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Capturing X-ray-Induced Photo-Reduction in Arsenite Oxidase: Implications for the Catalytic Mechanism

The molybdenum enzyme arsenite oxidase (Aio), is a promising biocatalyst for the detoxification and biosensor applications [1]. So far, its structural characterization and complete reaction mechanism understanding has been limited to artifactually reduced states of the Mo cofactor (Moco) caused by X-ray photoreduction (PDB 1g8k, 4aay) [2-4].

Here, we present the first crystallographic evidence of X-ray-induced photo-reduction in *Alcaligenes faecalis* Aio, tracking active-site geometric changes across incremental radiation doses (100–1000 kGy) at the PETRA III P14 beamline. High-resolution structures (1.5 Å) reveal that photo-reduction triggers the loss of a labile oxo ligand (O1), while displacing the Mo atom toward the dithiolene plane.

Low-dose data (100 kGy) allowed us to capture, for the first time, the Mo(VI) centre in a six-coordinated geometry with asymmetric cis-dioxo coordination (Mo–O1: 1.8 Å; Mo–O2: 2.1 Å). Reduction of the Moco also flattened the twist angle of the pterin from 31° to 17°, modulating the active site catalysis.

Our work demonstrates that radiation damage artifacts—prevalent in metalloenzyme crystallography—can obscure mechanistic insights. These findings underscore the need for dose-optimized structural data to refine AI-driven models of metalloenzyme mechanisms and **oxidised to reduced, rational engineering** **Reduced: 1000 kGy** **ing** and bioremediation.

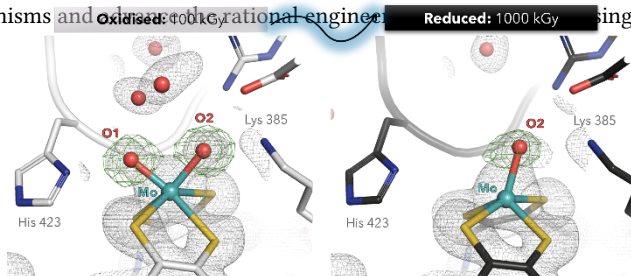


Figure 1: enter image description here

References

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- [2] Warelow T.P. et al., (2013) *PLoS One*, 8(8): e72535.
- [3] Ellis, P.J. et al. (2001) *Structure*, 9(2), 125-132.
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