Bacterial Dynamics During the Degradation of Starch-based Bioplastic and Oxo- degradable Plastic in Compost Soil

Joshua Abednego Wicaksono
Atma Jaya Catholic University of Indonesia
Tresnawati Purwadaria
Atma Jaya Catholic University of Indonesia
Adi Yulandi
Atma Jaya Catholic University of Indonesia
Watumesa Agustina Tan (tan.watumesa@atmajaya.ac.id)
Atma Jaya Catholic University of Indonesia

Research Article

Keywords: Starch-based bioplastic, Oxo-degradable plastic, Degradation, Soil metagenome

Posted Date: February 14th, 2022

DOI: https://doi.org/10.21203/rs.3.rs-1315062/v1

Abstract

Background: Plastic waste accumulation is one of the main ecological concerns in the past decades. A new generation of plastics that are easier to degrade in the environment compared to conventional plastics, such as starch-based bioplastic and oxo-degradable plastic, is perceived as a solution to this issue. However, the main degraders of such materials and their fate in the environment are unclear. In this study, we monitored the dynamics of bacterial community in soil upon introduction of commercial starch-based bioplastic and oxo-degradable plastic. The plastics were buried separately in compost soil and incubated for 30, 60, 90, and 120 days. Following incubation, soil pH and temperature as well as the weight of remaining plastics were measured. Bacterial diversity in soil surrounding the plastics was analyzed using Illumina high-throughput sequencing of the V3-V4 region of 16SrDNA.

Results: Starch-based bioplastic degraded quicker than oxo-degradable plastic in soil. The bacterial composition in soil fluctuated over time with or without the introduction of either type of plastic. While the major bacterial phyla remained similar for all treatment in this study, different types of plastics led to different soil bacterial community structure. None of these bacteria were abundant continuously, but rather they emerged at specific time points. The introduction of plastics into soil increased not only the population of bacteria with the ability to directly utilize plastic component for their growth, but also the abundance of those that may interact with direct degraders. Interestingly, there were no specific genera that were prevalent throughout the burial period.

Conclusions: The degradation of both starch-based bioplastic and oxo-degradable plastic involves a complex set of bacteria that continues to shift over time.

Background

Petroleum-based polyethylene has been used widely for consumer products due to its durability and low cost. About 50% of polyethylene products were single-use and this has led to plastic waste this has led to plastic waste accumulation issues [1]. In 2015, the global cumulative plastic production has risen to over 7.82 billion tons per year. Only 19.5% of the plastic waste was recycled, 25.5% incinerated, and the rest 55% ended up in landfills or polluting the ocean [2]. Recent studies have also demonstrated that overtime plastic waste will be degraded into smaller fragments known as microplastics [3, 4]. Multiple studies have also indicated the presence of microplastics in marine animals that are used for human consumptions, such as in fish, seashells, and crabs [5, 6]. To date, there is no clear information on the impact of prolonged microplastic ingestion on health.

One approach to combat petroleum-based plastic waste accumulation is by designing a new generation of plastics that is easier to degrade than petroleum-based plastics. This was achieved by engineering renewable plant or bacteria-based polymers, commonly known as bioplastics. The resources for bioplastics can be derived from plants like cassava, corn, and potato starch [7] or polymers derived from microorganisms such as poly(3-hydroxyalkanoate) and poly(3-hydroxybutyrate) [8]. These materials are

considered biodegradable and will be decomposed by bacteria and fungi into simpler molecules and used for metabolisms [9]. The starch component in starch–based bioplastics, among which, can be directly utilized as a carbon source by bacteria and fungi [10]. Starch is a polymeric carbohydrate that consists of repeated glucose molecules chained with glycosidic bonds. Amylase cleaves the glycosidic bonds in starch to generate oligo-, di-, or monosaccharides, all of which play a role as growth substrates for microorganisms [11].

Another strategy is to incorporate metallic salt additives in the petroleum-based low-density polyethylene to promote its degradation, more commonly known as oxo-degradable plastic [12]. Metallic salts act as pro-oxidants to initiate photo- or thermo- oxidation of polyethylene [12]. These reactions increase the hydrophilicity of the long -chained polyethylene polymer and break it down into lower molecular weight fragments there are presumably more susceptible to microbial attack [13, 14]. The biodegradability of oxo-degradable plastic, however, is debatable as it is unclear whether the plastic is truly decomposed into simpler molecules or merely broken down into microplastic fragments [15].

There is limited information on specific microbes that play a role in the decomposition process of the above easier-to-degrade plastics and how they may affect the microbial composition in the environment. Therefore, this research is aimed to analyze changes in soil bacterial community during the introduction of commercially available starch-based bioplastic and oxo-degradable plastic. Such information is of importance in determining particular bacterial groups that may be used to accelerate both plastic waste decomposition as part of a holistic approach in combating plastic pollution issues.

Methods

Sample preparation

Commercial cassava starch-based bioplastic and oxo-degradable plastic bags available for sale in Indonesia were used in this study. They were cut into random-sized sheets, weighed to 75 g, UV sterilized and buried at 10 cm depth in compost soil (Sahabat Tani brand; contains a mixture of guano, humus, manure, roasted rice husks, dolomite and cocopeat) in separate plastic pots (30-cm diameter). The pots were incubated outdoor in a shaded area for 30, 60, 90 and 120 days, respectively. Soil with no plastic collected from the same depth at each time point served as controls. The inner temperature and pH of soil were measured using a digital thermometer and a pH meter at each time point, respectively. At the end of each treatment, remaining plastic was weighed to evaluate plastic degradation. Soil surrounding the plastic was sampled and stored at 4ºC prior to further analysis.

Soil DNA extraction

Total DNA from soil surrounding the plastic was extracted using the Presto™ Soil DNA Extraction Kit (GeneAid, Taiwan) according to its manufacturer's protocol and stored at -20°C. Soil samples that were not directly analyzed were stored at 4°C for maximum 14 days. Total DNAs were visualized on a 1% agarose gel and quantified using NanoDrop™ 2000 (Thermo Fischer Scientific, USA).

V3-V4 library preparation and high-throughput sequencing

The V3-V4 region of 16S rDNA was amplified using the primer pair 341f (5'–GTGCCAGCMGCCGCGGTAA-3') and 806r (5'-CCGTCAATTCCTTTGAGTTT-3') [16]. Amplicons were generated and tagged with unique sequence barcode for each samples with NEBNext® UltraTM DNA Library Prep Kit for Illumina. Sequencing was done using the Illumina platform at NovogeneAIT, Singapore.

Bioinformatics analysis

Overlapping DNA fragments were merged using FLASH (V1.2.7) [17]. Quality filtering using specific conditions [18] was done to achieve high quality clean tags according to QIIME (V1.7.0) [19]. Chimera sequences were removed by comparing the tags to SILVA database (http://www.arb-silva.de/) using UCHIME algorithm [20, 21]. Uparse software (V7.0.1090) was used to assign the sequences that have ≥97% similarity into the same OTU [22]. Shannon and Simpson indices were calculated using QIIME (Version 1.7.0) and displayed with R software (Version 2.15.3). Representatives of each OTUs were screened with QIIME (Version 1.7.0) [23] using Mothur against SSUrRNA database of SILVA Database for species annotation at each taxonomic rank [24, 25]. The taxonomic relationship of all representative sequences was analyzed using MUSCLE (Version 3.8.31) [26].

Results And Discussion

Weight decrease of degradable plastic in soil

There was a contrast between the degradation rate of starch-based bioplastic and oxo-degradable plastic. After 30 days, bioplastic weight was reduced by 55.61%. The weight reduction was less drastic afterward, with a 73.65% weight decrease by day 120 (Fig. 1). A study done by Accinelli et al. [27], in which a corn starch-based bioplastic bags was buried at 25°C in the dark, also reported a 43% decrease in plastic weight after 90 days of incubation in compost soil. In contrast, oxo-degradable plastic weight was only reduced by 2.84% even after 120 days. Similarly, Mantia et al. [28] showed that the oxo-degradable plastic degradation rate was only around 1-1.5% after 120 days burial in soil, even in combination with photooxidation pre-treatment.

The starch content in bioplastic may be more readily used as a carbon source by soil bacteria [10] compared to the organic component in oxo-degradable plastic, and therefore the former was degraded faster in soil. The degradability of starch-based bioplastic also dependent on its starch content [29]. A study by Liew and Khor [30] showed that higher starch content led to higher degradation rates of starch-based bioplastic. Microbial activities, particularly degradation in this context, are affected by many factors, including soil moisture, temperature, and humidity [31]. The soil pH (8.6±0.2) and temperature (25°C) of soil in this study, however, remained constant at all time points, meaning that the introduction of neither starch-based bioplastic nor oxo-degradable plastic affected both environmental parameters. In the case of oxo-degradable plastic degradation, it should also be noted that the UV sterilization step prior to plastic burial might led to slight physical degradation. The metallic ions added in oxo-degradable

plastic acted as prooxidants [12] that are further photo-oxidized by UV light. Martín-Closas et al. [31] demonstrated that UV light exposure induced the oxidative cleavage of the polyethylene chain in oxo-degradable plastic.

Diversity indices

The diversity and richness of bacterial OTUs were quantified and expressed as Shannon and Simpson indices, respectively [32]. While the Shannon index fluctuated over time in soil with no plastic treatment, the bacterial diversity within soil introduced to both starch-based bioplastic and oxo-degradable plastic was directly proportional to the incubation time, in which the lowest diversity index was observed on day 30 (Bt30 and Ot30) and the highest on day 120 (Bt 120 and Ot120) (Table 1). Compared to the control group, soil treated with both types of plastic showed higher diversity over time, except on day 60. The Simpson index in soil treated with both types of plastic also showed that their presence led to a slight increase in bacterial richness over time.

Table 1. Diversity and richness of bacteria in soil following starch-based bioplastic and oxo-degradable plastic burial

Sample name	Treatment	Shannon index	Simpson index
Ct0	No plastic exposure (control)	7.263	0.952
Ct30		7.386	0.970
Ct60		8.014	0.985
Ct90		7.749	0.982
Ct120		7.537	0.972
Bt30	Starch-based bioplastic	7.503	0.969
Bt60		7.801	0.977
Bt90		7.908	0.982
Bt120		8.055	0.984
Ot30	Oxo-degradable plastic	7.407	0.971
Ot60		7.589	0.972
Ot90		8.232	0.987
Ot120		8.591	0.990

Numbers following each sample code indicate the length of plastic burial in days. The value of Shannon index represents bacterial diversity, in which higher numbers indicate higher diversity. The Simpson index

has a maximum value of 1, which signifies that all members of the population are present in equal abundance.

Bacterial community in compost soil introduced with starch-based bioplastic

The most dominant phyla in control soil and soil introduced with starch-based bioplastic were Bacteroidota, Proteobacteria, and Actinobacteria. This is in line with a report by Meng et al. [33] that explored microbial succession during cow manure and corn straw composting. Proteobacteria and Actinobacteria abundance increased in soil over time regardless of the introduction of starch-based bioplastic (Fig. 2a). The increase of Actinobacteria abundance in control soil was relatively higher than those in soil introduced with starch-based bioplastic, especially on day 90 (Ct90) and day 120 (Ct120). Firmicutes and Acidobacteria were relatively more abundant in soil introduced with starch-based bioplastic than in control soil. Cyanobacteria in the initial control soil (C0) is relatively abundant compared to all soil samples. Nanoarchaeota was detected in all samples (Fig. 2a), but it was more abundant on day 30 (Bt30) and 60 (Bt60) in soil with bioplastic than in the control soil and decreased on day 90 (Bt90) and 120 (Bt120).

Prevalent bacterial genera varied across all time points and some of these genera are known for their ability to degrade starch (Fig 2b). Various species belonging to *Klebsiella,* which was detected in abundance on day 30, are known to produce enzymes for starch degradation, such as α-cyclodextrin glycosyltransferase, α-amylase, and pullulanase [34]. *Longispora* was abundant on day 90 and 120. *Longispora fulfa* tested positive in starch hydrolysis assay [35]. Meanwhile, *Longispora, Bacillus,* and *Nitrospira* were abundant on day 120. Many *Bacillus* species are known to hydrolyze starch through the activity of α-amylase, an enzyme that cleaves the α-1,4 glycosidic bonds in starch [36, 37]. A study on functional genes among the tropical peat swamp bacterial community by Kanokratana et al. [38] indicated the prevalence of amylolytic genes belonging to *Nitrospira*, which indicates that members of this group play a role in starch degradation in the environment.

In contrast, there is a lack of evidence for starch degradation for some other bacterial genera that increased in abundance following the introduction of starch-based bioplastic. Past studies have shown that *Zobellela* and *Truepera* (day 30) play a role in the decomposition of high-carbohydrate organic materials. Maity et al. [39] demonstrated the use of *Zobellela tiwanensis* strain DD5 to produce polyhydroxy butyrate using the starch-rich banana peels as a substrate. Meanwhile, *Truepera* was reported to thrive in compost enrichment samples [40, 41]. On day 60, there was an increase of *Saccharimonadales* abundance. There is limited information on how *Saccharimonadales* may play a role in starch degradation. *Saccharimonadales* belongs to phylum Saccharibacteria, which is a member of superphylum Patescibacteria. Functional genome analysis showed that some members of Saccharibacteria were missing genes for de novo biosynthesis of essential amino acids, nucleotides, fatty acids, and cofactors [42], which indicated that they might require co-metabolism with other bacteria to survive in the environment. A member of Saccharibacteria isolated from wild oats rhizosphere showed that it feeds of plant exudates and its genome also indicates the prevalence of starch/glycogen and

trehalose breakdown gene for D-glucose production [43]. With the exception of *Longispora*, *Bacillus* and *Nitrospira*, all of the other genera that increased in abundance on day 120, including *Megamonas, Steroidobacter, Veillonella*, and *Bryobacter,* have not been associated with starch hydrolysis thus far. There is, however, an indication that they made indirect contribution to starch degradation and/or organic material decomposition in general. *Megamonas* is mainly found in human faeces and the human gut microbiome. In an *in vitro* pea starch digestion model, Cui et al. [44] reported that *Megamonas* was found in a large number after 8 hours of digestion. *Veillonella* thrives in the gut of gnotobiotic rats fed with amylomaize starch by utilizing starch degradation products derived by the amylolytic bacteria *Eubacterium* [45]. *V. atypica* was also reported to co-exist and communicate with *Streptococcus godonii* during the early formation of dental plaque biofilm [46]. This study showed that the presence of *V. atypica* increased the expression of the amylase-encoding gene *amyB* in *S. godonii. Bryobacter aggregatus* gen. nov., sp. nov., was reported to grow on starch, glucose, and maltose medium [47]. While *Bryobacter* have no reported amylase activity [47], it is possible that *Bryobacter* in soil with starch-based bioplastic used the starch degradation products such as maltose and glucose produced by starch degrading bacteria. Overall, this suggests that even though the above bacterial genera might not be involved in starch-based bioplastic degradation directly, they might thrive in soil by utilizing starch degradation intermediates or through other interactions within the bacterial community.

Bacterial community in compost soil introduced with oxo-degradable plastic

Similar to the observation for starch-based bioplastic treatment, dominant phyla in soil introduced with oxo-degradable plastic were Bacteroidota, Proteobacteria, and Actinobacteria. While there was an increase in Proteobacteria and Actinobacteria abundance, Bacteriodota abundance fluctuated over time (Fig. 3a). The introduction of oxo-degradable plastic affected non-dominant phyla within the bacterial community. The phyla that were increased in abundance following oxo-degradable plastic introduction were Patescibacteria, Planctomycetes, and Acidobacteria while a decrease was observed for Myxococcota and Firmicutes.

Several bacterial genera that may be associated with the degradation of the polyethylene backbone of oxo-degradable plastic emerged following its introduction in soil. *Serratia,* which was abundant on day 60 (Fig. 3b), was reported to degrade polyethylene [48, 49]. Faster degradation of polyethylene was achieved when the cell-free supernatant of *Serratia marcescens,* compared to the viable bacterial cells, was applied to the plastic film [48]. This showed that the bacterium produced extracellular enzyme(s) to degrade polyethylene.

Despite the lack of information on their direct contribution to polyethylene degradation, several genera that have been detected in polyethylene- or microplastic-rich environment also arose at various time points. *Saccharimonadales* (day 60) was detected in microplastic-contaminated soil [50]. Similarly, *Rheinheimera* (day 90) was reported in abundance in microplastic-infested water [51]. *Pantoea* (day 120) was found in the gut of the polyethylene-degrading *Galleria mellonella* and *Tenebrio molitor* larvae that were kept on polyethylene-rich diet [52, 53]. *Portibacter* (day 90 and 120) was identified among major

bacterial colonizers of polyethylene plastic debris [54]. Interestingly, as observed for starch-based bioplastic, no particular bacteria thrived continuously across all time points in soil introduced to oxo-degradable plastic. This indicate that both starch-based bioplastic and oxo-degradable plastic degradation require a multitude of bacteria that will continue to shift over time.

This study is focused on bacterial community profiles during the degradation of commercially available starch-based bioplastic and oxo-degradable plastic, yet this process may involve a plethora of other microorganisms such as fungi. Fungal diversity analysis was not included as the availability of database and non-bias universal primers are lacking for this group of microorganisms at the time this study was conducted. In the future, prolonged incubation up to the point that the plastics are fully degraded will provide a more in-depth view on microbial dynamics, particularly for the oxo-degradable plastic, which requires longer time to degrade.

Conclusion

Starch-based bioplastic degraded quicker than oxo-degradable plastic in soil. Our results indicated that the bacterial composition in soil changed over time with or without the introduction of either types of plastic. While the major bacterial phyla remained similar for all treatment in this study, the addition of both types of plastic led to a different shift in soil bacterial community. Various genera emerged at specific time points and none of them dominated the soil bacterial community continuously. They represent bacteria that might be directly involved in breaking down the plastic polymers, as well as those that survive by interacting with the degraders. Overall, this study suggests that the degradation of both starch-based bioplastic and oxo-degradable plastic involve a complex set of microbes that continues to change over time.

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Availability of data and materials

The datasets generated and/or analysed during the current study are available in the NCBI repository, accession number PRJNA803316.

Competing interests

The authors declare that they have no competing interests.

Funding

This study is supported by the 2021 Science and Technology Research Grant from Indonesia Toray Science Foundation.

Authors' contributions

WAT and TP designed the experiment. JAW was involved in data acquisition and analysis as well as manuscript preparation under the advisory of WAT, TP, and AY. All authors have read and approved the manuscript.

Acknowledgements

The authors would like to thank the Indonesian Toray Science Foundation for supporting this study.

References

1. Hopewell J, Dvorak R, Kosior E. Plastics recycling: Challenges and opportunities. Philosophical Transactions of the Royal Society B: Biological Sciences. 2009;364:2115–26.
2. Geyer R, Jambeck JR, Law KL. Production, use, and fate of all plastics ever made [Producción, uso y destino de todos los plásticos jamás fabricados]. Science Advances. 2017;3:e1700782.
3. Andrady AL. The plastic in microplastics: A review. Marine Pollution Bulletin. 2017;119:12–22.
4. Yang Y, Li Z, Yan C, Chadwick D, Jones DL, Liu E, et al. Kinetics of microplastic generation from different types of mulch films in agricultural soil. Science of The Total Environment. 2022;814:152572.
5. Li J, Qu X, Su L, Zhang W, Yang D, Kolandhasamy P, et al. Microplastics in mussels along the coastal waters of China. Environmental Pollution. 2016;214:177–84.
6. Smith M, Love DC, Rochman CM, Neff RA. Microplastics in Seafood and the Implications for Human Health. Current environmental health reports. 2018;5:375–86.
7. Abdullah AHD, Chalimah S, Primadona I, Hanantyo MHG. Physical and chemical properties of corn, cassava, and potato starchs. IOP Conference Series: Earth and Environmental Science. 2018;160.
8. Luengo JM, García B, Sandoval A, Naharro G, Olivera ER. Bioplastics from microorganisms. Current Opinion in Microbiology. 2003;6:251–60.
9. Abe MM, Branciforti MC, Brienzo M. Biodegradation of hemicellulose-cellulose-starch-based bioplastics and microbial polyesters. Recycling. 2021;6.
10. Koyama A, Wallenstein MD, Simpson RT, Moore JC. Carbon-Degrading Enzyme Activities Stimulated by Increased Nutrient Availability in Arctic Tundra Soils. PLOS ONE. 2013;8:null.
11. Maran JP, Sivakumar V, Thirugnanasambandham K, Sridhar R. Degradation behavior of biocomposites based on cassava starch buried under indoor soil conditions. Carbohydrate Polymers. 2014;101:20–8.

12. Thomas NL, Clarke J, McLauchlin AR, Patrick SG. Oxo-degradable plastics: Degradation, environmental impact and recycling. Proceedings of Institution of Civil Engineers: Waste and Resource Management. 2012;165:133–40.
13. Sebaa M, Servens C, Pouyet J. Natural and artificial weathering of low-density polyethylene (LDPE): Calorimetric analysis. Journal of Applied Polymer Science. 1993;47:1897–903.
14. Niu L, Li Y, Li Y, Hu Q, Wang C, Hu J, et al. New insights into the vertical distribution and microbial degradation of microplastics in urban river sediments. Water Research. 2021;188:116449.
15. O'Brine T, Thompson RC. Degradation of plastic carrier bags in the marine environment. Marine Pollution Bulletin. 2010;60:2279–83.
16. Klindworth A, Pruesse E, Schweer T, Peplies J, Quast C, Horn M, et al. Evaluation of general 16S ribosomal RNA gene PCR primers for classical and next-generation sequencing-based diversity studies. Nucleic acids research. 2013;41:e1.
17. Magoč T, Salzberg SL. FLASH: Fast length adjustment of short reads to improve genome assemblies. Bioinformatics. 2011;27:2957–63.
18. Bokulich NA, Subramanian S, Faith JJ, Gevers D, Gordon JI, Knight R, et al. Quality-filtering vastly improves diversity estimates from Illumina amplicon sequencing. Nature methods. 2013;10:57–9.
19. Caporaso JG, Kuczynski J, Stombaugh J, Bittinger K, Bushman FD, Costello EK, et al. QIIME allows analysis of high-throughput community sequencing data. Nature methods. 2010;7:335–6.
20. Edgar RC, Haas BJ, Clemente JC, Quince C, Knight R. UCHIME improves sensitivity and speed of chimera detection. Bioinformatics. 2011;27:2194–200.
21. Haas BJ, Gevers D, Earl AM, Feldgarden M, Ward D v, Giannoukos G, et al. Chimeric 16S rRNA sequence formation and detection in Sanger and 454-pyrosequenced PCR amplicons. Genome research. 2011;21:494–504.
22. Edgar RC. UPARSE: highly accurate OTU sequences from microbial amplicon reads. Nature Methods. 2013;10:996–8.
23. Altschul SF, Gish W, Miller W, Myers EW, Lipman DJ. Basic local alignment search tool. Journal of molecular biology. 1990;215:403–10.
24. Quast C, Pruesse E, Yilmaz P, Gerken J, Schweer T, Yarza P, et al. The SILVA ribosomal RNA gene database project: Improved data processing and web-based tools. Nucleic Acids Research. 2013;41:590–6.
25. Wang Q, Garrity GM, Tiedje JM, Cole JR. Naïve Bayesian classifier for rapid assignment of rRNA sequences into the new bacterial taxonomy. Applied and Environmental Microbiology. 2007;73:5261–7.
26. Edgar RC. MUSCLE: Multiple sequence alignment with high accuracy and high throughput. Nucleic Acids Research. 2004;32:1792–7.
27. Accinelli C, Saccà ML, Mencarelli M, Vicari A. Deterioration of bioplastic carrier bags in the environment and assessment of a new recycling alternative. Chemosphere. 2012;89:136–43.

28. Mantia FPL, Ascione L, Mistretta MC, Rapisarda M, Rizzarelli P. Comparative investigation on the soil burial degradation behaviour of polymer films for agriculture before and after photo-oxidation. Polymers. 2020;12.
29. German DP, Chacon SS, Allison SD. Substrate concentration and enzyme allocation can affect rates of microbial decomposition. Ecology. 2011;92:1471–80.
30. Liew KC, Khor LK. Effect of different ratios of bioplastic to newspaper pulp fibres on the weight loss of bioplastic pot. Journal of King Saud University - Engineering Sciences. 2015;27:137–41.
31. Martín-Closas L, Costa J, Cirujeda A, Aibar J, Zaragoza C, Pardo A, et al. Above-soil and in-soil degradation of oxo- and bio-degradable mulches: A qualitative approach. Soil Research. 2016;54:225–36.
32. Spellerberg IANF, Fedor PJ. Tribute To Claude Shannon (1916-2001). Global Ecology and Biogeography. 2003;:177–9.
33. Meng Q, Yang W, Men M, Bello A, Xu X, Xu B, et al. Microbial community succession and response to environmental variables during cow manure and corn straw composting. Frontiers in Microbiology. 2019;10 MAR:1–13.
34. Wöber G. Pullulanase is a characteristic of many Klebsiella species and functions in the degradation of starch. European Journal Of Applied Microbiology. 1976;3:71–80.
35. Shiratori-Takano H, Yamada K, Beppu T, Ueda K. Longispora fulva sp. nov., isolated from a forest soil, and emended description of the genus Longispora. International Journal of Systematic and Evolutionary Microbiology. 2011;61:804–9.
36. Konsula Z, Liakopoulou-Kyriakides M. Hydrolysis of starches by the action of an α-amylase from Bacillus subtilis. Process Biochemistry. 2004;39:1745–9.
37. Abd-Elhalem BT, El-Sawy M, Gamal RF, Abou-Taleb KA. Production of amylases from Bacillus amyloliquefaciens under submerged fermentation using some agro-industrial by-products. Annals of Agricultural Sciences. 2015;60:193–202.
38. Kanokratana P, Uengwetwanit T, Rattanachomsri U, Bunterngsook B, Nimchua T, Tangphatsornruang S, et al. Insights into the Phylogeny and Metabolic Potential of a Primary Tropical Peat Swamp Forest Microbial Community by Metagenomic Analysis. Microbial Ecology. 2011;61:518–28.
39. Maity S, Das S, Mohapatra S, Tripathi AD, Akthar J, Pati S, et al. Growth associated polyhydroxybutyrate production by the novel Zobellellae tiwanensis strain DD5 from banana peels under submerged fermentation. International Journal of Biological Macromolecules. 2020;153:461–9.
40. Eichorst SA, Joshua C, Sathitsuksanoh N, Singh S, Simmons BA, Singer SW. Substrate-specific development of thermophilic bacterial consortia by using chemically pretreated switchgrass. Applied and Environmental Microbiology. 2014;80:7423–32.
41. Heiss-Blanquet S, Fayolle-Guichard F, Lombard V, Hébert A, Coutinho PM, Groppi A, et al. Composting-like conditions are more efficient for enrichment and diversity of organisms containing cellulase-encoding genes than submerged cultures. PLoS ONE. 2016;11:1–22.

42. Lemos LN, Medeiros JD, Dini-Andreote F, Fernandes GR, Varani AM, Oliveira G, et al. Genomic signatures and co-occurrence patterns of the ultra-small Saccharimonadia (phylum CPR/Patescibacteria) suggest a symbiotic lifestyle. Molecular ecology. 2019;28:4259–71.
43. Starr EP, Shi S, Blazewicz SJ, Probst AJ, Herman DJ, Firestone MA, et al. Stable isotope informed genome-resolved metagenomics reveals that Saccharibacteria utilize microbially processed plant derived carbon. bioRxiv. 2017;:1–12.
44. Cui W, Ma Z, Li X, Hu X. Structural rearrangement of native and processed pea starches following simulated digestion in vitro and fermentation characteristics of their resistant starch residues using human fecal inoculum. International Journal of Biological Macromolecules. 2021;172:490–502.
45. Deiahaye EP, Bouchet B, Andrieux C, Raibaud P, Szylit O, Gallant DJ. Breakdown of amylomaize starch granules in gnotobiotic rats associated with four bacterial strains isolated from conventional rat microflora. Journal of the Science of Food and Agriculture. 1994;65:487–95.
46. Egland PG, Palmer RJ, Kolenbrander PE. Interspecies communication in Streptococcus gordonii-Veillonella atypica biofilms: Signaling in flow conditions requires juxtaposition. Proceedings of the National Academy of Sciences of the United States of America. 2004;101:16917–22.
47. Kulichevskaya IS, Suzina NE, Liesack W, Dedysh SN. Bryobacter aggregatus gen. nov., sp. nov., a peat-inhabiting, aerobic chemo-organotroph from subdivision 3 of the acidobacteria. International Journal of Systematic and Evolutionary Microbiology. 2010;60:301–6.
48. Azeko ST, Etuk-Udo GA, Odusanya OS, Malatesta K, Anuku N, Soboyejo WO. Biodegradation of Linear Low Density Polyethylene by Serratia marcescens subsp. marcescens and its Cell Free Extracts. Waste and Biomass Valorization. 2015;6:1047–57.
49. Azeko ST, Odusanya OS, Malatesta K, Anuku N, Soboyejo WO. Bacterial remediation of polyethylene by Serratia marcescens sub sp. marcescens and its supernatant. Advanced Materials Research. 2015;1132:238–51.
50. Feng X, Wang Q, Sun Y, Zhang S, Wang F. Microplastics change soil properties, heavy metal availability and bacterial community in a Pb-Zn-contaminated soil. Journal of Hazardous Materials. 2022;424:127364.
51. McCormick AR, Hoellein TJ, London MG, Hittie J, Scott JW, Kelly JJ. Microplastic in surface waters of urban rivers: Concentration, sources, and associated bacterial assemblages. Ecosphere. 2016;7.
52. Cassone BJ, Grove HC, Elebute O, Villanueva SMP, LeMoine CMR. Role of the intestinal microbiome in low-density polyethylene degradation by caterpillar larvae of the greater wax moth, Galleria mellonella. Proceedings of the Royal Society B: Biological Sciences. 2020;287:9–11.
53. Przemieniecki SW, Kosewska A, Ciesielski S, Kosewska O. Changes in the gut microbiome and enzymatic profile of Tenebrio molitor larvae biodegrading cellulose, polyethylene and polystyrene waste. Environmental Pollution. 2020;256:113265.
54. Cheng J, Jacquin J, Conan P, Pujo-Pay M, Barbe V, George M, et al. Relative Influence of Plastic Debris Size and Shape, Chemical Composition and Phytoplankton-Bacteria Interactions in Driving

Seawater Plastisphere Abundance, Diversity and Activity. Frontiers in Microbiology. 2021;11 January:1–17.

Figures

Figure 1

Starch-based bioplastic (blue) and oxo-degradable plastic (orange) weight reduction following their burial in compost soil over 120 days. Numbers in bracket represent weight decrease in percentage.

Figure 2

Abundance of bacterial phyla (a) and genus (b) in soil following burial of starch-based bioplastic (Bt) compared to control soil with no plastic addition (Ct). Numbers following sample codes indicate the length of treatment in days. Each bacterial phylum is represented by a specific color.

Figure 3

Abundance of bacterial phyla (a) and genus (b) in soil following burial of oxo-degradable plastic (Ot) compared to control soil with no plastic addition (Ct). Numbers following sample codes indicate the length of treatment in days. Each bacterial phylum is represented by a specific color.