Plastic Additives:
Understanding Threats to Human Health and Bioremediation Strategies

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Plastic additives in its ubiquity remain poorly managed and even less understood

•Plastic production is expected to **double** in the next 20 years
•Estimated 60-99 **million tons** of plastics worldwide were inappropriately discarded in 2015
•Natural removal of plastic takes decades to centuries
•It is unknown which plastic additives, in which polymers and concentration levels, induce toxic effects in organisms
•Public databases (e.g., IARC, EcoTox, IRIS) lack useful information on the toxicity of ~**90%** plastic additives

Objectives

The aim of our research team is two-fold:
Demystifying the health effects of additives
- **To test** the carcinogenic potential of selected additives with unknown classification
- **To identify** specific genes and pathways impacted by additives, clarifying how additives affect human health
- **To depict** relations between additives of known carcinogenicity and understudied, ill-documented additives
- **To underline** gaps between genes that are most studied and most salient to the toxic effects of additives

Validating lab-grown solutions
- **To test** newly identified enzymes capable of rapidly degrading plastic
- **To find** new plastic degrading enzymes testable in lab

Procedure

Building an expansive library of potentially carcinogenic additives	Demystifying additives with unknown toxic classification	Exploring new ways of degrading plastic
• Literature review • Analyze IRIS, IARC, and EcoTox databases for known carcinogenic plastic additives	• Experimental: • Dose Response Assays • Computational: • Enrichment analyses and clustering	• Experimental: • Validate Pseudonomas stutzeri • Computational: • Find new PET and MHET degrading enzymes

Additives and Human Health

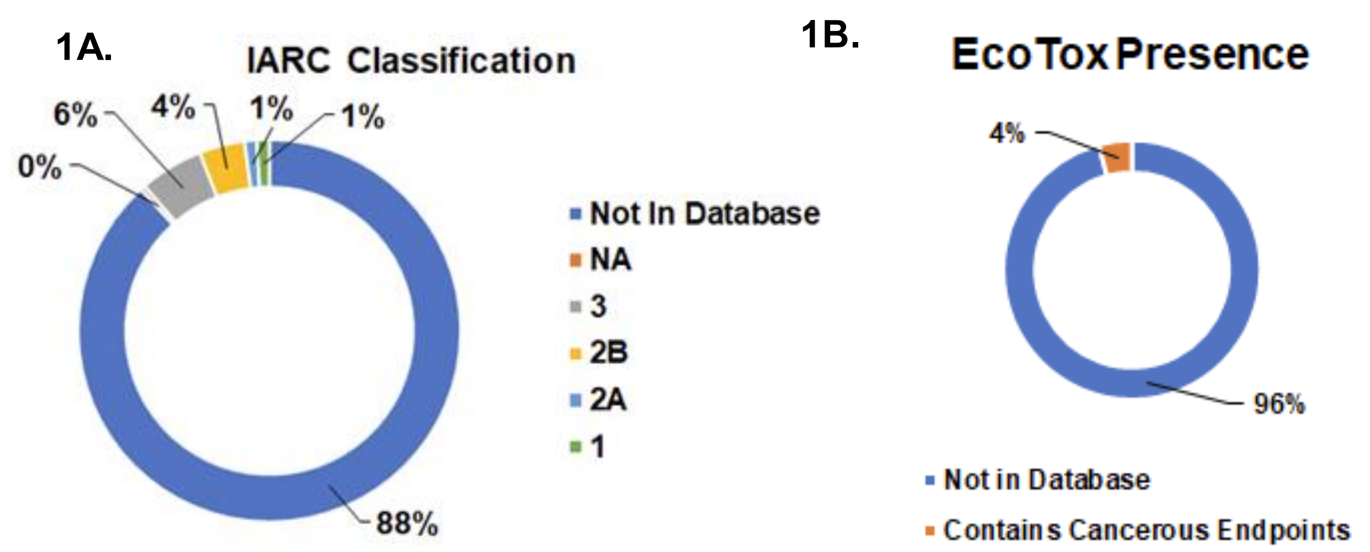

Figure 1. (A) Classifications of the 1322 plastic additives in the IARC database. *NA*: No Classification, *3*: Not classifiable as to its carcinogenicity to humans, *2B*: Possibly carcinogenic, *2A*: Probably carcinogenic, *1*: Carcinogenic. (B) Classifications of the 1322 plastic additives in the EcoTox database.

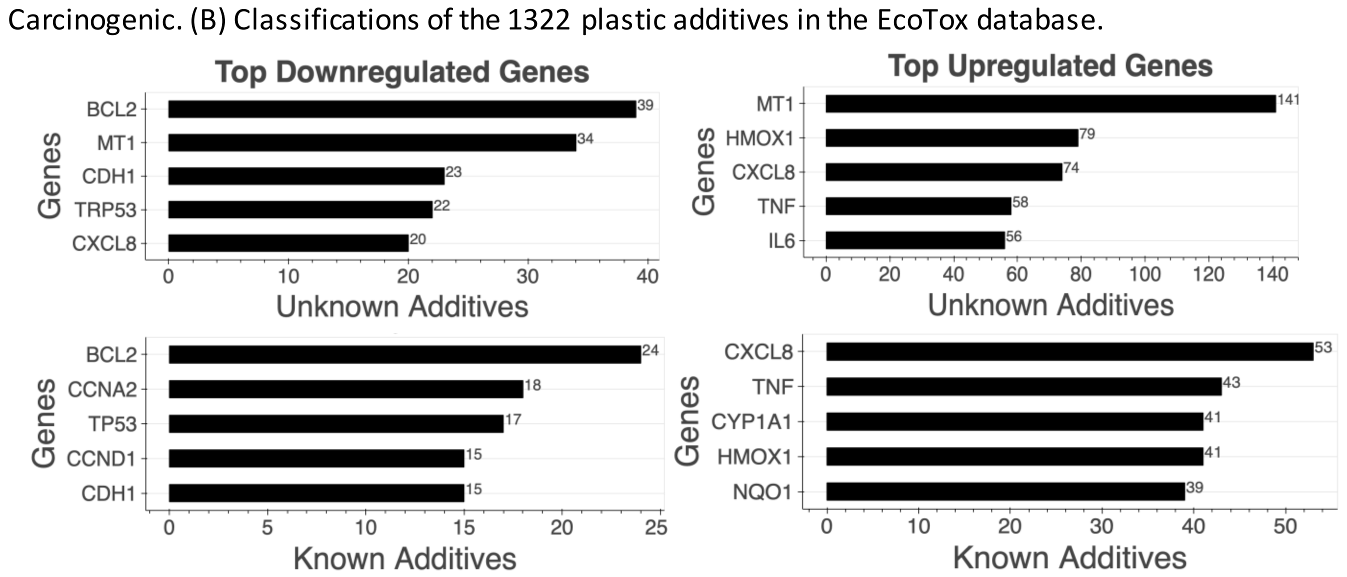

Figure 2. Comparison of top upregulated and downregulated genes in dataset of additives with unknown carcinogenic classification vs. additives with known carcinogenic classification.

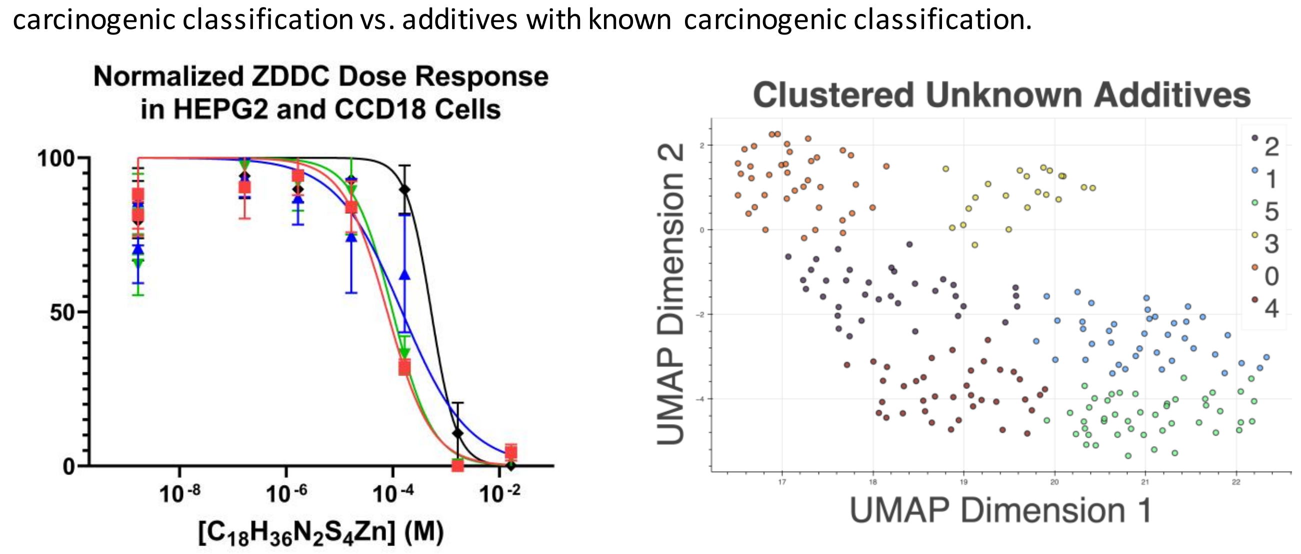

Figure 3. Experimental and computational methods for understanding the health effects of additives. (A) Normalized dose response curve in HEPG2 and CCD18 cells to zinc dibutyldithiocarbamate. (B) UMAP projection of unknown-toxicity additives dataset categorizes samples based on enriched biological pathways. Five resulting clusters are color-coded.

Conclusions

- Known plastic degrading enzymes have conserved sequence and structural elements that can be used to find new enzymes
- There is a severe lack of knowledge and publicity regarding plastic additive toxicity
- Certain groups of additives display similar gene expression patterns and fall into expression clusters with one another

Exploring Plastic Degrading Enzymes

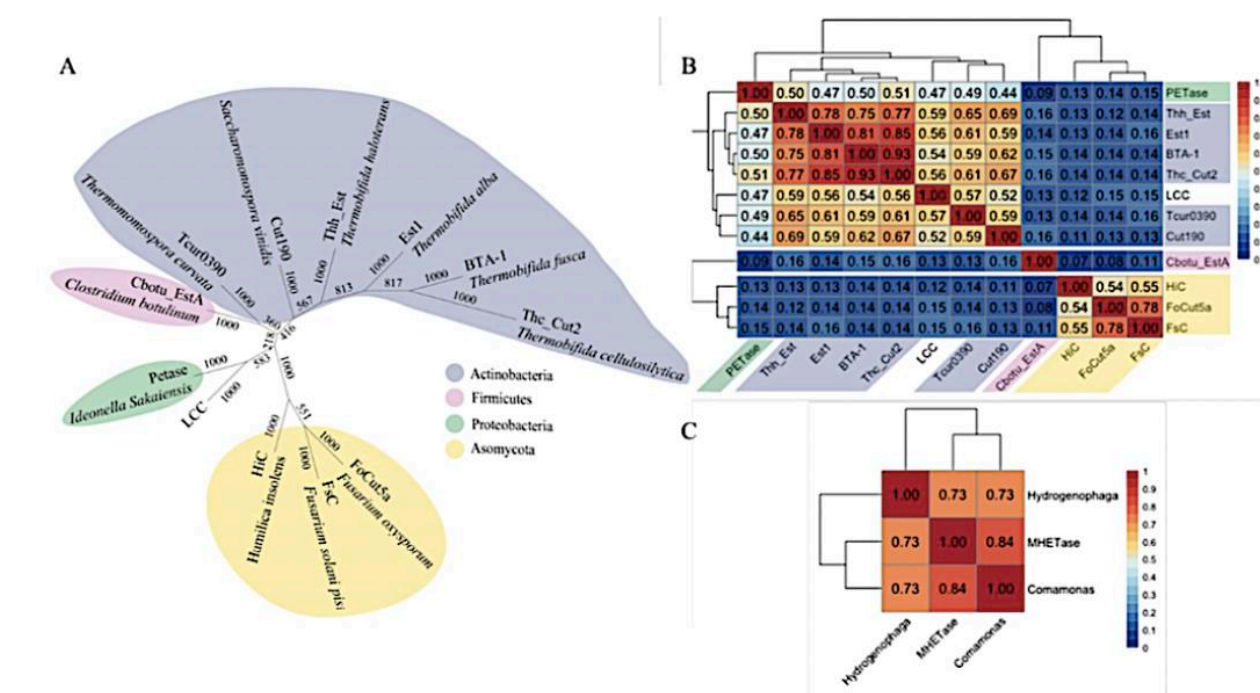

Figure 4. (A) Unrooted maximum likelihood phylogenetic tree of proven PET hydrolases. (B) Percent identity heat map of the experimentally validated PET hydrolases. (C) Percent identity heat map of the three MHET hydrolases.

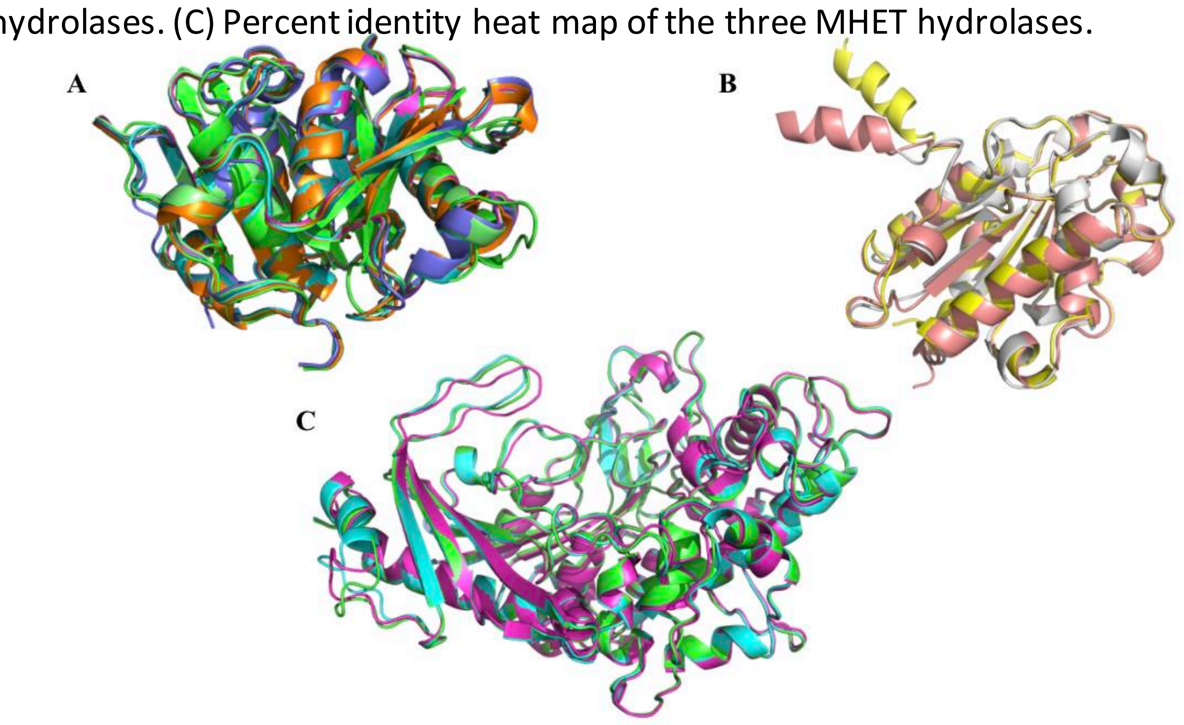

Figure 5. (A) Nine bacterial species and (B) three fungal species that are experimentally proven PET hydrolases superpositions. (C) Three experimentally proven MHET hydrolases superposition. Structures obtained with AlphaFold.

Next Steps

- Validate P. stutzeri as a plastic degrader in the lab using biological and chemical assays
- Verify additive toxicity in the lab
- Perform network analysis and predict net toxic effects from single polymers

References

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