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STRUCTURAL BASIS FOR THE MAGNESIUM-DEPENDENT ACTIVATION OF TRANSKETOLASE FROM *CHLAMYDOMONAS REINHARDTII*

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In photosynthetic organisms, transketolase (TK) is involved in the Calvin-Benson cycle and participates to the regeneration of ribulose-5-phosphate. Previous studies demonstrated that TK catalysis is strictly dependent on thiamine pyrophosphate (TPP) and divalent ions such as Mg^{2+}. TK from the unicellular green alga *Chlamydomonas reinhardtii* (CrTK) was recombinantly produced and purified to homogeneity. Biochemical properties of the CrTK enzyme were delineated by activity assays and its structural features determined by CD analysis and X-ray crystallography. CrTK is homodimeric and its catalysis depends on the reconstitution of the holo-enzyme in the presence of both TPP and Mg^{2+}. Activity measurements and CD analysis revealed that the formation of fully active holo-CrTK is Mg^{2+}-dependent and proceeds with a slow kinetics. The 3D-structure of CrTK without cofactors ($CrTK_{apo}$) shows that two portions of the active site are flexible and disordered while they adopt an ordered conformation in the holo-form. Oxidative treatments revealed that Mg^{2+} participates in the redox control of CrTK by changing its propensity to be inactivated by oxidation. Indeed, the activity of holo-form is unaffected by oxidation whereas CrTK in the apo-form or reconstituted with the sole TPP show a strong sensitivity to oxidative inactivation. These evidences indicate that Mg^{2+} is fundamental to allow gradual conformational arrangements suited for optimal catalysis. Moreover, Mg^{2+} is involved in the control of redox sensitivity of CrTK. The importance of Mg^{2+} in the functionality and redox sensitivity of CrTK in the physiological context will be discussed.