STRUCTURAL BASES OF P_{II} SIGNALING IN PHOTOSYNTHETIC ORGANISMS

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In microorganisms and plants the homotrimeric P_{II} protein is a key mediator of energy, carbon and nitrogen interactions, being a central regulator of carbon:nitrogen metabolism. P_{II} experiences allosteric changes triggered by 2-oxoglutarate and ATP, and can be uridylylated or phosphorylated. In photosynthetic organisms, under conditions of carbon and nitrogen abundance, P_{II} associates with N-acetyl-L-glutamate kinase (NAGK), the arginine feed-back inhibited enzyme which controls arginine biosynthesis. P_{II} binding renders NAGK more active and less sensitive to feed-back inhibition by arginine. In this way, arginine can accumulate and nitrogen is stored as arginine. The crystal structure at 2.75 Å-resolution of the complex of two P_{II} trimers with one NAGK hexamer (a doughnut-like trimer of dimers) of the cyanobacterium *Synechococcus sp.* explains these effects of P_{II} on NAGK. Both P_{II} molecules and NAGK bind with their threefold axes aligned. Each P_{II} trimer sits on an opposite face of the NAGK ring. One P_{II} subunit contacts only one NAGK subunit. The protruding T-loop of each P_{II} subunit plays a key role in the interactions with NAGK, and adopts a novel compact conformation. Since ADP favors an extended conformation of the T-loop, it triggers dissociation of the complex. 2-Oxoglutarate does not change the T-loop shape but may prevent complex formation by rendering P_{II} more negatively charged. Charge effects as well as steric clash, and the loss of hydrogen bonds with NAGK, explain why the phosphorylation of S49 of P_{II} prevents complex formation. Through a dense network of predominantly polar interactions with both domains of the NAGK subunit, the T-loop and the B-loop of P_{II} glue together both domains of each NAGK subunit, favoring the contact of the two substrates of NAGK (each substrate binds in one domain), thus countering arginine inhibition, which is mediated by increasing the distance between the sites of the two substrates. In this way, P_{II} and arginine favor alternative contracted/active and expanded/inactive NAGK ring conformations. The contacts of the two proteins explain why NAGK-P_{II} complex formation is restricted to photosynthetic organisms.

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