

Electronic Supplementary Information

Cutinase-Catalyzed Polyester-Polyurethane Degradation: Elucidation of the Hydrolysis Mechanism

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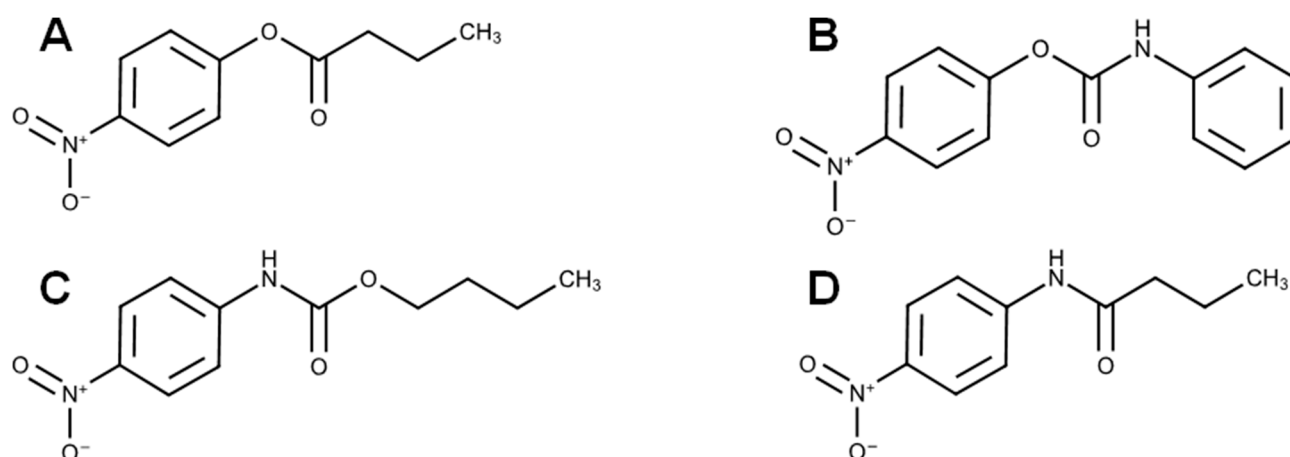


Figure S1. Model substrates used for the hydrolytic activity assay; **(A)** para-nitrophenyl-butyrates (pNPB); **(B)** para-nitrophenyl-N-benzylcarbamate (pNPC); **(C)** butyl-N(para-nitrophenyl)carbamate (pNPBC); **(D)** para-nitrobutyranilide (pNPA).

Table S1. Values of M_n , M_w and \bar{D} before and after enzymatic hydrolysis of PU as evaluated via GPC analysis.

	M_n	M_w	\bar{D}
T0	22000	107800	4.89
24 h	6400	90600	14.20
48 h	4100	74000	17.96
72 h	3400	62700	18.38
120 h	3600	64700	17.99
168 h	3500	62200	17.89

Table S2. Integrated values of the peaks corresponding to the carbon atoms linked to the ester bonds at the beginning and at the end of the incubation (all the peaks were manually integrated using the signal from the methylene group of the methylenedianiline as the reference).

δ (ppm)	2.3 ("j")	4.05 ("i")	4.15 ("g")
Blank	13.6429	12.4187	5.3357
T0	11.5995	10.8860	4.6157
24 h	7.7351	8.4851	3.0704
48 h	5.9597	7.3946	2.4300
72 h	4.7645	6.4077	1.9587
120 h	3.2922	5.5477	1.2741
168 h	5.2897	7.0965	2.1912

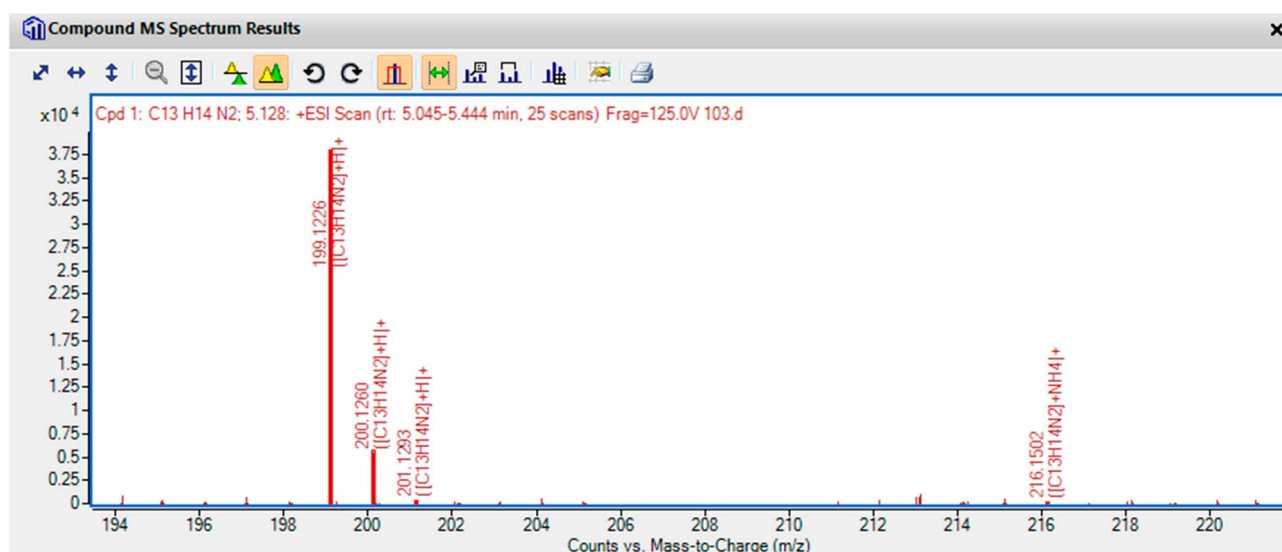


Figure S2. LC-TOF/MS analysis of the released product 3,3'-methylenedianiline, chemical formula: C₁₃H₁₄N₂. Exact mass: 198.12 Da.

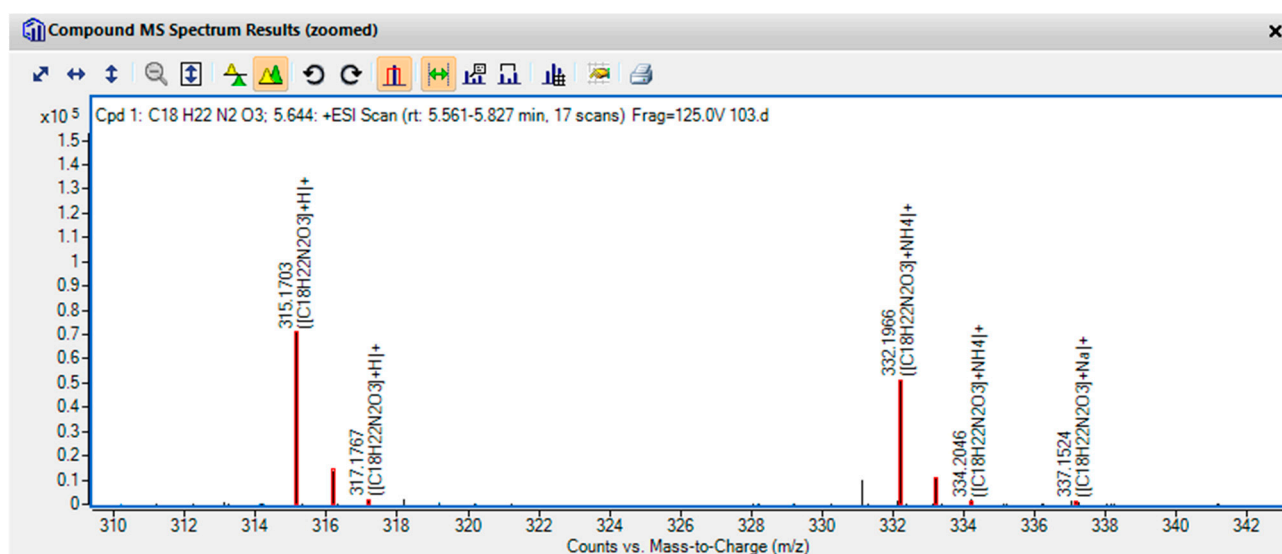


Figure S3. LC-TOF/MS analysis of the released product 4-hydroxybutyl (3-(3-aminobenzyl)phenyl)carbamate, chemical formula: C₁₈H₂₂N₂O₃. Exact mass: 314.16 Da.

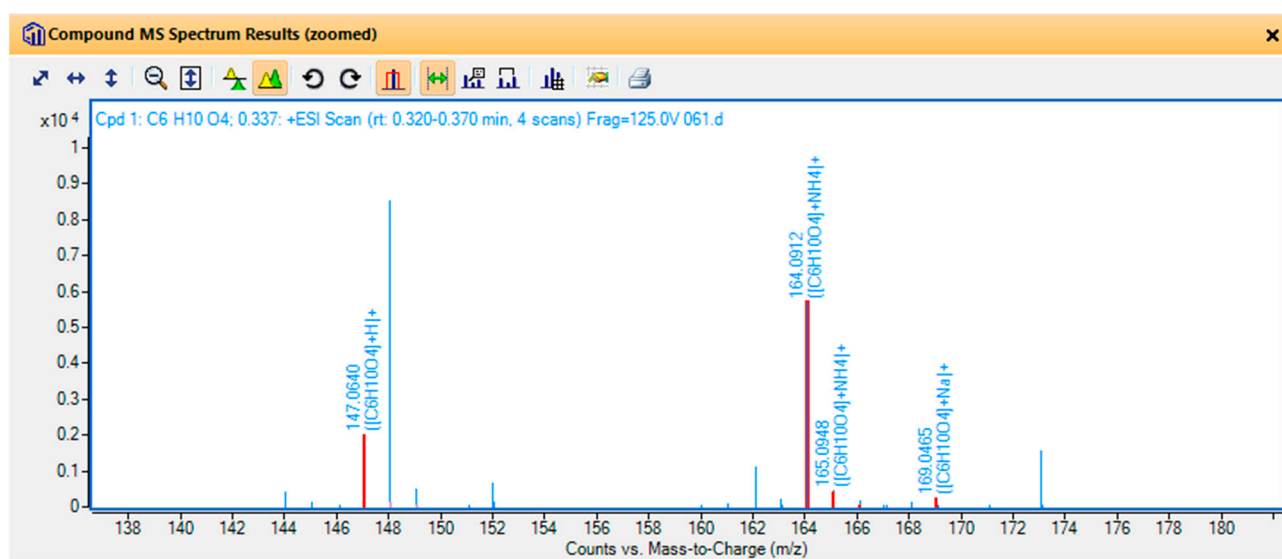


Figure S4. LC-TOF/MS analysis of the released product adipic acid, chemical formula: C₆H₁₀O₄. Exact mass: 146.06 Da.

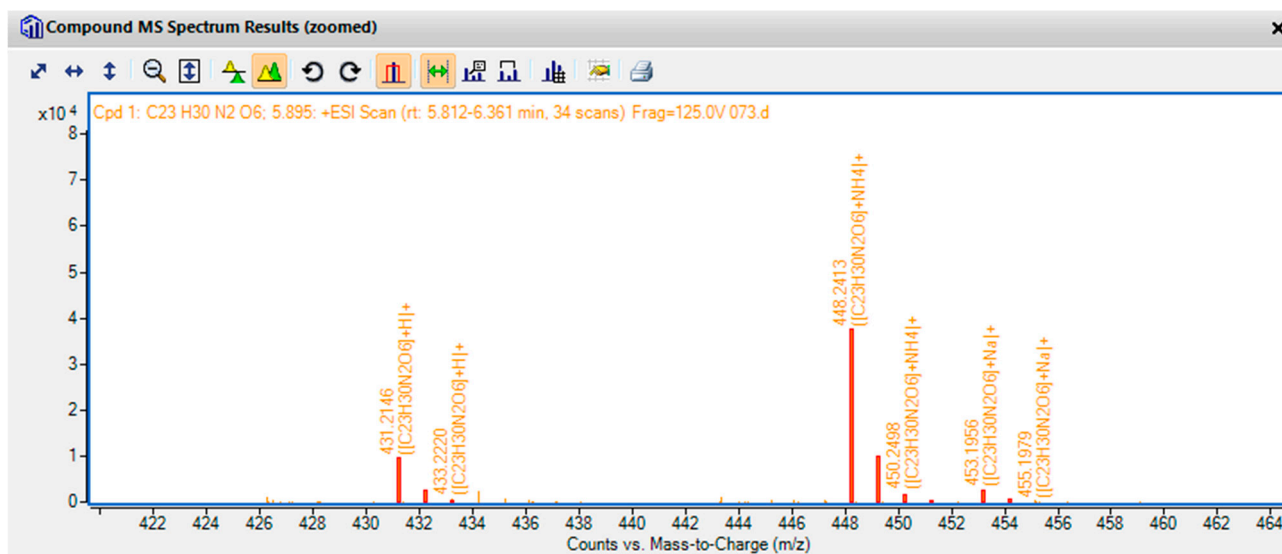


Figure S5. LC-TOF/MS analysis of the released product bis(4-hydroxybutyl) (methylenabis(3,1-phenylene))dicarbamate, chemical formula: C₂₃H₃₀N₂O₆. Exact mass: 430.21 Da.

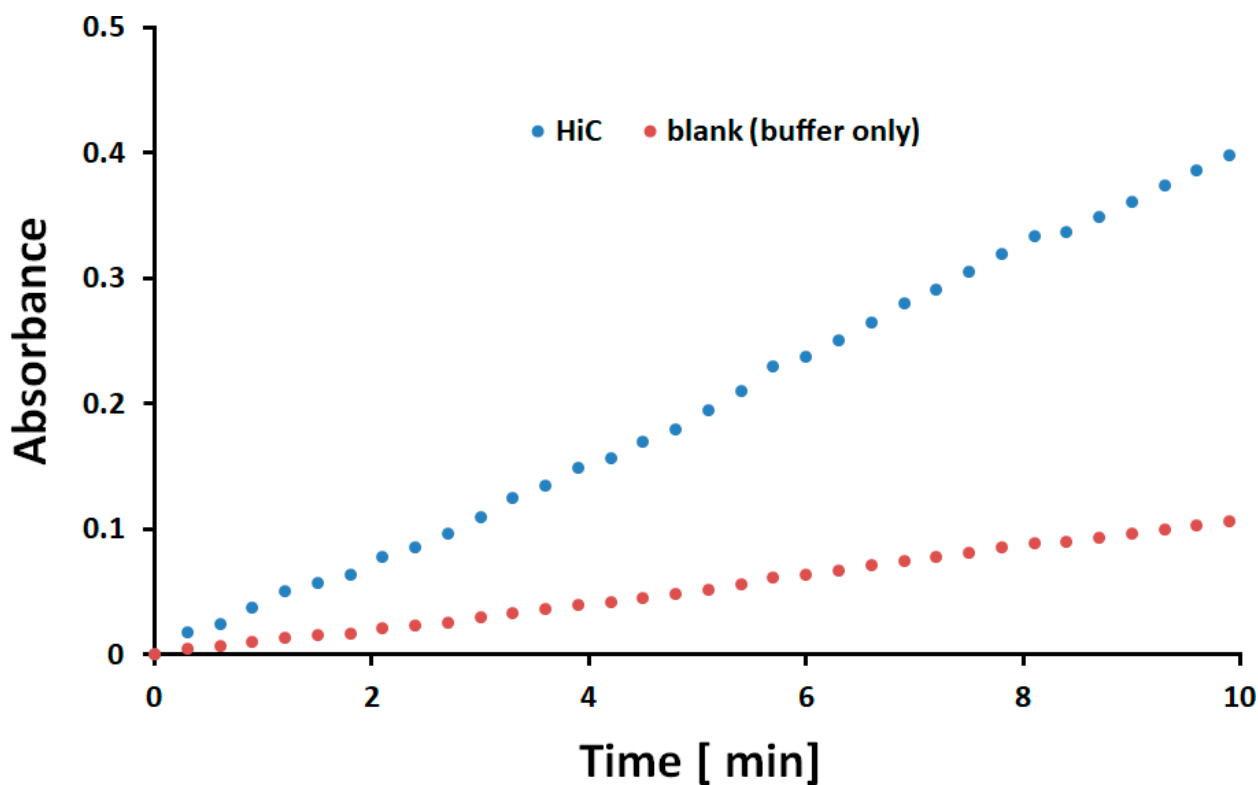


Figure S6. Activity of assay of HiC (blue dots) in presence of para-nitrophenyl-N-benzylcarbamate as substrate. The blank (red dot) was performed in presence of only buffer.

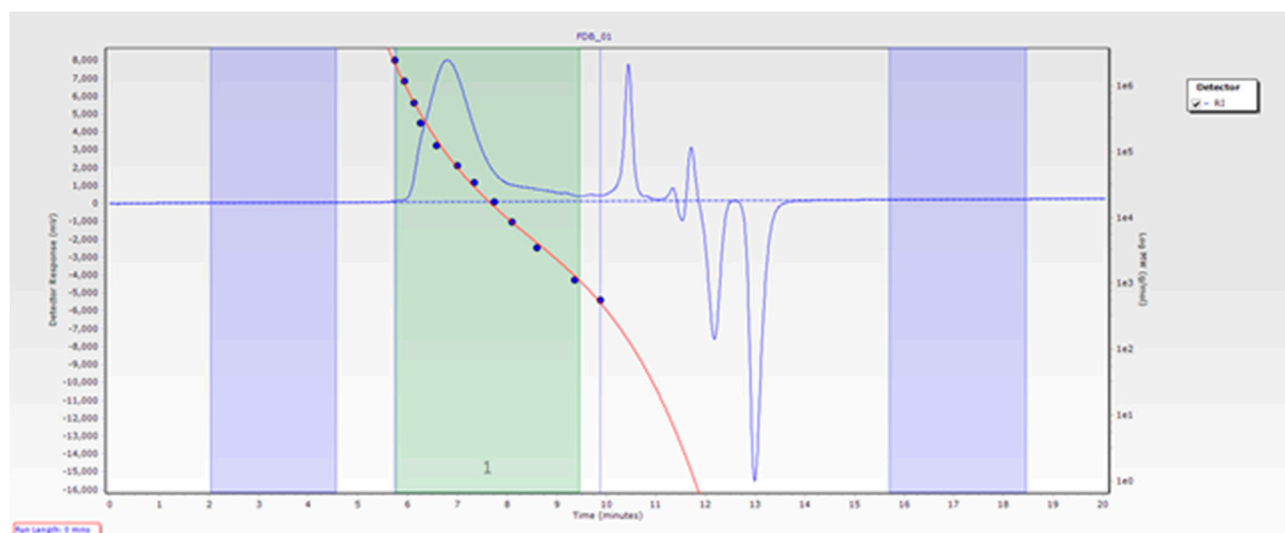


Figure S7. GPC chromatogram of the PU-PE film after 0h of incubation

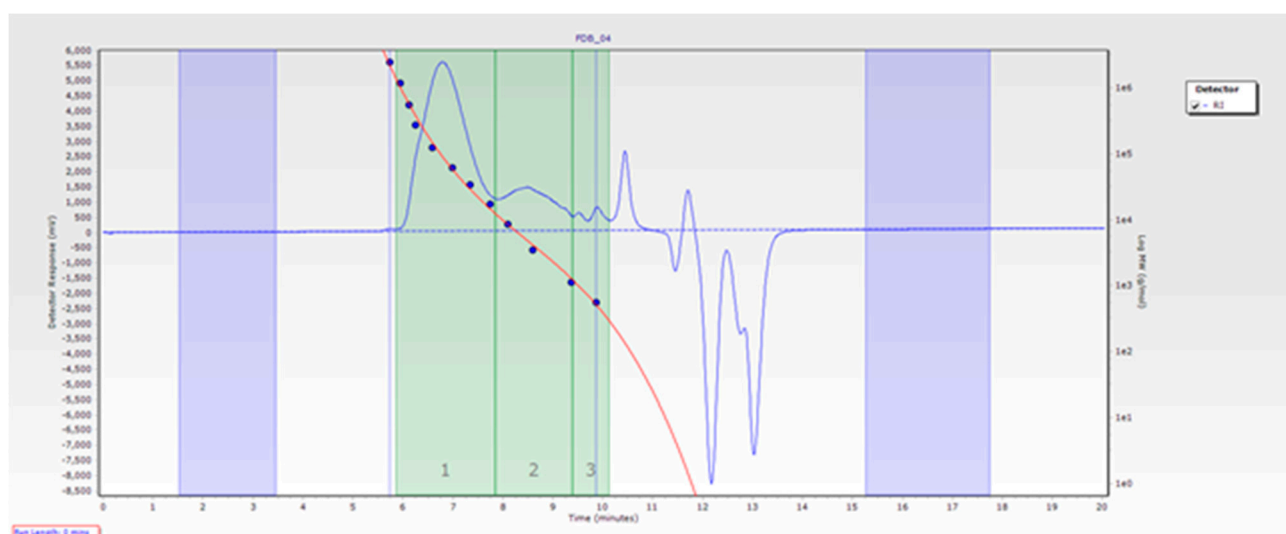


Figure S8. GPC chromatogram of the residual PU-PE film after 24h of incubation

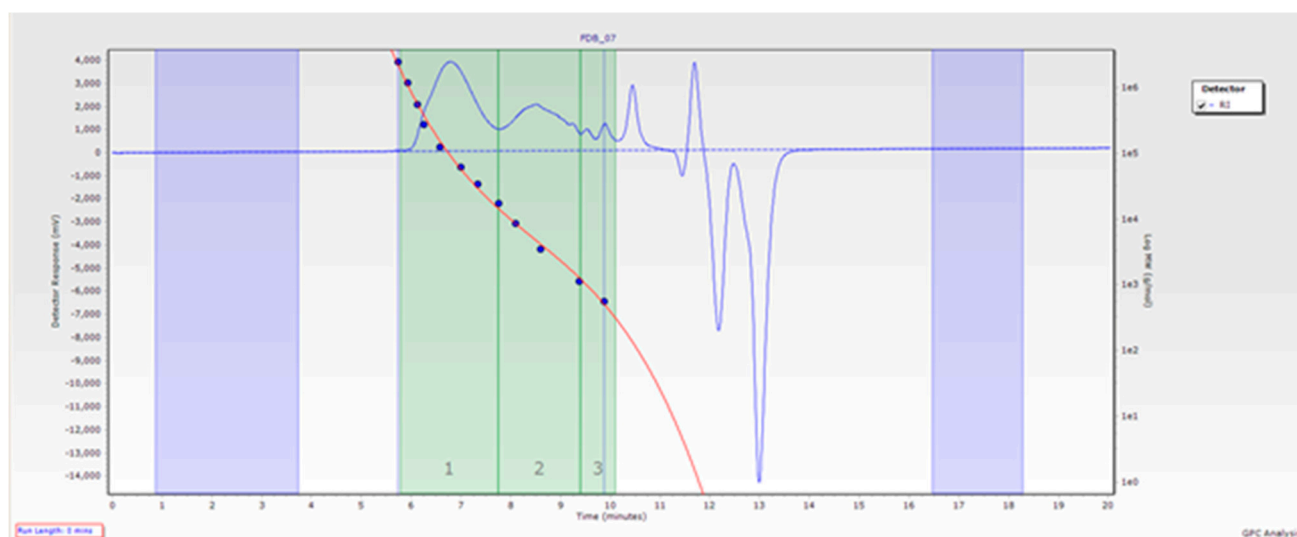


Figure S9. GPC chromatogram of the residual PU-PE film after 48h of incubation

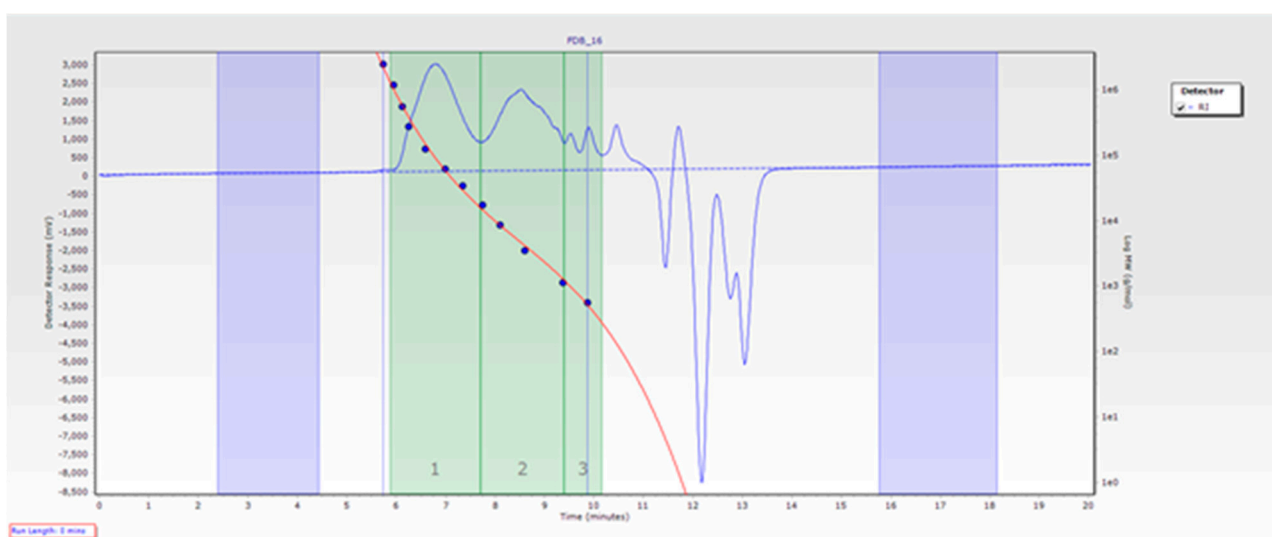


Figure S10. GPC chromatogram of the residual PU-PE film after 168h of incubation

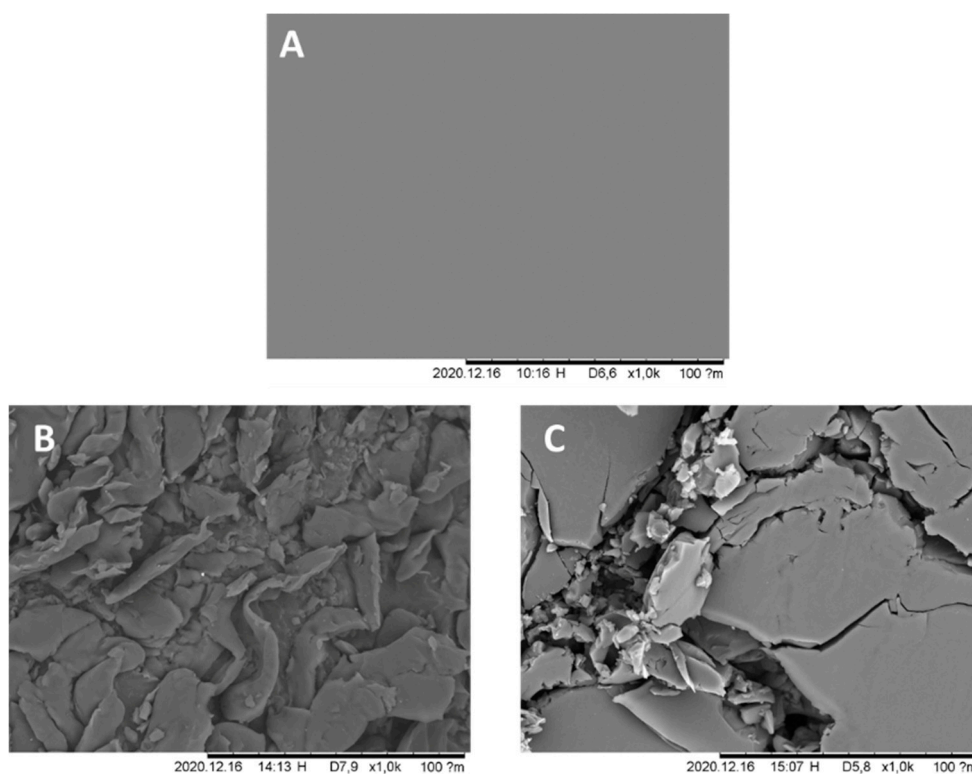


Figure S11. SEM images of the PU-PE films during enzymatic hydrolysis. (A) Blank; (B) 72 h of incubation; (C) 168 h of incubation. Magnification 1000x.