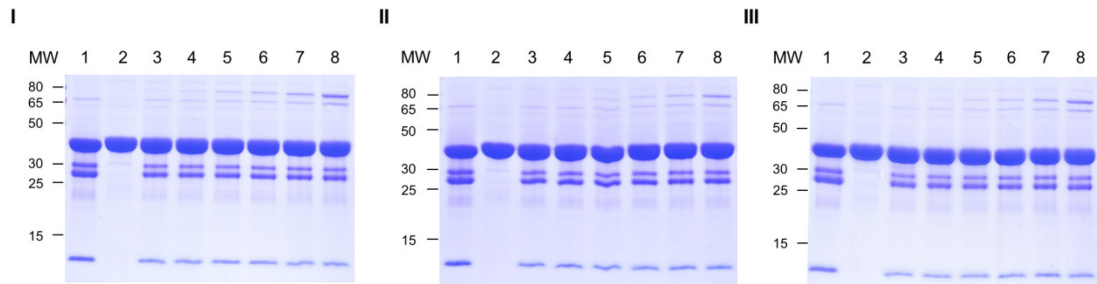


A



B



S9 Fig: Rates of isopeptide bond formation of 3kptC^T-MBP variants on the minute scale.

(A) Sequence alignment of different 3kptC^T-MBP variants (I: 3kptC^T wildtype (residues T505-K518), II: N-terminal N506-Q507 extension of 3kptC^T, III: C-terminal GWI instead of PTK in 3kptC^T). (B) Comparative covalent intermolecular bond formation assay between different 3kptC^T-MBP variants and mCherry-3kptC^C (0min-30min). Purified 3kptC^T-MBP variants and mCherry-3kptC^C proteins were mixed each at 15 μM (final conc.) for 30min at 25°C with shaking at 500 rpm before boiling (10min, 95°C) and SDS-PAGE with Coomassie staining. Interaction I: mCherry-3kptC^C + 3kptC^T (wildtype), interaction II: mCherry-3kptC^C + 3kptC^T (NQ), interaction III: mCherry-3kptC^C + 3kptC^T (GWI). (lane 1: mCherry-catcher input (30μM), lane 2: tag input (30μM), lane 3: 0min, lane 4: 1min, lane 5: 3min, lane 6: 5min, lane 7: 10min, lane 8: 30min). Same volume was loaded on the gel. MW stands for molecular weight (kDa).