Kinetic and Structural Studies on Metallo-β-Lactamase ImiS

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Bacterial resistance to antibiotics is a growing public health concern. Zn(II)-containing β-lactamases (metallo-β-lactamases, MβL's) contain 1-2 moles of Zn(II) per mole of enzyme, hydrolyze all known cephalosporins, carbapenems, and penicillins, are not inhibited by clavulanic acid and other classical β-lactamase inhibitors, and have no known clinically-useful inhibitor towards them. Previous studies have shown that there is significant structural and mechanistic diversity among the MβL's, leading to the grouping of the enzymes into three distinct subclasses. The objective of our research is to probe the mechanism and structure of a representative enzyme from each of the distinct MβL subclasses in an effort to uncover common structural and mechanistic properties of the enzymes. This work describes our studies on a subclass B2 MβL, ImiS from *Aeromonas veronii bv. sobria*. EPR, ^{1}H NMR, UV-Vis, and EXAFS studies reveal that the catalytic site in ImiS is the consensus Zn_2 site in the MβL's. Presteady state kinetic studies reveal that the B2 MβL's utilize a novel reaction mechanism to catalyze the hydrolysis of carbapenems.