Oral presentation

Oxidative damage on Mo/W Formate dehydrogenases and their innate protection mechanism

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The reversible CO_2/formate interconversion by Mo/W-Formate dehydrogenases (Fdhs) is a promising route not only for green-house gas sequestration but also to sustainably produce fuel. Formate is a safe option for hydrogen storage/delivery (53g H_2/L) in cell power applications [1].

W-FdhAB (periplasmic heterodimer; W active site: bisMGD, Se(Cys), S ligand; 4x[4Fe-4S]) is the main responsible for CO_2 reduction in *D. vulgaris*[2] and a suitable model for CO_2 reduction biocatalytic applications due to its robustness and high catalytic activity [3, 4].

Mo/W Fdhs are O_2 sensitive which hampers industrial use as biocatalysts. Nonetheless, the chemical/structural consequences of O_2- induced damage remain unknown yet are crucial for devising protective mechanisms. Our recent study[5], combining biochemical, spectroscopic, and structural studies of *Dv*FdhAB reveals that O_2 inactivation is promoted by the presence of substrate and results in the formation of a new active site species, consistently captured in the crystal structures. The process involves the displacement of the catalytic SeCys from tungsten coordination, replaced by a O_2 or H_2O_2 molecule (Figure 1). In addition, we proved that oxidative inactivation does not require Mo/W reduction, as widely assumed, occurring in the oxidized state in the presence of CO_2 [5].

Nonetheless, *Dv*FdhAB is more oxygen-tolerant than other Fdhs and can be purified aerobically in the absence of substrates [3]. Our team found that the formation of a conserved disulfide bond [6], reduces enzyme activity and protects it from oxidative inactivation. *Dv*FdhAB can protect itself from transient O_2 damage when exposed to physiological concentrations of formate (low μM). Our structural studies disclosed the allosteric mechanism responsible for transducing the signal from the surface exposed disulfide bond to the deeply buried active site [6].

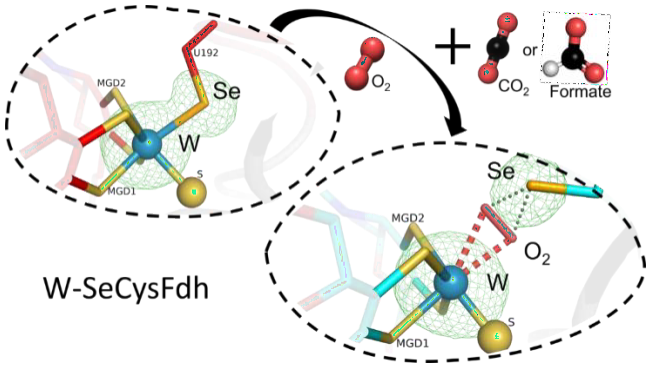

Figure 1. SeCys displacement on W-FdhAB. Anomalous difference Fourier map in green mesh (contoured at 5σ).

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This work was supported by FCT - PTDC/BII- BBF/2050/2020 (DOI:10.54499/PTDC/BII-BBF/2050/2020) and R&D units MOSTMICRO-ITQB (UIDB/04612/2020 and UIDP/04612/2020) and UCIBIO (UIDP/04378/2020 and UIDB/04378/2020) and Associated Laboratories LS4FUTURE (LA/P/0087/2020) and i4HB (LA/P/0140/2020).