Kinetic Characterizations of Nitrocefin, Cefoxitin, and Meropenem Hydrolysis

by $\beta$-lactamase of *Mycobacterium tuberculosis*

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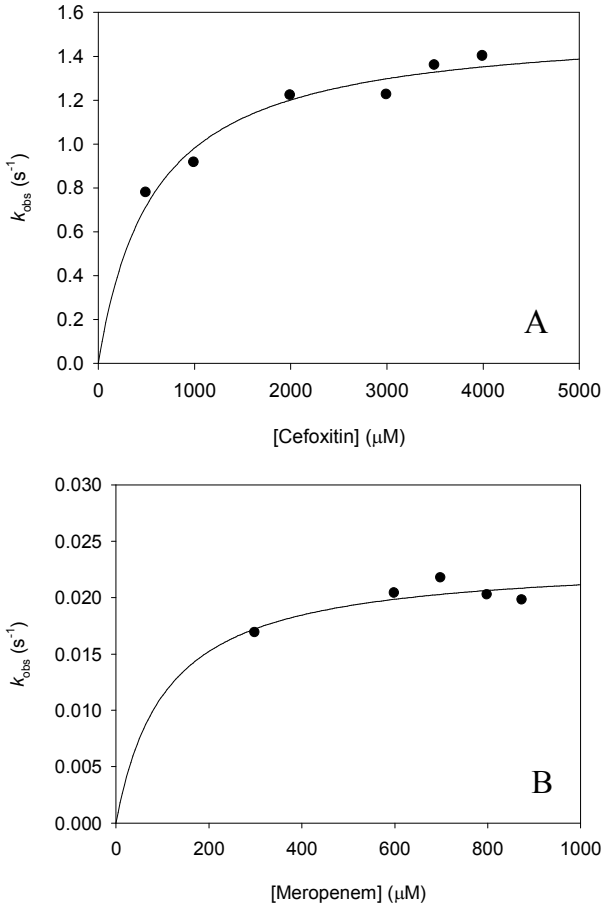


Figure S1. Saturation curves for (A) cefoxitin and (B) meropenem hydrolysis. When $k_{obs}$ reaches saturation, $k_1[S] >> k_2$ and BlaC is considered sufficiently saturated to produce pseudo-first-order kinetics for cefoxitin hydrolysis. Concentrations of meropenem below 600 $\mu$M resulted in poor fit likely due to contributions from binding and $k_{obs}$ values could not be obtained. At concentrations of meropenem above 600 $\mu$M, $k_{obs}$ values were relatively constant, suggesting that the enzyme is sufficiently saturated.

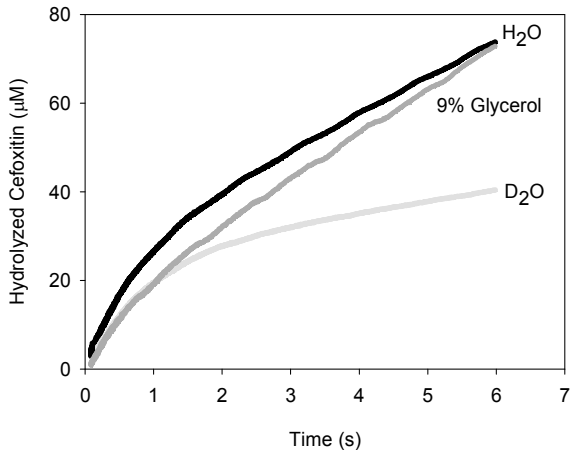


Figure S2. Burst kinetic isotope effects on cefoxitin in $H_2O$ (black), $D_2O$ (light gray), and 9% glycerol (gray).

**Table S1.** Pre-steady state kinetic fitting

| Substrate | Cefoxitin | Meropenem |
|---|---|---|
| $k_2$ ($s^{-1}$) | 1.4 ± 0.01 | (1.9 ± 0.3) x $10^{-2}$ |
| $k_3$ ($s^{-1}$) | 0.4 ± 0.001 | (4.0 ± 1.0) x $10^{-4}$ |
| $A_o$ | 0.67 ± 0.001 | 0.95 ± 0.01 |
| Pre-steady state $k_{cat}$ ($s^{-1}$) | 0.30 ± 0.01 | (4.0 ± 1.0) x $10^{-4}$ |

$k_2$, rate of acylation; $k_3$, rate of deacylation; $A_o$, burst amplitude; $k_{cat}$, turnover number.