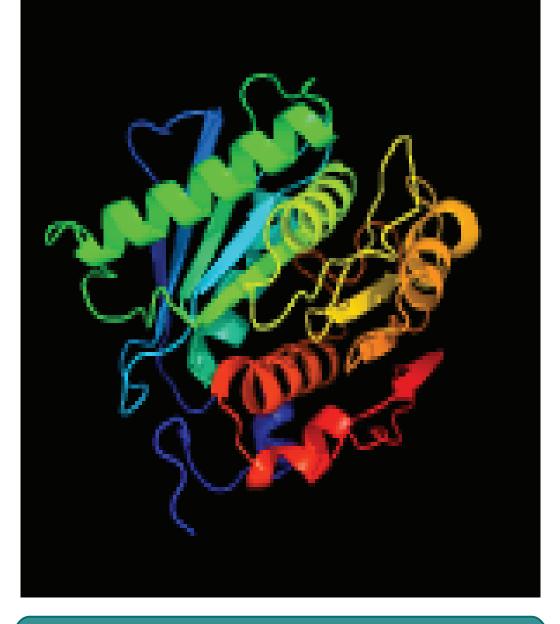


WeSTEM

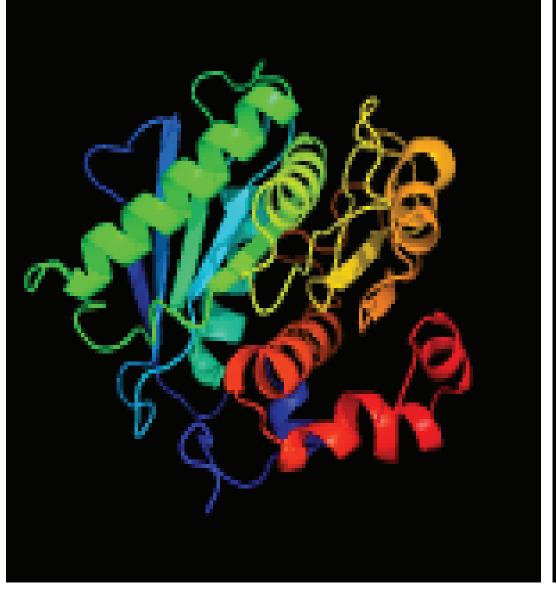
Polyethylene terephthalate (PET) is a clear plastic designed for single-use consumer packaging. PET plastics are 100% recyclable but only 31% of PETs are recycled^[1]. These durable plastics do not decompose for up to 450 years in disposal facilities, resulting in the clogging of natural landscapes with phthalate leaching litter. *Ideonella sakaiensis*, a bacteria discovered in 2016 in the sludge a plastic-recycling facility in Japan, exhibited successful decomposition and metabolization of PETs by binding to the surface of plastic consumer products and delivering enzymes to catabolize the plastic into its original structural components^[2]. The discovery of PETase in fungi, *Pestalotiopsis microspora*, by Yale in 2008 opened the door for new models of plastic degradation^[3]. By exploiting the lineage between fungi, endophytes, and marine plants with endophytic microbiomes, we could determine if natural PETase activity could be the solution to the hydrolysis of microplastics in our natural local wetland environment.

> Plastic surface

		18	0					
A0A0K8P6T7_Ideonella_sakaiensis		-	S	R	L	ΜQ) A (
E9LVH8_Thermobifida_cellulosilytica	-	-	S	R	L	SΑ	S	1
G9BY57_Unk.cutinase_homolog_leafbranch_metagenomics	_	_	s	R	L	sν	s '	i
D4Q9N1_Thermobifida_alba	-	-	s	R	L	GΑ	D	i
E9LVH9_T.cellulosilytica_PETH1_THECS	_	_	s	R	L	ΑV	/ M	1
F7IX06_T.alba_locusPETH2	_	_	s	R	L	ΑV	/ M	1
G8GER6_Thermobifida_fusca	D	s	s	R	L	ΑV	M N	1
Q48RJ7_T.fuscaYX	D	s	s	R	L	ΑV	/ M	i - 1
Q47RJ6_Nocardiopsacea_sp.	-	-	s	R	L	SΑ	S	i
Q6A0I4_T.fusca_sp.NRRL_B8184	-	-	s	R	L	SΑ	S	1
	E	Ν	D	S	Ι	ΑF	٧	
6ANE_B_I.sakaiensis_CHAINB	: Е	Ν	D	S	Ι	ΑF	٧	
6ANE_A_I.sakaiensis_CHAINA	E	Ν	D	s	Ι	ΑF	٧	
6EQH_C_Unc.Marinobacter	-	-	s	R	L	мç) A (
6EQH_B_Unc.Marinobacter	_	-	s	R	L	мç) A (
6EQH_A_Unc.Marinobacter	_	-	s	R	L	мç) A (
6EQG_C_PseudomonasCHAINC	_	-	s	R	L	мç) A (
6EQG_B_PseudomonasCHAINB						МÇ		
6EQG_A_PseudomonasCHAINA						МÇ	-	_
Alignment_Consensus						хx	-	



Ideonella sakaiensis (PETase)



Rhizobacter gummiphilis (PETase)

METHODS

Literature Search

Collect sequencing data from GenBank, a NCBI Database[5].

Alignment & Development

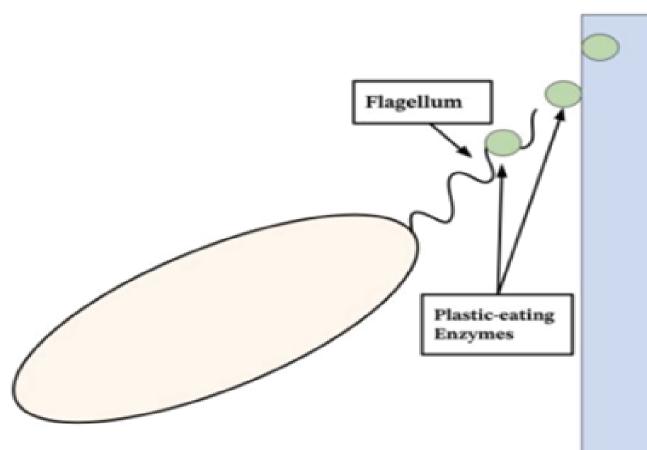
Test for regions of similarity using BLAST. Align sequences in Discovery Studio. Transform MSA into a phylogenetic tree using MegaX[4]. Determine major linkages. **3D Homology Models**

Use PHYRE2 to develop 3D homology models for folding comparison.

[1] Yoshida, S., Kazumi, H., Takehana, T., Taniguchi, I., Yamaji, H., Maeda, Y., Toy bacterium that degrades and assimilates poly(ethylene terephthalate). Science 351: 1196-[2] Hemani, G., Zheng, J., Wade, K.H., Laurin, C., Elsworth, B., Burgess, S., Bowder The MegaX platform supports systematic causal inference across the human phenome. el [3] Kelley, L., Mezulis, S., Yates, C. et al. (2015). The PHYRE2 web portal for protein

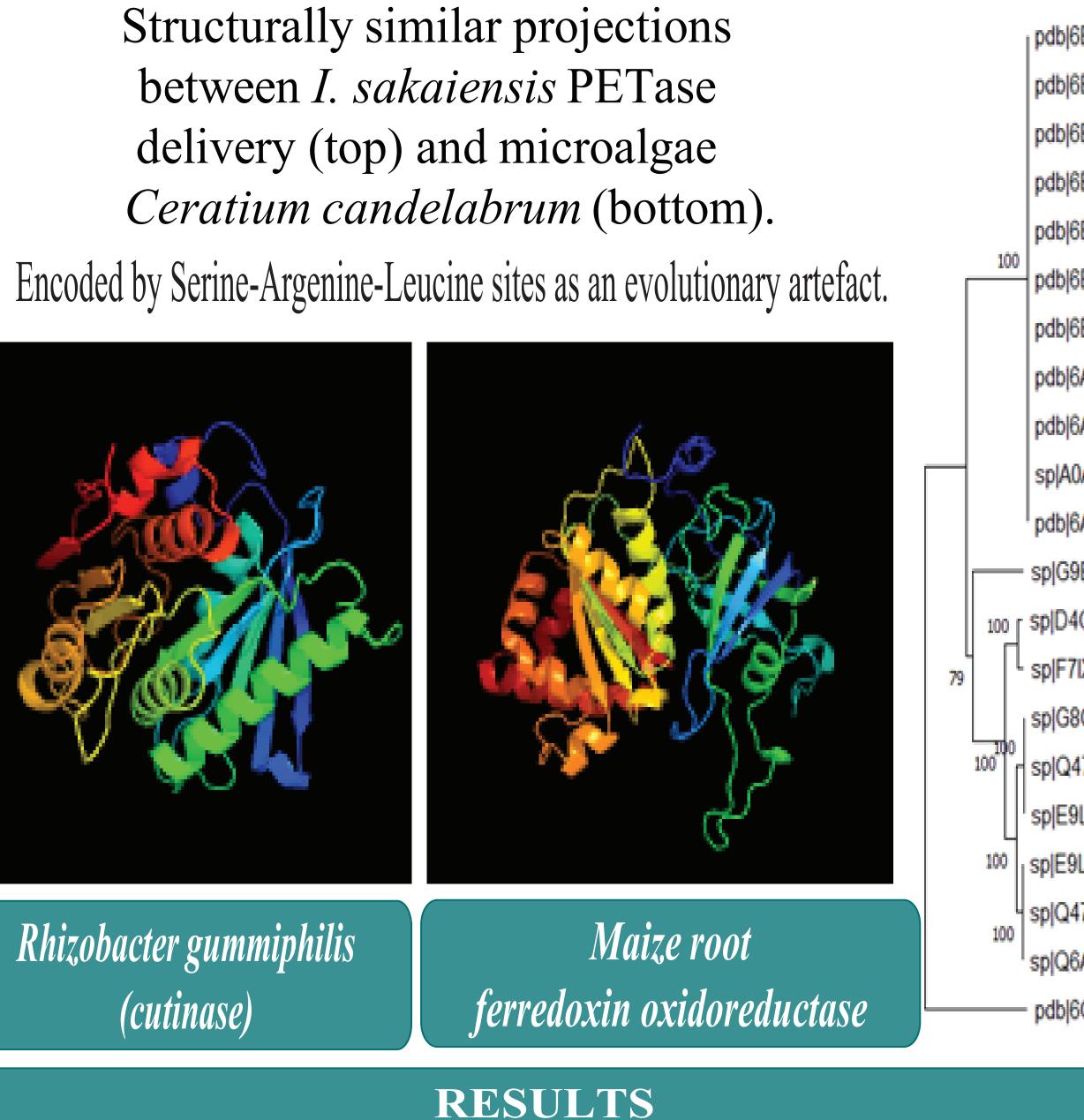
Evolutionary Analysis of Plastic-Degrading Enzyme Polyethylene Terephthalate Hydrolase (PETase) in the Endophytic Microbiome of Viridiplantae for Phytoremediation Marissa Mazariego & Michelle M. Barthet, Coastal Carolina University Department of Biology

INTRODUCTION





Carty, S. (2014). Freshwater Dinoflagellates of North America. Cornell University Press: 1 ed.



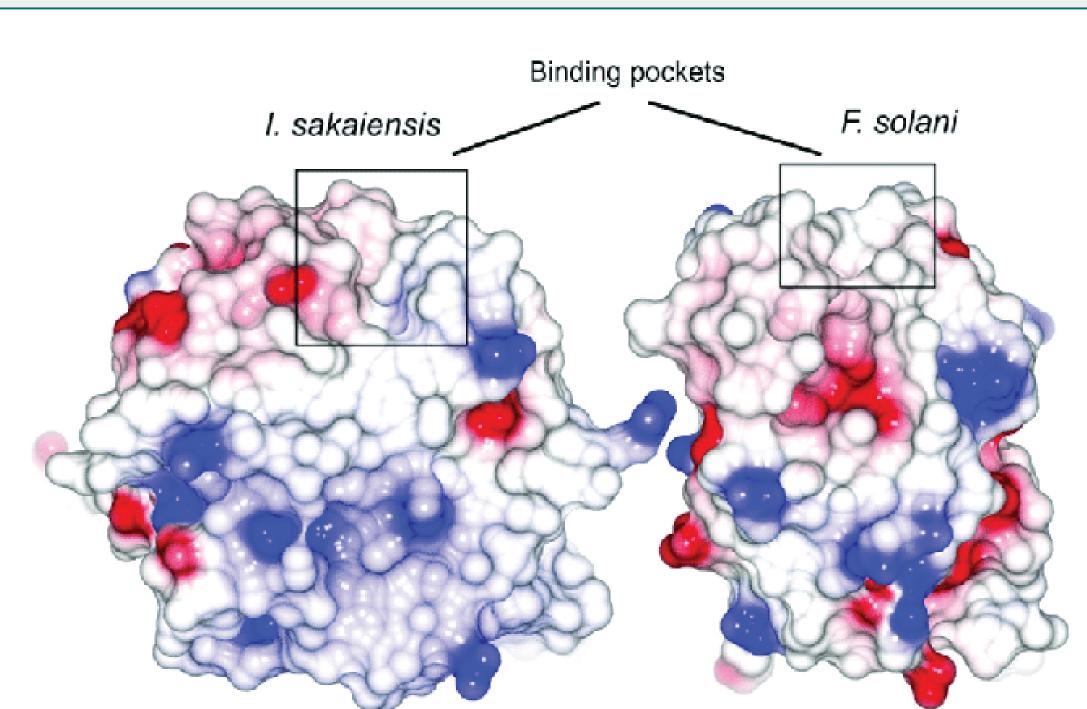
SRL Alignment as an Artefact of Microbial Evolution SRL, an amino acid sequence associated with "dinoflagellates", exploits the linkage between microalgae and bacterial enzyme delivery.

Bacterial to Fungi Linkage

Major active bacteria and fungi identified are: Aspergillus sp., Bacillus amyloliquefaciens, Nocardia sp., & Thermobifida sp.

SOU	SOURCES			
oyohara, K., Miyamoto, K. Kimura, Y., Oda, K 2016). A 5–1199. en, J., Langdon, R., Tan, V., Yarmolinsky, J., F, Hashem. (2018). eLife.	 [4] Wilm A.F., Dine Molecular Systems: [5] Sayers E.W., Bo Lanczycki C., Lath 			
modeling, prediction and analysis. Nature Protocols. 10: 845 858.	K.D., Sherry S.T (

pdb/6EQG/A Chain A Poly(ethylene terephthalate) hydrolase pdb|6EQF|A Chain A Poly(ethylene terephthalate) hydrolase pdb|6EQG|B Chain B Poly(ethylene terephthalate) hydrolase pdb|6EQG|C Chain C Poly(ethylene terephthalate) hydrolase pdb/6EQH/A Chain A Poly(ethylene terephthalate) hydrolase pdb/6EQH/B Chain B Poly(ethylene terephthalate) hydrolase pdb|6EQH|C Chain C Poly(ethylene terephthalate) hydrolase pdb/6ANE/A Chain A Poly(ethylene terephthalate) hydrolase pdb[6ANE|B Chain B Poly(ethylene terephthalate) hydrolase



H., Lihui, L., Changcheng, H., Liu, L., Yunzi, B., Rui. (2018). Protein Crystallography and Site-Direct Mutagenesis Analysis of the Poly(ethylene terephthalate) Hydrolase PETase from Ideonella sakaienss. ChemBioChem; 19: 10-1002.

sp|A0A0K8P6T7.1|PETH IDESA RecName: Full=Poly(ethylene terephthalate) hydrolase Short=PET hydrolase Short=PETase AltName: Full=PET-digesting enzyme Flags: Precursor pdb/6ANE/C Chain C Poly(ethylene terephthalate) hydrolase

sp[G9BY57.1]PETH UNKP RecName: Full=Leaf-branch compost cutinase Short=LC-cutinase Short=LCC AltName: Full=PET-digesting enzyme AltName: Full=Poly(ethylene terephthalate) hydrolase 100 [sp]D4Q9N1.2|PETH1 THEAE RecName: Full=Cutinase est1 AltName: Full=Poly(ethylene terephthalate) hydrolase Short=PET hydrolase Short=PETase Flags: Precursor L sp[F7IX06.1]PETH2 THEAE RecName: Full=Cutinase est2 AltName: Full=Poly(ethylene terephthalate) hydrolase Short=PET hydrolase Short=PETase AltName: Full=TaCut2 Flags: Precursor sp/G8GER6.1/PETH1 THEFU RecName: Full=Cutinase cut1 AltName: Full=Poly(ethylene terephthalate) hydrolase Short=PET hydrolase Short=PETase Flags: Precursor sp|Q47RJ7.1|PETH1 THEFY RecName: Full=Cutinase AltName: Full=Poly(ethylene terephthalate) hydrolase Short=PET hydrolase Short=PETase Flags: Precursor sp[E9LVH9.1]PETH2 THECS RecName: Full=Cutinase 2 AltName: Full=Poly(ethylene terephthalate) hydrolase Short=PET hydrolase Short=PETase ¹⁰⁰ sp[E9LVH8.1]PETH1 THECS RecName: Full=Cutinase 1 AltName: Full=Poly(ethylene terephthalate) hydrolase Short=PET hydrolase Short=PETase sp|Q47RJ6.1|PETH2 THEFY RecName: Full=Cutinase AltName: Full=Poly(ethylene terephthalate) hydrolase Short=PET hydrolase Short=PETase Flags: Precursor splQ6A0I4.1 [PETH2 THEFU RecName: Full=Cutinase cut2 Short=TfCut2 AltName: Full=Acetylxylan esterase AltName: Full=BTA-hydrolase 1 AltName: Full=Poly(ethylene terephthalate) hydrolase Sh pdb/6QZ4/B Chain B Mono(2-hydroxyethyl) terephthalate hydrolase

DISCUSSION

Limitations of Novel Enzyme Lack of information about sequencing made production of alignment difficult. **Artificial Manipulation** Majority of sequences in GenBank are lab-created strains of "unknown origin".

Better Together Looking Forward... extraction.

neen D.G., Gibson T.J., Karplus K., Li W., Lopez R., McWilliam H., Remmert M., Söding J., Thompson J.D., Higgins (2022). : Fast, scalable generation of high-quality protein multiple sequence alignments using Clustal Omega. Sievers Biology. 7: 539. Bolton E.E., Brister J.R., Canese K., Chan J., Comeau D.C., Connor R., Funk K., Kelly C., Kim S., Madej T., Marchler Bauer A., throp S., Lu Z., Thibaud-Nissen F., Murphy T., Phan L., Skripchenko Y., Tse T., Wang J., Williams R., Trawick B.W., Pruitt (2022). Database resources of the national center for biotechnology information. Nucleic Acids Res. 50: D20-D26.

Literature suggests endophytes cease degradation when supplemented with growth-promoting nutrients. Identifying organisms in local environments for sampling and DNA