

Supplementary Information

Highly efficient β -lactamase assay applying Poly-Dimethylacrylamide-based surface functionalization with β -lactam antibiotics and β -lactamase inhibitors

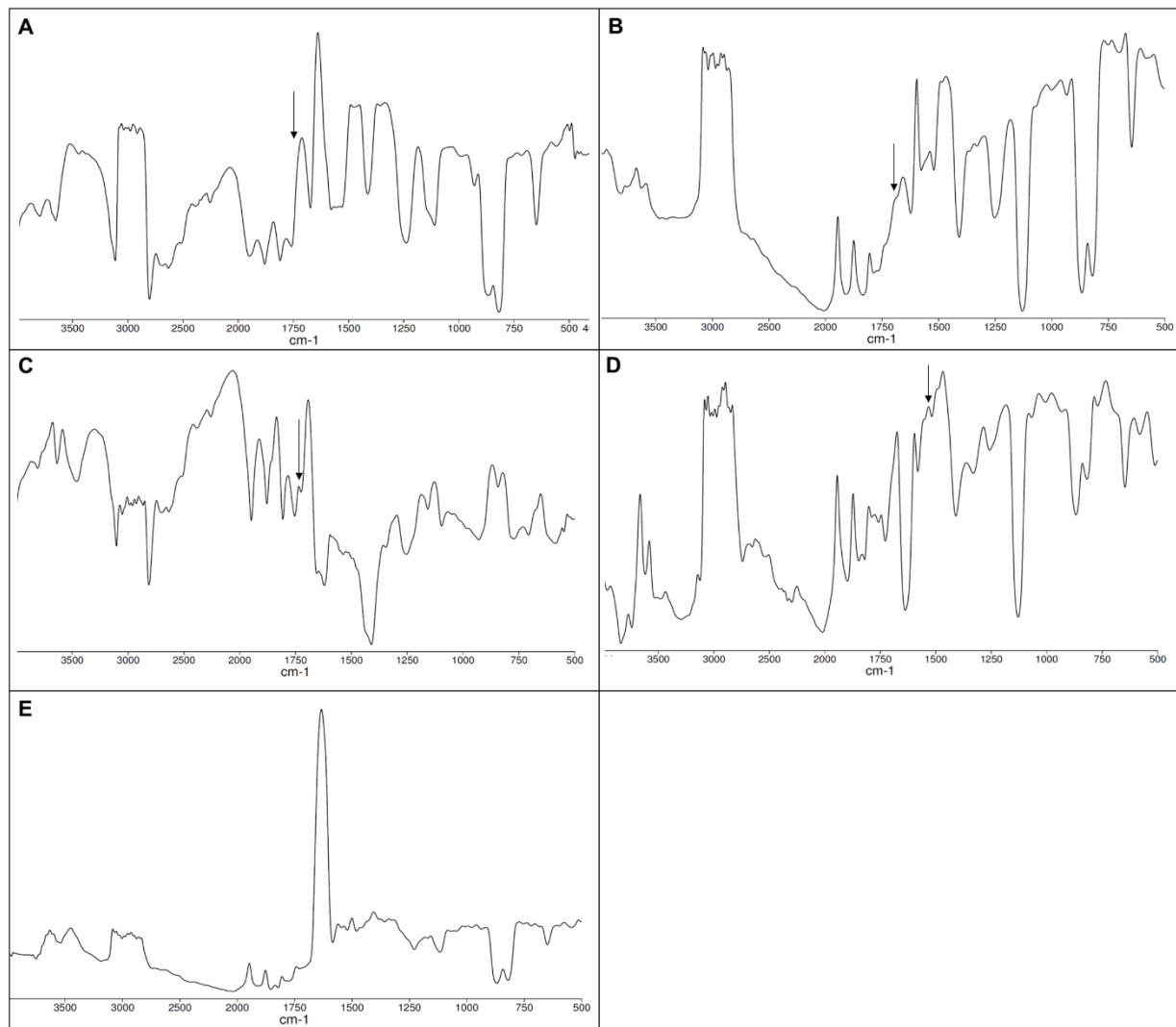


Fig. S1 Recorded Fourier transform-infrared spectra of β -lactam antibiotics and β -lactamase inhibitors crosslinked with PDMA-MBP on polypropylene plates. (A) ampicillin, (B) cefoperazone, (C) sulbactam and (D) relebactam. Spectra were recorded in the range of 4000 – 400 cm^{-1} . Relevant peaks are marked with arrows.

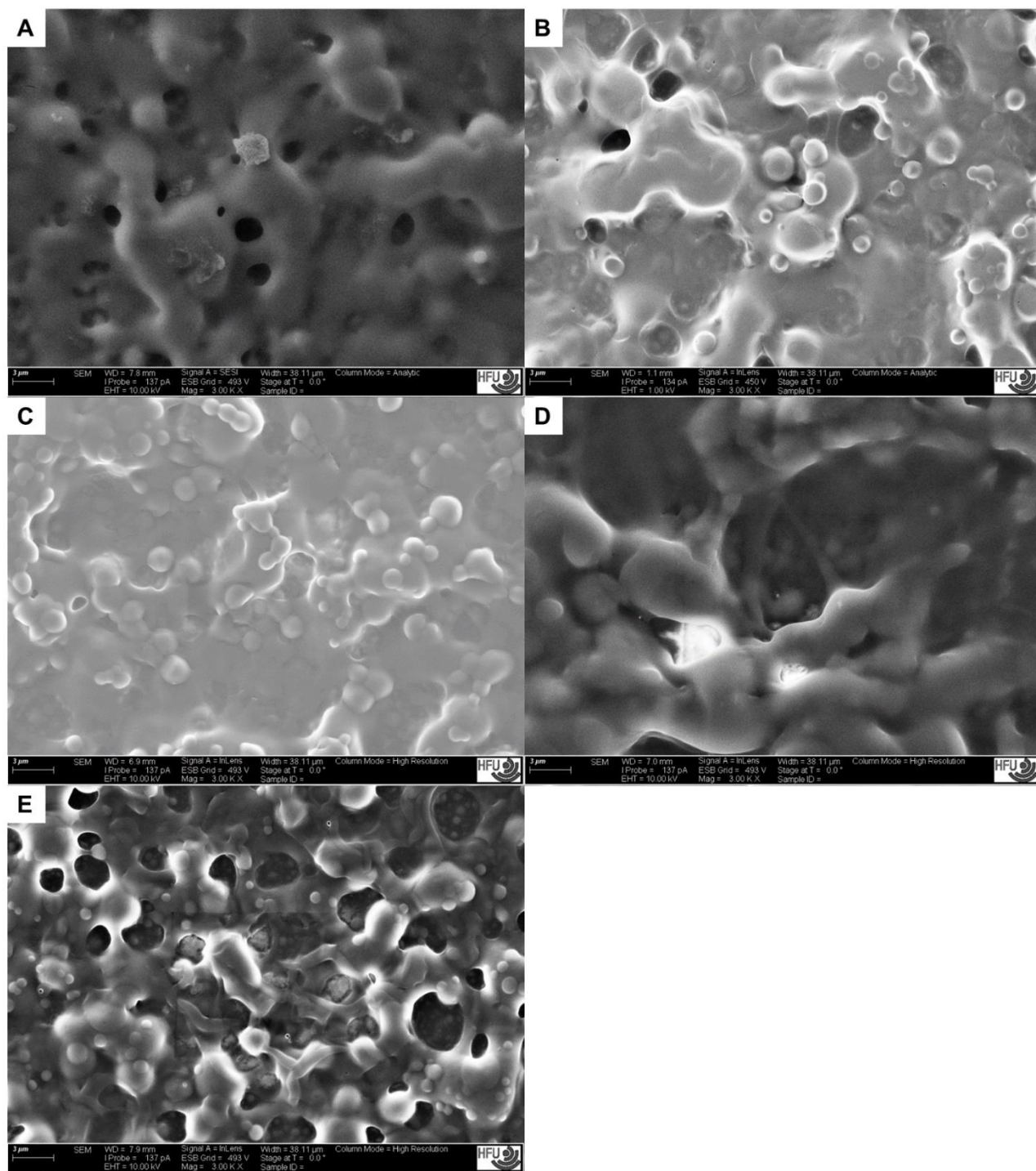


Fig. S2 SEM images of the PDMA coatings crosslinked with different β -lactam antibiotics and β -lactamase inhibitors. (A) PDMA, (B) PDMA with meropenem, (C) PDMA with aztreonam, (D) PDMA with ampicillin, (E) PDMA with cefoperazone.

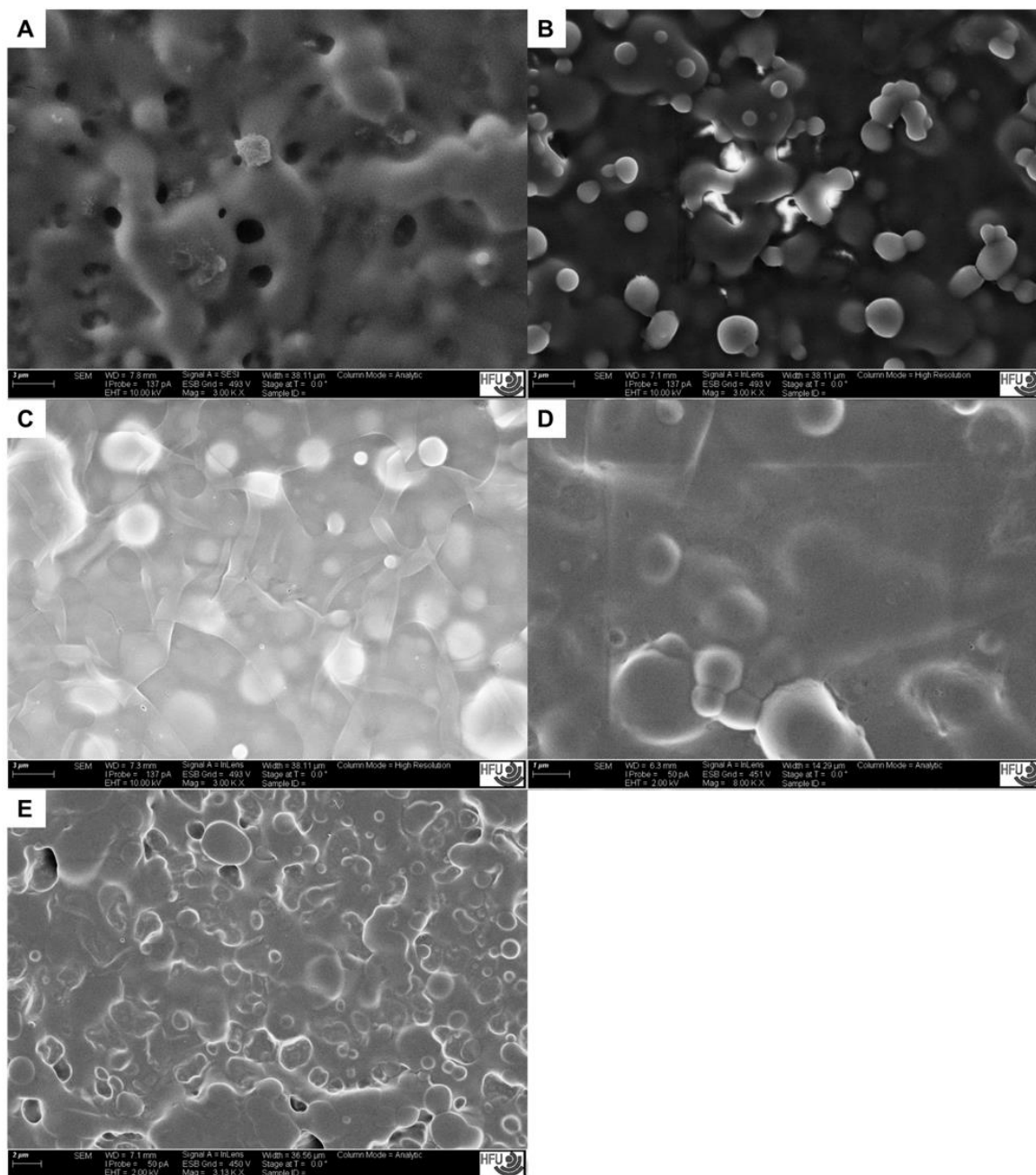


Fig. S3 SEM images of the PDMA coatings crosslinked with different β -lactamase inhibitors. (A) PDMA, (B) PDMA with avibactam, (C) PDMA with tazobactam, (D) PDMA with sulbactam and (E) PDMA with relebactam.

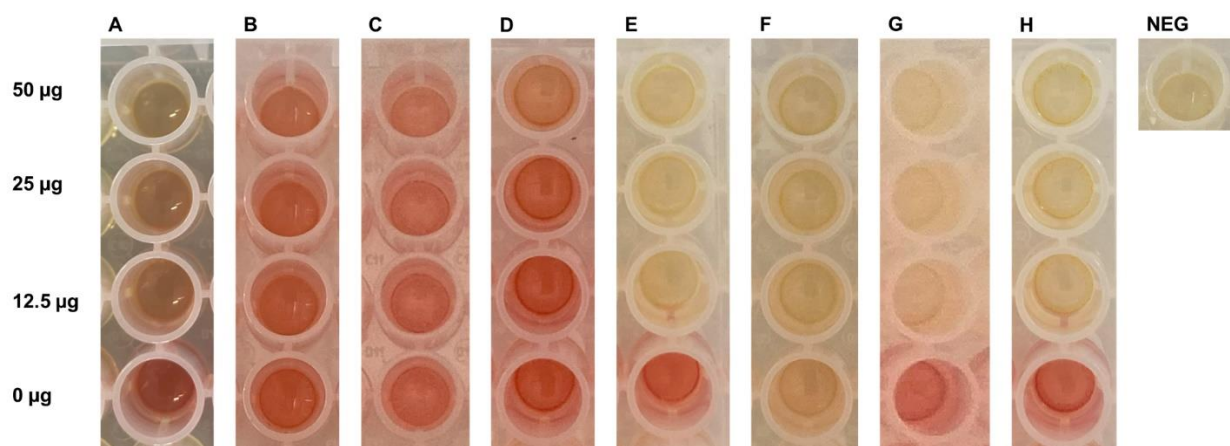


Fig. S4 Images of the β -lactamase inhibition assays for the determination of the effect of different amounts of immobilized β -lactam-antibiotics and β -lactamase inhibitors. Colour change of nitrocefin substrate turnover of different recombinant β -lactamases after preincubation in microplate wells modified with 50, 25 and 12.5 μg of various β -lactam antibiotics and β -lactamase inhibitors prepared by UV-crosslinking via PDMA. (A) ampC incubated on meropenem, (B) OXA-23 on aztreonam, (C) ampC on ampicillin, (D) OXA on cefoperazone, (E) ampC on avibactam, (F) “SiA1” lactamase blend on tazobactam, (G) ampC incubated on sulbactam and (H) ampC on relebactam.

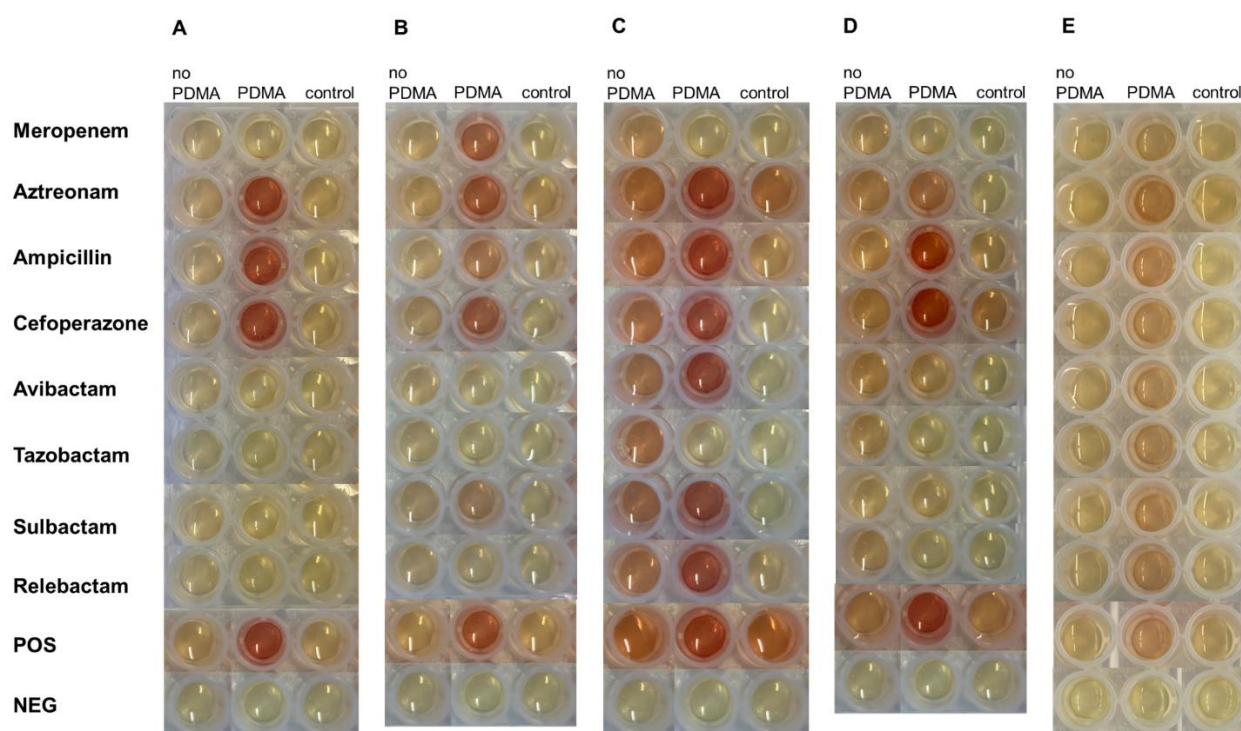


Fig. S5: Images of β -lactamase (recombinant) inhibition assay in microplate wells functionalized with β -lactam-antibiotics and β -lactamase inhibitors. Colour change of nitrocefin substrate turnover of different recombinant β -lactamases (A) TEM-1, (B) “SiA1”-lactamase blend, (C) OXA-23, (D) ampC and (E) IMP-4 after preincubation in microplate wells modified with 50 μg of various β -lactam antibiotics and β -lactamase inhibitors prepared by UV-crosslinking via PDMA (“PDMA”). To verify the functionality of the wells modified with PDMA and β -lactam-antibiotics/ β -lactamase inhibitors, inhibition assays in solution were performed as reference experiments (“control”). Therefore, β -lactamases were added to each well of an untreated 96-well plate and preincubated with β -lactam antibiotic/ β -lactamase inhibitor in solution, followed by the addition of nitrocefin. To exclude nonspecific binding of the β -lactam antibiotics/ β -lactamase inhibitors to the wells, wells without PDMA were prepared as controls (“no PDMA”). Therefore, only the β -lactam antibiotics/ β -lactamase inhibitors were added and crosslinked with UV in the wells. High

activity of lactamases was detected in each of these “no PDMA”-control wells. Therefore, nonspecific binding of the antibiotics/inhibitors to the wells can be excluded.

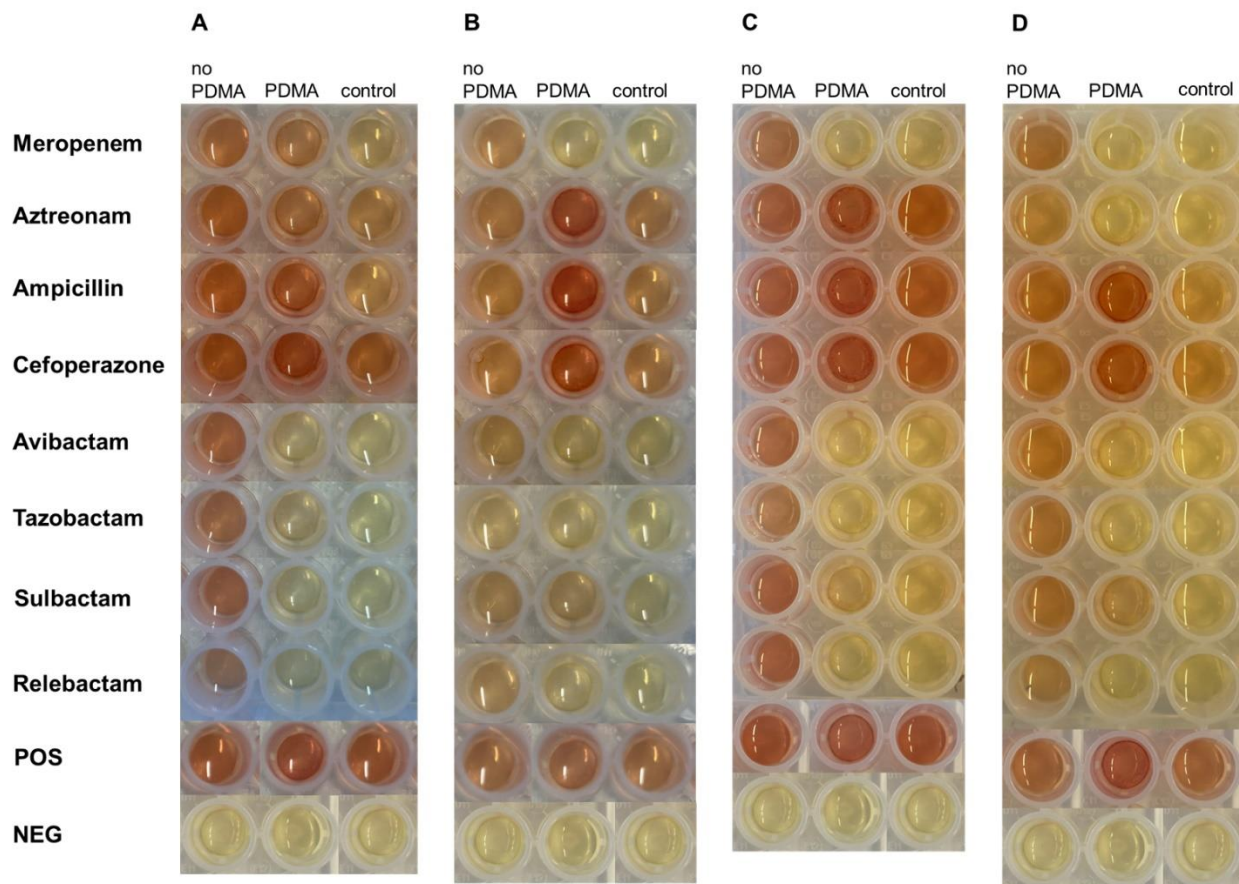


Fig. S6 Images of β -lactamase (recombinant) inhibition assay in microplate wells functionalized with β -lactam-antibiotics and β -lactamase inhibitors. Colour change of nitrocefin substrate turnover of different recombinant β -lactamases (A) *Acinetobacter baumannii*, (B) *Escherichia coli*, (C) *Klebsiella pneumoniae* and (D) *Pseudomonas aeruginosa* after preincubation in microplate wells modified with 50 μ g of various β -lactam antibiotics and β -lactamase inhibitors prepared by UV-crosslinking via PDMA (“PDMA”). To verify the functionality of the wells modified with PDMA and β -lactam-antibiotics/ β -lactamase inhibitors, inhibition assays in solution were performed as reference experiments (“control”). Therefore, β -lactamases were added to each well of an untreated 96-well plate and pre-incubated with β -lactam antibiotic/ β -lactamase inhibitor in solution, followed by the addition of nitrocefin. To exclude nonspecific binding of the β -lactam antibiotics/ β -lactamase inhibitors to the wells, wells without PDMA were prepared as controls (“no PDMA”). Therefore, only the β -lactam antibiotics/ β -lactamase inhibitors were added and crosslinked with UV in the wells. High activity of lactamases was detected in each of these “no PDMA”-control wells. Therefore, nonspecific binding of the antibiotics/inhibitors to the wells can be excluded.