

Heterologous functionality and roles of conserved cysteine motifs of the [NiFe]-hydrogenase accessory protein, HupK/HoxV

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Abstract

Numerous auxiliary proteins participate in the complex posttranslational modification process of [NiFe]-hydrogenases. In *Thiocapsa roseopersicina*, the HupK protein is important for the formation of active membrane-bound hydrogenases. The HupK proteins of various origins have moderate similarity to each other and to the large subunits of [NiFe]-hydrogenases. Site directed mutagenesis experiments were performed to disclose the role of the highly conserved cysteines in HupK. Cys54 was shown to be indispensable for the proper function of HupK and recreation of a large subunit like cysteine profile had negative effect on the maturation processes. Although, the results of the mutagenesis study slightly differed from that obtained for *Ralstonia eutropha* HoxV, it was clearly demonstrated that HupK from *T. roseopersicina* and HoxV from *R. eutropha* can substitute each other. It was also demonstrated that HoxV could be involved in the maturation of both Hup and Hyn hydrogenases in *T. roseopersicina*.

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