



**Supplemental Figure S2. Purification of wt and mutant BlaC enzymes.** The wt and BlaC mutant enzymes were all purified by Ni-NTA chromatography followed by size exclusion chromatography. An equal amount of each purified enzyme was loaded onto an SDS-PAGE gel, which was subsequently stained with Coomassie blue to visualize proteins. (a) Following purification, the N-terminal 6x-His tag was removed by thrombin cleavage. (b) For nitrocefin analysis, the BlaC(I105T/T145dI/V263I) mutant was purified in addition to wt and BlaC(I105F) mutants. MW, molecular weight ladder. The expected size of BlaC with 6x-His tag is ~32 kDa.