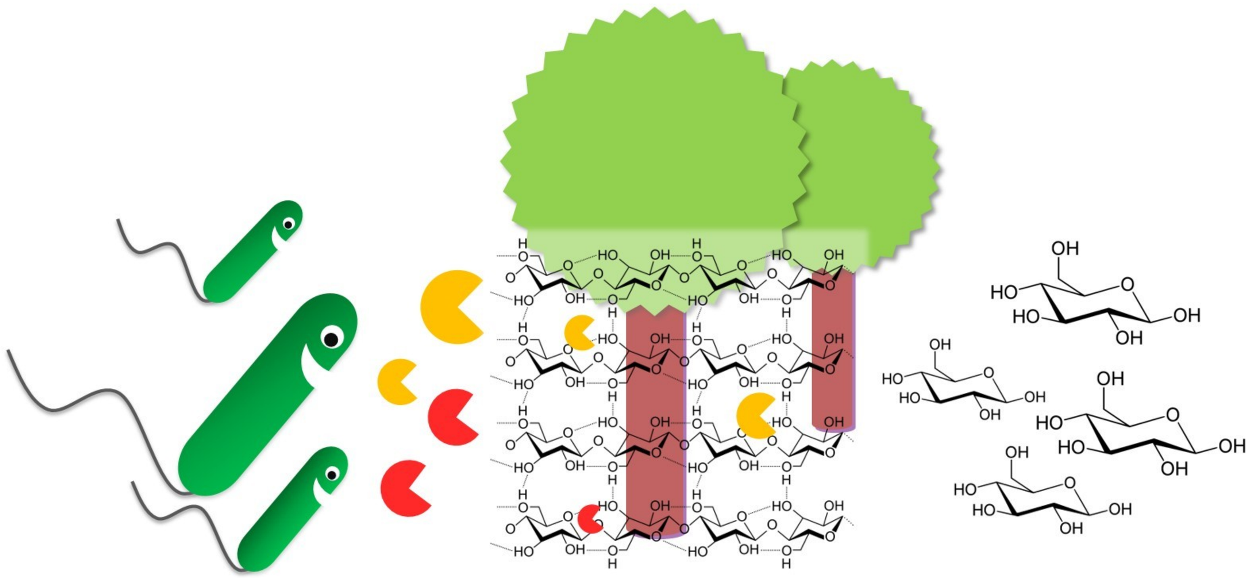


Plastic-degrading enzymes: the Microbiology

1. ***Employing microorganisms and their products to help us.*** Many of our everyday products contain substances originally made by microorganisms. The antibiotic penicillin for treatment of bacterial infections was first discovered to be produced by a fungus. Many laundry detergents contain microbial protein- or fat-degrading enzymes. Microorganisms produce these compounds to survive and thrive in the environment, to break down their food, or chase away other microorganisms that are interested in their dinner. These natural abilities of microorganisms have – unknowingly – been exploited by humans over thousands of years for production of food and beverages such as bread, cheese and wine, or medical treatment. With modern technologies, mankind learned more and more about these abilities, and improved the exploitation of microbial processes for various applications in the food industries, but also in paper and textile production, pharmaceutical development etc. To fulfil the world population´s growing and changing needs, while respecting the essential implementation of sustainable solutions, these biotechnological processes become more and more important in today's life.

2. ***Environmental microorganisms degrade natural polymers***. Generally, organic substances produced in nature are degraded by microorganisms and thereby recycled. Even very durable polymers such as cellulose or chitin providing structure to plants and animals are degraded over time. Microorganisms secrete enzymes to break down these long polymer chains into smaller pieces that they then can take up and utilize in their own metabolism. These enzymes, namely cellulases and chitinases, can also be produced industrially. Cellulases are utilized in the pulp and paper industries, for food processing, as nutritional supplement or to produce the medically relevant chitosan. Chitinases are used in agrochemical and medical applications. In nature, all organisms are competing for food and they evolve to extract their nutrition from any kind of material that

they have available, even if they have to invest some of their energy to produce and secrete enzymes to degrade persistent materials.



Bacteria producing cellulases that degrade the cellulose from trees into its monomer, glucose.

3. ***The plastic problem.*** Since the introduction of mass production mid last century, plastic production has been growing continuously, leading to an estimated sum of 8300 million metric tons by 2015. In 2015, only around 9% of waste was recycled while 79% was deposited in landfills and the environment. It has been estimated that by 2050, there will be more plastic than fish in the oceans (by weight), if no significant countermeasures are taken. Plastic is a synthetic or semi-synthetic material consisting of different types of inert organic polymers. In the environment, large plastic items can be fragmented by abiotic parameters such as light, oxidation, thermal and mechanical actions down to a size below 5 mm, the microplastics. The fate and effects on the health diverse organisms in the biosphere of these microparticles is unresolved.

4. ***Plastic-degrading microorganisms***. A possible sink for microplastics is microbial degradation, the breakdown of a complex chemical structure by microorganisms. The microbial composition on marine plastic debris is unique in comparison to that of the surrounding environment (seawater, non-plastic debris), hence indicating a specificity for living on and, possibly, off plastic. In the laboratory, several microorganisms have been found to degrade different types of plastic. A polyethylene terephthalate (PET)-degrading enzyme, named PETase, was isolated from a marine bacterium, *Ideonella sakaiensis*. The PETase is proposed to have evolved from cutinases that catalyze hydrolysis of cutin, a major component of the plant cuticle. This enzyme has been engineered to become a more efficient plastic-degrading enzyme variant.

Being exposed to PET in the environment, *I. sakaiensis* will have gained an evolutionary advantage against competitors by evolving its enzymes to specifically attack PET. This microbial adaptation to its environment gives hope that there could be a biological solution to the plastic problem.

5. *Biodiscovery of plastic-degrading microorganisms*. Like explorers hunting for hidden treasures, microbiologists use many tools and strategies to bio-discover novel microbial bioactivities such as plastic degradation. Today, both cultivation-dependent and cultivation-independent approaches are employed for biodiscovery. Cultivation and isolation of microorganisms can be based on classical plating of environmental samples on solid media in Petri dishes containing different nutrient compositions and concentrations. There are also more elaborate approaches utilizing the actual ecological parameters of the microorganisms such as iChip (in which the microorganisms are cultivated on a chip in the environment) or dilution-to-extinction (where the microorganisms are separated from microbial competitors using dilution) that can extract a greater or different microbial diversity from nature into the laboratory. These cultivation approaches could directly be targeted to plastic-degrading microorganisms, by providing plastic as the only energy source. Cultivation-based techniques will however always only recover a fraction of the actual biodiversity, since some microorganisms will require unknown factors (e.g. specific microbial interactions), and hence will not grow in the laboratory.

Therefore, cultivation-independent biodiscovery approaches have become very useful. Here, one queries nucleotide or protein data (from genomes or metagenomes or –transcriptomes) for specific features such as plastic-degrading enzymes. The efficiencies of these enzymes can then be tested by heterologous expression in laboratory host organisms (like *Escherichia coli* or yeast) without even knowing the natural producer of the enzyme.

Having discovered plastic-degrading microorganisms or enzymes, they can be further optimized by modern metabolic or protein engineering, as has been demonstrated for the PETase; this represents the current stage (2020) of development of the plastic-degrading activities and making the enzymes more efficient will be key to their application.

6. *Development of plastic-degrading microorganisms and their enzymes*. With highly efficient plastic-degrading microorganisms or enzymes at hand, their applications would need to go through careful risk assessment. Utilization of any strains or enzymes in the environment should be closely monitored to evaluate a possible uncontrolled spread of their bioactivities. Biofilters entrapping live, plastic-degrading microorganisms or their enzymes could provide an applicable method for bioremediation or wastewater treatment. Biotechnological implementation of plastic-degrading metabolic pathways for the utilization of plastic as a feedstock for cell factories could be controlled in closed bioreactors.

While the plastic problem in the environment and in landfills is a great challenge, plastic remains a very important and useful material and uncontrolled destruction of it will have serious consequences and needs to be avoided in any way.

Relevance for Sustainable Development Goals and Grand Challenges

The microbial dimension of developing novel enzymes for plastic degradation relates to several SDGs (*microbial aspects in italics*), including

- **Goal 12. Ensure sustainable consumption and production patterns**. Currently plastic usage is a one-way street as it is produced from fossil fuels and ends in the environment/landfills or is incinerated. Enzymatic processes could contribute to reuse or recycle plastic so that it can enter a circular economy.

- **Goal 13. Take urgent action to combat climate change and its impacts.** Production of virgin plastic has a significant carbon impact and recycling of the plastic could help reduce this footprint.
- **Goal 14. Conserve and sustainably use the oceans, seas and marine resources for sustainable development.** Virgin plastic is produced from fossil feedstocks and exploitation of fossil fuel can lead to drastic contamination of the environment. Finding sustainable solution to recycle plastic will protect biodiversity and marine resources.

Pupil participation

1. ***Class discussion***
   a. Which items that you use every day contain plastic?
   b. What if these things would suddenly disappear?
   c. Which household products contain enzymes?
   d. Where would you search for novel bioactivities and why?
   e. Can you identify other areas where microorganisms or their enzymes could replace current (chemical) techniques?
   f. How could we remove the existing plastic waste from the environment?

2. ***Pupil stakeholder awareness***
   a. Why is sustainability important in everyday life?
   b. How can we as society reduce plastic waste?
   c. Who would be affected when plastic is removed from the environment?

3. ***Exercise***
   a. Why is plastic persistent in the environment?
   b. How can we discover novel microbial activities?
   c. How could we employ plastic-degrading enzymes in an application?

The evidence base, further reading and teaching aids

Can Microbes Clean Up Our Oily Mess? – by the Scientific American https://youtu.be/a_HWlFzgQiM

German scientists identify microbe that could help degrade polyurethane-based plastics – frontiers Science News https://blog.frontiersin.org/2020/03/27/german-scientists-identify-microbe-that-could-help-degrade-polyurethane-based-plastics/

Plastic Degrading Microbes For a Cleaner Future – by Sarah Wettstadt https://sarahs-world.blog/bacteria-degrade-plastic/

Danso D, Chow J, Streit WR (2019) Plastics: Environmental and Biotechnological Perspectives on Microbial Degradation. Appl Environ Microbiol, 85 (19): 1–14. https://doi.org/10.1128/AEM.01095-19
Ellen MacArthur Foundation (2016) The New Plastics Economy: Rethinking the future of plastics http://www3.weforum.org/docs/WEF_The_New_Plastics_Economy.pdf
Roager L, Sonnenschein EC (2019) Bacterial Candidates for Colonization and Degradation of Marine Plastic Debris. Environ Sci Technol 53 (20), 11636–11643. https://doi.org/10.1021/acs.est.9b02212
Yoshida S, Hiraga K, Takehana T, et al (2016) A bacterium that degrades and assimilates poly(ethylene terephthalate). Science 351:1–5. https://doi.org/10.1126/science.aad6359

Glossary

Enzyme: proteins that act as biological catalysts
Polymer: macromolecules that are composed of many repeating subunits
Biotechnology: the research and development of products using living organisms
Bioremediation: the process to clean-up contaminated environments using living organisms
Biodiscovery: the discovery of novel biological molecules or organisms