Structural and mechanistic insights in dimeric chlorite dismutase - Impact of pH and the dynamics of the catalytic arginine

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Chlorite dismutases (Clds) are heme \(b\) containing oxidoreductases, which catalyse the conversion of chlorite into harmless chloride and molecular dioxygen. They differ in oligomerisation, subunit architecture and the hydrogen bonding network of the catalytic arginine. Many questions about the molecular reaction mechanism have remained unanswered, including binding of anionic angulate ligands like nitrite and the substrate chlorite or the electronic nature of the active oxoiron(IV) intermediate and its interaction with the catalytically essential arginine. Here, we have investigated dimeric Cld from *Cyanothec* sp. PCC7425 (CCld) and two variants having the catalytic arginine R127 (i) hydrogen-bonded to glutamine Q74 (wild-type CCld), (ii) arrested in a salt bridge with a glutamate (Q74E) or (iii) being fully flexible (Q74V). We present the X-ray crystal structures of Q74V and Q74E\(^1\) and demonstrate the pH-induced changes in the environment and coordination of the heme iron by UV–vis, electron paramagnetic resonance spectroscopies\(^2\) as well as differential scanning calorimetry. Sequential stopped-flow studies reveal the initial and transient appearance of Compound I in the reaction between CCld and chlorite at pH 5.0 and 7.0 and the dominance of an oxoiron(IV) species during the chlorite degradation period. The dynamics of R127 does not affect hypochlorite mediated Compound I formation, but has an influence on Compound I stability which decreases rapidly with increasing pH. Compound I has been shown to oxidize iodide, chlorite and serotonin but not hypochlorite. Serotonin is able to attenuate oxidative damage and inactivation of CCld at neutral and alkaline pH. Presented data are discussed in terms of the molecular reaction mechanism by Clds and the pronounced pH-dependence of chlorite degradation.

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References

\(^1\) Schmidt D, Serra I, et al, doi: 10.1021/acs.biochem.0c00910.