Nha1-type antiporters with different substrate specificity (or affinity to individual substrates) and functions. One of them is involved in the detoxification of cells and the other could play an important role in the maintenance of stable intracellular potassium content, cell volume and pH of the cytoplasm thanks to its ability to transport K^+.

At this time, no yeast species possessing solely a Na^+/H^+ antiporter with narrow substrate specificity (for Na^+ and Li^+) is known; therefore, the physiological role of Nha1-type antiporters seems to be complex with a participation both in the elimination of toxic cations from cells and in the maintenance of stable intracellular K^+ concentration, cell volume and cytoplasmic pH.

4. Summary

The main results of the thesis can be summarized as:

- The studies of sequences of yeast alkali metal cation/H^+ antiporters (Nha1, Nhx1 and Kha1 families) *in silico* showed that the structure of all three groups of these proteins probably contains 12 transmembrane segments, we found some conserved amino acid residues or motifs likely to be important for proper functioning of antiporters, and our phylogenetic study revealed that Nhx1 proteins are related to both plant and mammalian Na^+/H^+ antiporters, Kha1 antiporters are close to the bacterial relatives and Nha1-type proteins form a separate group so far characterized only in yeasts or fungi.
- The *D. hansenii NHA1* gene encodes a plasma membrane Na^+/H^+ antiporter with broad substrate specificity (for Na^+, Li^+, K^+ and Rb^+) playing a complex role in cell physiology (participating both in the elimination of toxic cations from cells and in the maintenance of stable intracellular K^+ concentration, cell volume and cytoplasmic pH). We also revealed the sequence of *NHA1* gene from *D. hansenii* CBS 1793 strain that was sent to the EMBL database (Acc. No. AJ876409).
- *Y. lipolytica* possesses two plasma membrane Na^+/H^+ antiporters with different functions in cell physiology. *Yl*Nha1p is able to transport K^+ and therefore could play a role in the maintenance of stable intracellular potassium content, cell volume and pH of the cytoplasm; *Yl*Nha2p is a very efficient system for elimination of toxic Na^+ or Li^+ from cells. Heterologous expression of *YlNHA2* gene could be used to improve the sodium tolerance of some *S. cerevisiae* industrial strains.

- Similarly to *Y. lipolytica*, *S. pombe* possesses two plasma membrane Na^+/H^+ antiporters differing in their substrate specificity and physiological roles. In contrast to the previously described *Sp*sod2p, the newly characterized *Sp*sod22 antiporter is able to transport K^+ and thus can be involved in the maintenance of stable intracellular K^+ concentration, cell volume and cytoplasmic pH.
- The *Z. rouxii ZrNHA1* gene encodes a plasma membrane Na^+/H^+ antiporter with an ability to export both Na^+ and K^+ from cells. Therefore, *Z. rouxii* also possesses a Nha1-type protein that can play a role in the maintenance of stable intracellular potassium content, cell volume and pH of the cytoplasm.
- The presence of at least one plasma membrane Na^+/H^+ antiporter with an ability to transport the main intracellular cation K^+ is conserved among various non-related yeasts. Therefore, the physiological role of Nha1-type antiporters seems to be complex with participation both in the elimination of toxic cations from cells and in the maintenance of stable intracellular K^+ concentration, cell volume and cytoplasmic pH.
- The co-action of osmotic and high temperature stresses results in a growth improvement of *D. hansenii*. At a lower temperature (more convenient for *D. hansenii* cultivation), no significant growth stimulation in the presence of salts or sorbitol can be observed. Furthermore, this cross-effect of the presence of two non-related stress conditions can be found in *S. cerevisiae* and *S. pombe* as well.

5. References

Bañuelos MA, Sychrová H, Bleykasten-Grosshans C, Souciet JL, Potier S (1998) *Microbiology* **144**: 2749-2758

Bañuelos MA, Ramos J, Calero F, Braun V, Potier S (2002) *Yeast* **19**: 1365-1372

Brett CL, Tukaye DN, Mukherjee S, Rao RJ (2005) *Mol Biol Cell* **16**: 1396-1405

Dujon B *et al.* (2004) *Nature* **430**: 35-44

Flis K, Hinzpeter A, Edelman A, Kurlandzka A (2005) *Biochem J* **390**: 655-664

Gonzalez-Hernandez JC, Cardenas-Monroy CA, Pena A (2004) *Yeast* **21**: 403-412

Hill JE, Myers AM, Koerner TJ, Tzagoloff A (1986) *Yeast* **2**: 163-167

Jia ZP, McCullough N, Martel R, Hemmingsen S, Young PG (1992) *EMBO J* **11**: 1631-1640

Kamauchi S, Mitsui K, Ujike S, Haga M, Nakamura N, Inoue H, Sakajo S, Ueda M, Tanaka A, Kanazawa H (2002) *J Biochem* **131**: 821-831

Kinclová O, Potier S, Sychrová H (2001a) *J Biotechnol* **88**: 151-158

Kinclová O, Ramos J, Potier S, Sychrová H (2001b) *Mol Microbiol* **40**: 656-668