



S6 Fig: Covalent bond formation assays of 4oq1 active site mutants.

(A) $4oq1^C + 4oq1^T$ -MBP (wildtype interaction); (B) $4oq1^C + 4oq1^T(N252A)$ -MBP; (C) $4oq1^C(K155A) + 4oq1^T$ -MBP; (D) $4oq1^C(E222Q) + 4oq1^T$ -MBP. The wildtype interaction showed a covalent product increasing over time whereas no covalent product was detected in the active site mutants. Comparative covalent bond formation assays were performed with $15\mu\text{M}$ final concentration of the reactants. The reactive residues N252 in the $4oq1^T$ and K155 and E222 in the $4oq1^C$ were mutated and tested for covalent product formation via SDS-PAGE and subsequent Coomassie Brilliant blue staining. Samples were boiled at 95°C for 10min prior to gel loading. (A-D): lane 1: catcher input ($30\mu\text{M}$), lane 2: tag input ($30\mu\text{M}$), lane 3: 0h, lane 4: 1h, lane 5: 2h, lane 6: 3h, lane 7: 4h, lane 8: 24h). Same volume of samples were loaded. MW stands for molecular weight (kDa).