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A semi-automated discovery pipeline for compostable plastic degrading enzymes

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PURPOSE OF THE ABSTRACT

Annual global plastic production has reached a staggering 360 – 400 million metric tonnes in recent years, of which 60% is estimated to be lost into the environment [1]. This growing plastic waste problem has triggered greater interest in compostable plastic alternatives such as polylactic acid (PLA) and polybutylene adiptate terephthalate (PBAT). Despite their renewable nature, compostable plastics such as PLA and PBAT can be relatively resistant to microbial degradation in soil, and do not degrade optimally in existing waste management solutions such industrial composting and anaerobic digestors [2]. Given these issues, we aim to identify novel plastic degrading microbes and enzymes which could be used as innocula or microbial consortia to improve compostable plastic degradation in industrial composting and anaerobic digestion systems.

In pursuit of this goal, we performed soil enrichment cultures in the presence of PLA and PBAT plastic films. From these we isolated 23 PLA/PBAT degrading microbes all of which were Bacilli bacteria, not previously shown to degrade plastics. To identify and characterise the enzymes involved, we developed an in silico and wet lab semi-automated pipeline for PLA and PBAT degrading enzyme discovery. This approach utilizes and integrates existing bioinformatic search tools such SignalP and HMMER to filter candidate genes by sequence-based searches and comparison with literature-characterised enzymes. Through this, we narrowed our search to 105 candidate genes from an initial pool of 108,000 genes across the 23 strains. These genes were then cloned and characterized in a high-throughput approach using emulsified agar plate screens.

We envision that this pipeline will help further our understanding of the PLA/PBAT degradation activity observed in our strain isolates and inform us of which strains could be useful to generate synthetic microbial consortia for enhanced PLA/PBAT degradation.

FIGURES

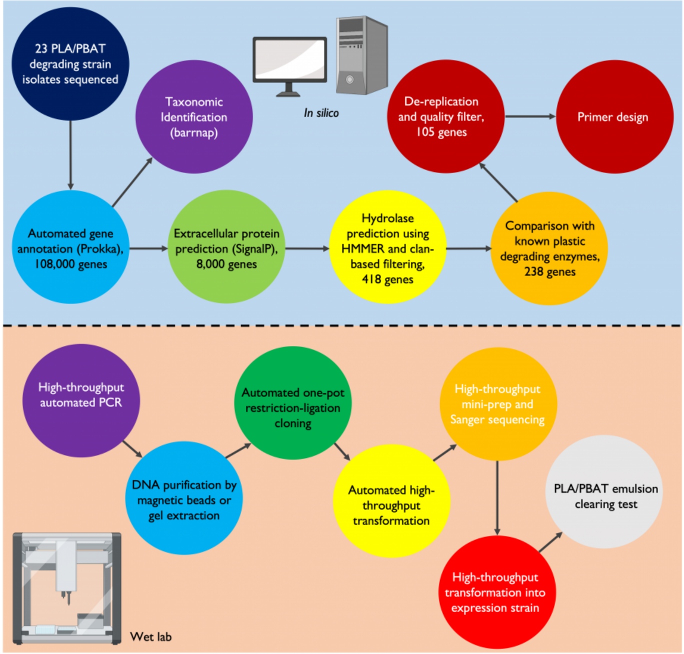

FIGURE 1

Figure 1

Schematic overview of the semi-automated discovery pipeline developed

FIGURE 2

KEYWORDS

Enzyme discovery and screening | Automation | Microbial Plastic degradation | Bioinformatics

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