Expression pattern of GPI-anchored non-specific lipid transfer proteins in *Physcomitrella patens*

Andrey Höglund

Supervisors: Monika Edstam & Johan Edqvist

Masters thesis Project in Molecular Genetics and Physiology

IFM Molecular Genetics, Linköpings University

Background & Aim

When the first plants started to colonize the terrestrial habitat approximately 450 million years ago, they faced numerous novel stresses including UV-radiation, desiccation and temperature stresses.

Non-specific lipid transfer proteins (nsLTP) are present in all land plants but not in any algae. Possibly, nsLTP evolved during the water-to-land transition and have useful properties to cope with these stressful abiotic conditions.

In the moss *Physcomitrella patens* a nsLTP subfamily is called type G, and contains 10 genes. They are characterized by the addition of the glycosylphosphatidylinositol (GPI) anchor, as a post-transcriptional modification. The GPI-anchor allows the protein to attach to the plasma membrane and face the connected protein outward to the extracellular side.

The aim of this study was to investigate the expression pattern of GPI-anchored non-specific lipid transfer proteins in *Physcomitrella patens* during abiotic stresses.

Method

- The experiments were carried out on three to four weeks old gametophytes (*Physcomitrella patens* stain Gransden 2004)
- The moss was stressed with either;
  - **NaCl**
  - **cold** (on ice)
  - **mannitol** (osmotic stress)
  - **UV-radiation**
  - **drought**
  - **copper** (heavy metal)
  - **abscisic acid** (plant hormone)
- After the stresses RNA was extracted from which cDNA was synthesized. The cDNA was used as template for the qRT-PCR.
- The results were analyzed with the software REST.

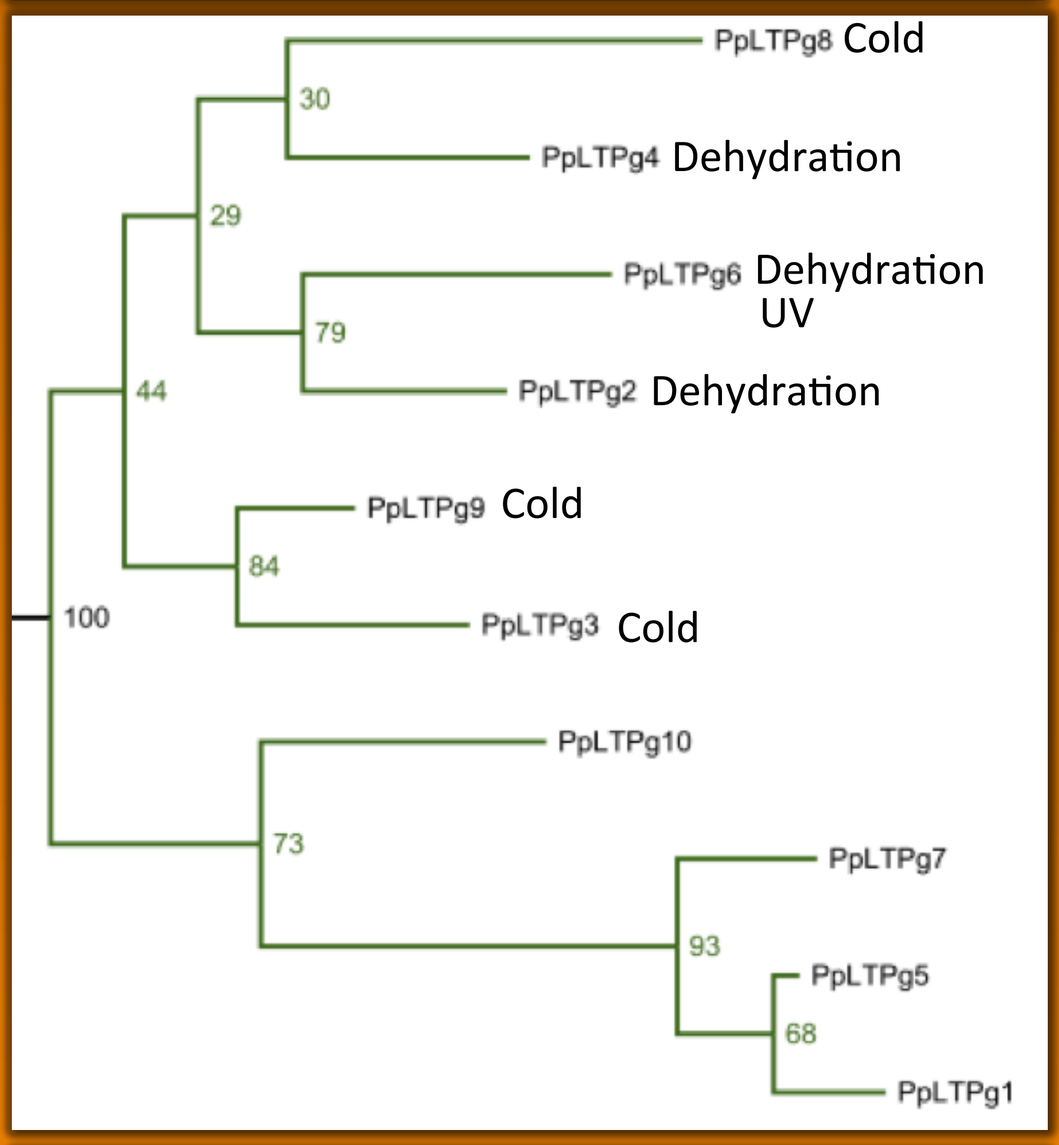

Figure 2: *A phylogenetic tree of the PpLTPg genes, constructed with the maximum likelihood method. The stresses that up-regulated the genes are indicated.*

Results

An up-regulation was seen in the gene *PpLTPg6* during UV-radiation stress, in *PpLTPg3*, *PpLTPg8* and *PpLTPg9* during cold stress, and in *PpLTPg2*, *PpLTPg4* and *PpLTPg6* during dehydration stress (figure 1). Additionally, a phylogenetic tree was constructed to show the relationship between the up-regulated genes (figure 2).

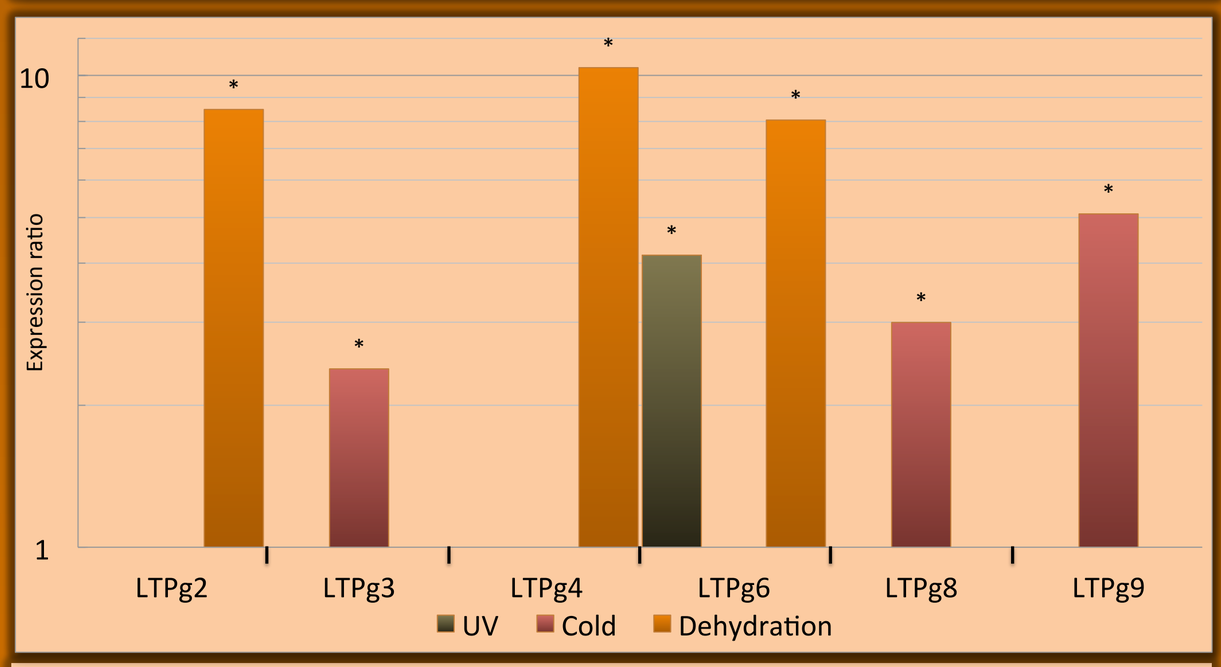

Figure 1: *An upregulation of the PpLTPg genes during various abiotic stresses. The values are normalized and compared to the control sample. The asterix (*) represent significance of p<0.05, calculated with permutation test.*

Conclusion

The up-regulated genes indicate that they might be important for the plants survival during the stressful conditions. Additional studies should aim to knock-out the up-regulated genes and compare the phenotypes with the wild-type.

Acknowledgement

I would like to thank my supervisors Johan Edqvist and Monika Edstam for their guidance and expertise, and my fellow students at the office for all the laughs.

Contact

Andrey Höglund

andho875@student.liu.se