Harnessing the catalytic potential of a ferulic acid esterase for MHET hydrolysis

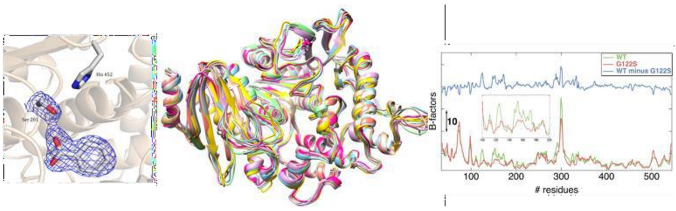
Panagiota Karampa[1], Konstantinos Makryniotis[2], Efstratios Nikolaivits[2], Evangelos Topakas[2], Vangelis Daskalakis[3], Maria Dimarogona[1]

[1]*Laboratory of Structural Biology and Biotechnology, Department of Chemical Engineering, University of Patras, Greece*
[2]*Industrial Biotechnology & Biocatalysis Group, Biotechnology Laboratory, School of Chemical Engineering, National Technical University of Athens, Greece*
[3]*Laboratory of Biomolecular Dynamics and Engineering, Department of Chemical Engineering, University of Patras, Greece*

E-mail: mdimarog@chemeng.upatras.gr

Keywords: plastic degradation, MHETase, feruloyl esterase, X-ray crystallography, docking

The enzymatic breakdown of plastics presents a promising approach to address their uncontrolled accumulation on Earth. Polyethylene terephthalate (PET) is a widely used polymer used in packaging, construction, and agriculture. Since 2000, numerous enzymes, capable of decomposing plastic, such as lipases and carboxyl ester hydrolases have been discovered: PETases target the polymer's ester bonds, producing mono-(2-hydroxyethyl) terephthalate (MHET) as the primary water-soluble degradation product and MHETases then cleave the ester bonds of MHET, yielding terephthalic acid (TPA) and ethylene glycol (EG). Ferulic acid esterases, and specifically those belonging to tannase-like family, are structural homologs of the well-studied bacterial MHETase from *Ideonella sakaiensis*, while their primary role in nature is to cleave the ester bonds between hydroxycinnamic acids and arabinose in the plant cell wall. We have previously demonstrated that *Fo*FaeC, a tannase-like feruloyl esterase, shows activity on PET oligomers as well as synergistic effect for PET degradation, when combined with PETases[1]. In the frame of the present work, an *Fo*FaeC variant, G122S, was created by structure-guided mutagenesis, in an effort to mimick MHETase active site. Compared to wild-type *Fo*FaeC, G122S variant exhibits increased catalytic activity against MHET. The crystallographic structure of both wild-type *Fo*FaeC and G122S variant were used for docking simulations aiming to acquire deeper understanding and interpretation of these biochemical findings. Shedding light on the structural determinants of PET-active enzymes will allow the production of robust biocatalysts for plastic degradation.



This research was implemented in the framework of H.F.R.I call "Basic research Financing (Horizontal support of all Sciences)" under the National Recovery and Resilience Plan "Greece 2.0" funded by the European Union – NextGenerationEU (H.F.R.I. Project Number: 15024).

ΕΛ.Ι.Δ.Ε.Κ. Ελληνικό Ίδρυμα Έρευνας & Καινοτομίας
Ελλάδα 2.0 ΕΘΝΙΚΟ ΣΧΕΔΙΟ ΑΝΑΚΑΜΨΗΣ ΚΑΙ ΑΝΘΕΚΤΙΚΟΤΗΤΑΣ
Με τη χρηματοδότηση της Ευρωπαϊκής Ένωσης NextGenerationEU

[1] G. Taxeidis, E. Nikolaivits, J. Nikodinovic-Runic and E. Topakas, "Mimicking the enzymatic plant cell wall hydrolysis mechanism for the degradation of polyethylene terephthalate" Environmental Pollution, vol. 356, 124347, 204