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Identification of protein partners of the Azotobacter vinelandii rhodanese-like protein RhdA by pull-down approaches

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Abstract

Rhodaneses (TST; EC 2.8.1.1) catalyze in vitro the transfer of sulfane sulfur from thiosulfate to cyanide with the concomitant formation of thiocyanate. Genome sequence analyses evidenced the presence of more than 14000 sequences coding for putative rhodanese-like proteins (RDPs). These sequences are distributed in all taxa thus suggesting RDPs involvement in biological processes different from cyanide detoxification. Studies aiming to unravel RDP biological functions highlighted the relationship between members of this superfamily and the maintenance of the intracellular redox homeostasis. This ability may rely in the peculiar RDP catalytic domain that consists of a cysteine residue centered in an electropositive environment that promotes the stabilization of the bio-active persulfide sulfur. The aim of this work was to gain more insights about RDP biological function by the identification of protein interaction partners. For this reason we used tagged recombinant versions of the biochemically characterized RDP RhdA from Azotobacter vinelandii, as a bait to isolate interaction partners by pull-down approaches. We identified IlvC, an oxidative sensitive ketol-acid reductoisomerase (KARI; EC 1.1.1.86). The partial inhibition of KARI activity in the A. vinelandii RhdA-null mutant strain suggests a functional interaction between RhdA and IlvC that can be related to the protection against oxidative stress events.