Electronic Supplementary Material (ESI) for Green Chemistry.
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Supplementary Information

Toward more Sustainable Enzyme Reactions: Enhancing Kinetics by Polydimethylacrylamide Implementation

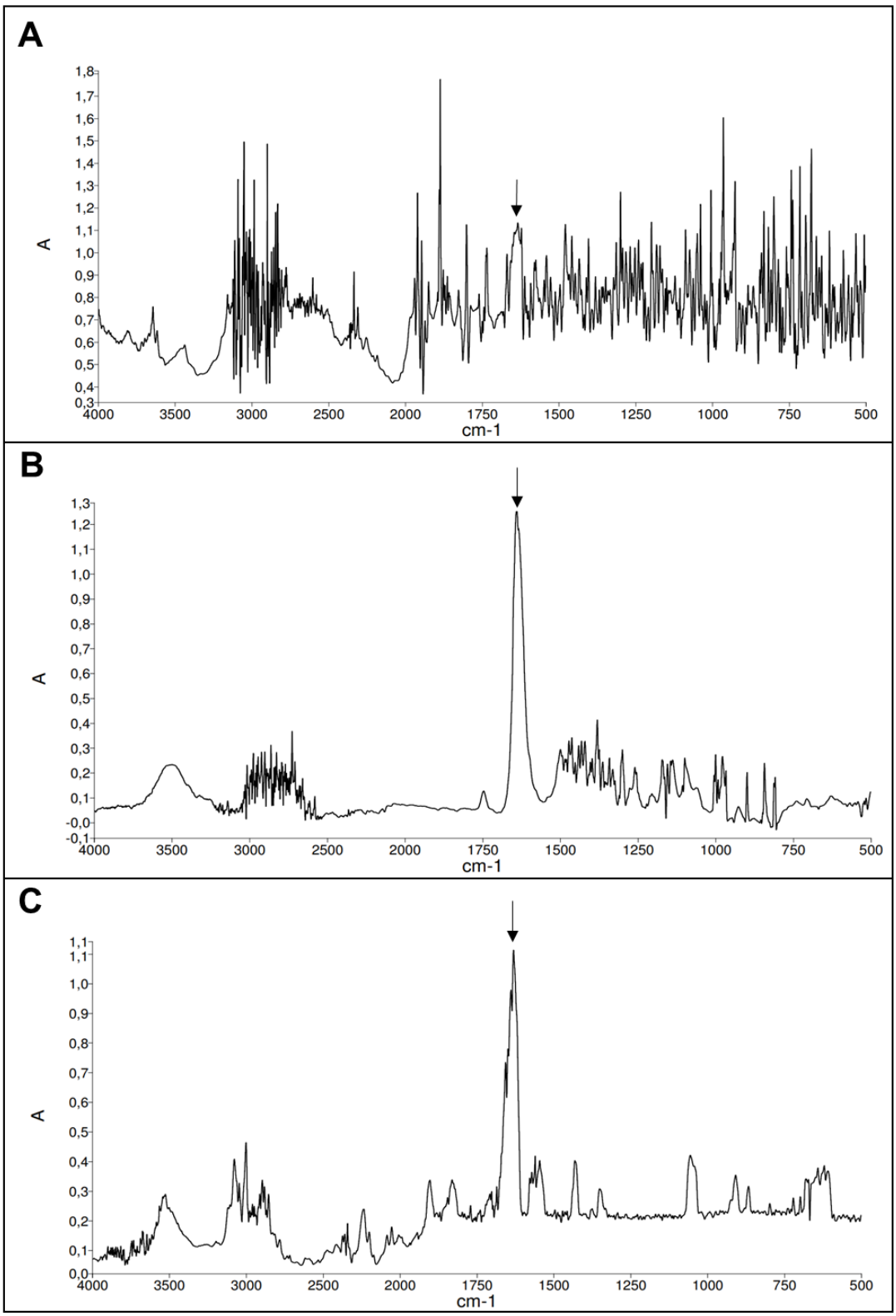

Figure S1 Recorded Fourier transform-infrared spectra of PDMA-functionalized microwell plates. *Spectra of the PDMA coatings on (A) polystyrene. (B) polypropylene and (C) polycarbonate were recorded in the range of 4000 – 400 cm^{-1}. The expected PDMA-amides can be seen clearly at 1650 cm^{-1}(relevant peak marked with arrows). In addition. the dominant peaks between 3000 – 3500 cm^{-1} can be assigned to the PDMA. Thus. the recorded Fourier transform-infrared spectra proved the presence of PDMA on the microtiter plates.*

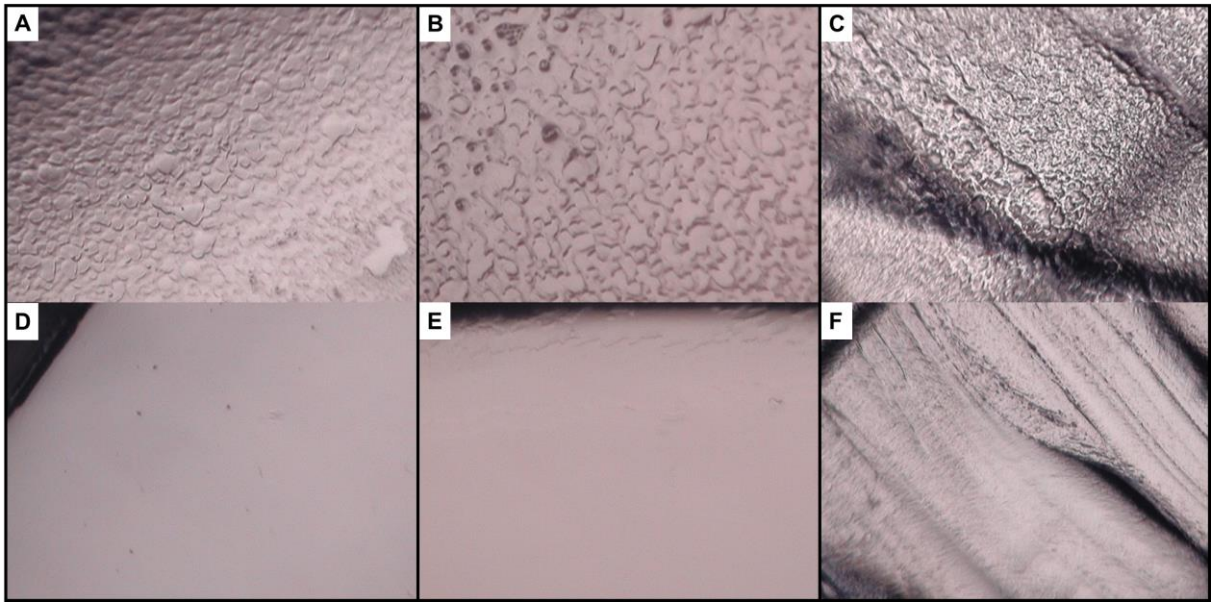

Figure S2 FTIR-Microscope images of PDMA functionalized microwell plates. *Images of (A) polystyrene (B) polypropylene and (C) polycarbonate functionalized with PDMA as well as non-functionalized (D) polystyrene. (E) polypropylene and (F) polycarbonate were caputred with a Spotlight 400 FT-IR microscope (PerkinElmer. Waltham. USA). Plane surfaces can be seen for unfunctionalized PS (D) and PP (E) and a slightly structured surface for unfunctionalized PC (F) originating from the 3D printing. The functionalization of the different polymers with PDMA results in smooth, wavy textured surfaces.*

Table S1 Nitrocefin substrate turnover (µM) of TEM-1 β-lactamase *with varied concentrations of nitrocefin substrate after 60 minutes incubation. Experiments were performed in polystyrene wells functionalized with PDMA and non-functionalized (control). Substrate turnover was measured with a TECAN Infinite 200 Pro at 492 nm. The amounts of substrates converted by the enzyme were determined from the measured absorbance values and the corresponding extinction coefficient of 17,400 M^{-1} cm^{-1} Each data point represents the mean ± SD of three different experiments.*

Nitrocefin [µM]					
	PDMA		control		
	Mean	SD	Mean	SD	p-value
750	349.29	6.02	195.56	15.59	0.0061
500	316.88	7.33	176.38	3.44	0.0019
250	224.27	9.48	102.38	3.88	0.0021
125	104.23	1.95	52.59	3.09	0.0002
62.5	50.04	1.42	33.49	1.11	0.0022
31.25	25.27	0.40	18.20	0.52	0.0022
0	0.00	0.00	0.00	0.00	

Table S2 TMB substrate turnover (µM) of HRP *with varied concentrations of TMB substrate after 60 minutes incubation. Experiments were performed in polystyrene wells functionalized with PDMA and non-functionalized (control). Substrate turnover was measured with a TECAN Infinite 200 Pro at 650 nm. The amounts of substrates converted by the enzyme were determined from the measured absorbance values and the corresponding extinction coefficient of 39,000 M^{-1} cm^{-1} Each data point represents the mean ± SD of three different experiments.*

TMB [µM]					
	PDMA		control		
	Mean	SD	Mean	SD	p-value
117.9	27.09	0.37	19.73	0.85	0.0067
99.46	26.99	0.13	20.95	2.33	0.0456
88.69	21.64	1.22	21.74	0.26	0.8906
81.58	21.71	1.27	21.16	0.59	0.5967
74.48	19.13	0.14	19.56	0.57	0.3848
67.375	18.94	0.70	19.77	0.58	0.1445

Table S3 PNPP substrate turnover (µM) of ALP *with varied concentrations of pNPP substrate after 60 minutes incubation. Experiments were performed in polystyrene wells functionalized with PDMA and non-functionalized (control). Substrate turnover was measured with a TECAN Infinite 200 Pro at 405 nm. The amounts of substrates converted by the enzyme were determined from the measured absorbance values and the corresponding extinction coefficient of 18,000 M^{-1} cm^{-1} Each data point represents the mean ± SD of three different experiments.*

pNPP [µM]					
	PDMA		control		
	Mean	SD	Mean	SD	p-value
1,000	140.82	18.68	46.25	9.40	0.0279
500	67.55	10.13	22.33	1.62	0.0205
250	37.28	2.67	17.39	0.80	0.0100
125	22.81	0.99	12.77	0.47	0.0069
100	20.29	1.00	11.85	0.20	0.0030
62.5	17.31	1.30	12.54	0.87	0.0391
0	0.00	0.00	0.00	0.00	

Table S4 Absorbance values of nitrocefin substrate turnover by TEM-1 β-lactamase *in polystyrene microtiter plate wells functionalized with different amounts of PDMA. Substrate turnover was measured with a TECAN Infinite 200 Pro at 492 nm. Each data point represents the mean ± SD of three different experiments.*

PDMA [µg]	Mean	SD
400	0.840	0.018
350	0.828	0.007
300	0.761	0.037
250	0.767	0.016
200	0.741	0.016
150	0.666	0.021
100	0.609	0.079
50	0.444	0.006
0	0.379	0.007

Table S5 Absorbance values of TMB substrate turnover by HRP *in polystyrene microtiter plate wells functionalized with different amounts of PDMA. Substrate turnover was measured with a TECAN Infinite 200 Pro at 650 nm. Each data point represents the mean ± SD of three different experiments.*

PDMA [µg]	Mean	SD
800	1.416	0.029
700	1.376	0.020
600	1.354	0.035
500	1.339	0.013
400	1.303	0.024
300	1.316	0.024
200	1.267	0.006
100	1.226	0.029
0	1.069	0.004

Table S6 Absorbance values of pNPP substrate turnover by ALP *in polystyrene microtiter plate wells functionalized with different amounts of PDMA. Substrate turnover was measured with a TECAN Infinite 200 Pro at 405 nm. Each data point represents the mean ± SD of three different experiments.*

PDMA [µg]	Mean	SD
400	0.639	0.042
350	0.619	0.063
300	0.530	0.072
250	0.505	0.075
200	0.450	0.031
150	0.344	0.053
100	0.256	0.043
50	0.183	0.053
25	0.181	0.014
12.5	0.193	0.023
0	0.158	0.013

Table S7 Initial reaction rates of nitrocefin substrate turnover by TEM-1 β-lactamase *in polystyrene microtiter plate wells. Substrate turnover was monitored as absorbance increase with a TECAN Infinite 200 Pro at 492 nm in 10 second intervals. Initial reaction rates were calculated from the linear range of substrate turnover at different substrate concentrations. Each data point represents the mean ± SD of three different experiments.*

Nitrocefin [µM]	V_0				
	PDMA on PS		control		
	Mean	SD	Mean	SD	p-value
600	3.308	0.074	1.753	0.220	0.0108
400	3.418	0.037	1.733	0.037	0.0006
200	3.101	0.016	1.319	0.032	0.0002
100	2.240	0.032	0.953	0.072	0.0006
50	1.402	0.072	0.756	0.047	0.0030
25	0.911	0.018	0.415	0.007	0.0005
0	0.000	0.000	0.000	0.000	

Table S8 Initial reaction rates of TMB substrate turnover by HRP *in polystyrene microtiter plate wells. Substrate turnover was monitored as absorbance increase with a TECAN Infinite 200 Pro at 650 nm in 15 second intervals. Initial reaction rates were calculated from the linear range of substrate turnover at different substrate concentrations. Each data point represents the mean ± SD of three different experiments.*

TMB [µM]	V_0				
	PDMA on PS		control		
	Mean	SD	Mean	SD	p-value
117.9	0.952	0.005	0.670	0.038	0.0059
99.46	0.861	0.007	0.674	0.057	0.0270
88.69	0.697	0.039	0.686	0.004	0.6796
81.58	0.668	0.029	0.643	0.015	0.3232
74.48	0.572	0.016	0.574	0.014	0.9104
67.375	0.553	0.025	0.561	0.017	0.7522

Table S9 Initial reaction rates of pNPP substrate turnover by ALP *in polystyrene microtiter plate wells. Substrate turnover was monitored as absorbance increase with a TECAN Infinite 200 Pro at 405 nm in 10 second intervals. Initial reaction rates were calculated from the linear range of substrate turnover at different substrate concentrations. Each data point represents the mean ± SD of three different experiments.*

pNPP [µM]	V_0				
	PDMA on PS		control		
	Mean	SD	Mean	SD	p-value
500	0.229	0.021	0.068	0.005	0.0085
250	0.140	0.007	0.051	0.003	0.0040
125	0.081	0.002	0.018	0.005	0.0041
100	0.063	0.007	0.012	0.002	0.0104
62.5	0.044	0.006	0.016	0.002	0.0105
0	0.000	0.000	0.000	0.000	

Table S10 Nitrocefin substrate turnover (µM) of TEM-1 β-lactamase *with varied concentrations of nitrocefin substrate after 60 minutes incubation. Experiments were performed in polypropylene wells functionalized with PDMA and non-functionalized (control). Substrate turnover was measured with a TECAN Infinite 200 Pro at 492 nm. The amounts of substrates converted by the enzyme were determined from the measured absorbance values and the corresponding extinction coefficient of 17,400 M^{-1} cm^{-1} Each data point represents the mean ± SD of three different experiments.*

Nitrocefin [µM]					
	PDMA on PP		control		
	Mean	SD	Mean	SD	p-value
750	385.33	56.25	193.43	13.21	0.0206
625	411.77	36.80	163.33	29.36	0.0151
500	330.98	30.66	132.16	6.21	0.0113
375	281.39	12.14	125.33	10.24	0.0040
250	195.57	13.39	90.00	0.30	0.0051
125	115.95	9.85	65.10	8.52	0.0369
62.5	60.55	6.16	48.93	4.04	
0	0.000	0.000	0.000	0.000	

Table S11 Nitrocefin substrate turnover (µM) of TEM-1 β-lactamase *with varied concentrations of nitrocefin substrate after 60 minutes incubation. Experiments were performed in polypropylene wells functionalized with PDMA and non-functionalized (control). Substrate turnover was measured with a TECAN Infinite 200 Pro at 492 nm. The amounts of substrates converted by the enzyme were determined from the measured absorbance values and the corresponding extinction coefficient of 17,400 M^{-1} cm^{-1} Each data point represents the mean ± SD of three different experiments.*

Nitrocefin [µM]					
	PDMA on PC		control		
	Mean	SD	Mean	SD	p-value
750	277.35	34.79	80.59	18.12	0.0139
625	238.95	4.23	81.76	8.65	0.0021
500	207.05	9.21	49.72	6.47	0.0032
375	168.37	5.73	49.47	5.56	0.0010
250	129.38	10.97	42.86	4.89	0.0032
125	78.82	5.32	18.60	6.86	0.0020
62.5	36.55	1.21	13.01	5.19	0.0182
0	0.000	0.000	0.000	0.000	

Table S12 Initial reaction rates of nitrocefin substrate turnover by TEM-1 β-lactamase *in polypropylene microtiter plate wells. Substrate turnover was monitored as absorbance increase with a TECAN Infinite 200 Pro at 492 nm in 10 second intervals. Initial reaction rates were calculated from the linear range of substrate turnover at different substrate concentrations. Each data point represents the mean ± SD of three different experiments.*

Nitrocefin [µM]	V_0				
	PDMA on PP		control		
	Mean	SD	Mean	SD	p-value
750	3.112	0.440	1.786	0.095	0.0232
625	3.118	0.156	1.576	0.227	0.0055
500	2.584	0.092	1.330	0.041	0.0037
375	2.348	0.020	1.264	0.142	0.0045
250	1.815	0.098	0.973	0.017	0.0046
125	1.522	0.077	0.824	0.084	0.0168
62.5	1.101	0.037	0.682	0.094	0.0307
0	0.000	0.000	0.000	0.000	

Table S13 Initial reaction rates of nitrocefin substrate turnover by TEM-1 β-lactamase *in polycarbonate microtiter plate wells. Substrate turnover was monitored as absorbance increase with a TECAN Infinite 200 Pro at 492 nm in 10 second intervals. Initial reaction rates were calculated from the linear range of substrate turnover at different substrate concentrations. Each data point represents the mean ± SD of three different experiments.*

Nitrocefin [µM]	V_0				
	PDMA on PC		control		
	Mean	SD	Mean	SD	p-value
750	2.179	0.206	0.847	0.197	0.0287
625	2.076	0.037	1.085	0.107	0.0017
500	1.967	0.043	0.859	0.092	0.0047
375	1.477	0.096	0.694	0.024	0.0071
250	1.288	0.061	0.659	0.019	0.0017
125	1.005	0.026	0.445	0.043	0.0046
62.5	0.614	0.047	0.262	0.010	0.0027
0	0.000	0.000	0.000	0.000	