Kinetic and Spectroscopic Characterization of TauD Inhibitors

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Taurine/a-ketoglutarate dioxygenase (TauD) is an *Escherichia coli* Fe(II)-containing enzyme that converts taurine (2-aminoethanesulfonate) to sulfite, which can be used as a sulfur source. This reaction is coupled with the conversion of a-ketoglutarate (aKG) to succinate (Figure 1). In order to clarify the structure and properties of reaction intermediates we have examined the kinetics of selected TauD inhibitors and tested their abilities to trap catalytic intermediates. N-oxalyl glycine (NOG), for example, a known inhibitor of related enzymes such as prolyl hydroxylase and FIH (factor inhibiting HIF, an asparaginyl hydroxylase), was examined for its interaction with TauD. We demonstrated that NOG is a weak inhibitor (K_i ~0.2 mM) that competes with aKG. Although NOG is known from crystallographic studies to chelate Fe(II) in the same manner as aKG when bound to FIH, NOG fails to produce the diagnostic aKG/Fe(II) chromophore (530nm) known for this enzyme family. Exposure of the NOG-TauD/Fe(II) sample to oxygen resulted in the formation of a ~700nm chromophore resembling that produced with the succinate-TauD/Fe(II) sample, but distinct from that generated with aKG-TauD/Fe(II) sample. EPR spectroscopy was used to confirm NOG binding to the TauD metal ion of anaerobic sample in the presence of the oxygen analogue NO. More specifically, the EPR signal of TauD/Fe(II)-NO change from being slightly rhombic, to become more axial upon addition of NOG, which further changed to become more rhombic upon addition of taurine.

Figure1: TauD reaction