Volume 50(3-4):169, 2006
Acta Biologica Szegediensis
http://www.sci.u-szeged.hu/ABS

DISSERTATION SUMMARY

The *Thiocapsa roseopersicina* genome project and the use of results in the hydrogenase research

Lívia Palágyi-Mészáros

Department of Biotechnology, University of Szeged, Szeged, Hungary, and Institute of Biophysics, Biological Research Center, Hungarian Academy of Sciences, Szeged, Hungary

Hydrogenases are metalloenzymes catalyzing the oxidation of molecular hydrogen and the reverse reaction (Vignais et al. 2001).

Thiocapsa roseopersicina BBS is a Gram-negative, photosynthetic purple, sulphur bacterium. The bacterium contains is able to fix atmospheric N_2, a process accompanied by H_2 production.

Thiocapsa roseopersicina harbours at least three different types of NiFe hydrogenases: HynSL, HupSL, HoxEFUYH. It is an intriguing question why the cells need so many distinct hydrogenases (Kovacs et al. 2005).

The NiFe hydrogenases contains a complex metallocenter composed of a Ni and Fe atom. The assemly of this center requires concerted action of numerous so-called accessory enzymes. Moreover, the hydrogen metabolism is linked to other bioenergetic/metabolic processes like photosynthesis, sulfur- and nitrogen metabolism, respiration etc. For characterization of the various metabolic pathways related to the hydrogen metabolism the whole genome sequencing of *T. roseopersicina* was started.

First of all a shotgun library was constructed. More then 15 000 clones were sequenced from both sides and the sequences assembled into 2400 contigs after clearance. As a next step a cosmid library was constructed which can be used to assemble the existing sequences into larger contigs and to fill the gaps between them. The sequencing of more than 3000 cosmid clones are in progress. Although the genome sequencing has not finished yet some of the important genes were found in the shotgun library.

In few microbes it was shown that the membrane associated hydrogenases are transported into the periplasmatic space via twin-arginin-translocation (Tat) pathway. The Tat pathway has a typical signal sequence, a conserved (S/T)-R-R-x-F-L-K motif, which can be found at the beginning of the small subunit of the hydrogenases also. The genes coding for the components of the Tat pathway was identified in the genome.

In the case of the membrane bound Hyn hydrogenase in *T. roseopersicina*, the structural genes are arranged unusually, since there two additional open reading frames (namely *isp1* and *isp2*) are located between the genes coding the small and the large subunit.

The function of the Isp proteins were studied by mutagenesis. It was shown that the Isp1 and Isp2 are required for the activity of the Hyn hydrogenase *in vivo* although they do not have on the hydrogenase activity *in vitro*. It was concluded that the Isp proteins were involved in the transmembrane electron transport. The Isp1 protein is a hem-containing integrated membrane protein in which characteristic signal sequence could not be observed. Hence, it is assumed that it is transported into the membrane via the Ffh/FtsY (SRP) pathway. The genes encoding the FtsY were recognized in the genome

The role of the SRP pathway in the transport of Isp1 and the Tat proteins in the export of the HynSL are going to be clarified. This would be the first case when the components of a functional membrane associated enzyme complex would be targeted by distinct pathways.

References

Vignais PM, Billoud B, Meyer J (2001) Classification and phylogeny of hydrogenases. FEMS Microbiol Rev 25(4):455-501.

Kovacs KL, Kovacs AT, Maroti G, Meszaros LS, Balogh J, Latinovics D, Fulop A, David R, Doroghazi E, Rakhely G (2005) The hydrogenases of Thiocapsa roseopersicina. Biochem Soc Trans 33(1):61-63.

Rakhely G, Colbeau A, Garin J, Vignais PM, Kovacs KL (1998) Unusual organization of the genes coding for HydSL, the stable [NiFe]hydrogenase in the photosynthetic bacterium Thiocapsa roseopersicina BBS. J Bacteriol 6:1460-1465.

Supervisors: Kornél Kovács, Gábor Rákhely
E-mail: meszaros@brc.hu