Counter-intuitive enhancement of degradation of solid plastic through engineering of lowered enzyme binding to solid plastic

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Abstract

Degradation of solid polyethylene terephthalate (PET) by leaf branch compost cutinase (LCC) produces various PET-derived degradation intermediates (DIs), in addition to terephthalic acid (TPA), which is the recyclable terminal product of all PET degradation. Although DIs can also be converted into TPA, in solution, by LCC, the TPA that is obtained through enzymatic degradation of PET, in practice, is always contaminated by DIs. Here, we demonstrate that the primary reason for non-degradation of DIs into TPA in solution is the efficient binding of LCC onto the surface of solid PET. Although such binding enhances the degradation of solid PET, it depletes the surrounding solution of enzyme that could otherwise have converted DIs into TPA. To retain a sub-population of enzyme in solution that would mainly degrade DIs, we introduced mutations to reduce the hydrophobicity of areas surrounding LCC's active site, with the express intention of reducing LCC's binding to solid PET. Despite the consequent reduction in invasion and degradation of solid PET, overall levels of production of TPA were ~3.6-fold higher, due to the partitioning of enzyme between solid PET and the surrounding solution, and the consequent heightened production of TPA from DIs. Further, synergy between such mutated LCC (F125L/F243I LCC) and wild-type LCC resulted in even higher yields, and TPA of nearly ~100% purity.

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