

Background. Uptake of water by organisms including plants requires a lower internal water potential than the environment. Recent forecasts of the FAO (Food and Agriculture Organization of the United Nations) predict water shortages due to climate change and at the same time, increased water consumption for agricultural production of human nourishment. Therefore, insufficient water uptake during drought periods may not only limit plant growth, but also affect the fertilisation process, especially the male gametophyte (the pollen) which is highly sensitive to drought stress. Failure in fertilisation directly reduces crop yields due to lower production of seeds and fruits. So far, we do not know how pollen and the pollen tubes in particular, adapt to reduced water availability in the stigma tissue because they need to take up water continuously to elongate on their way to the ovule.

Aims. The primary aim of this project is to study molecular mechanisms of osmoregulation which enable the pollen to cope with drought stress conditions and is focused on dynamic metabolic adaptations that generate a low cytosolic water potential for continuous water uptake.

Methods. Combining turgor pressure with simultaneous metabolite concentration measurements in single, living cells and additional pollen metabolomics enable the detection of molecular processes during osmoregulation in pollen. Genetically encoded nanosensors based on Förster resonance energy transfer (FRET) technology and specific for selected metabolites will be introduced into the cytosol of pollen grains and tubes using the turgor pressure probe, thus allowing simultaneous monitoring of turgor pressure and changes in metabolite concentrations upon osmotic treatments. In addition, the change in metabolite concentrations and in consequence, the adaptation of metabolic pathways during osmotic stress will be studied using a metabolomics approach. Metabolic osmoregulation will be studied in the model organisms *Lilium* and *Arabidopsis* pollen, thus taking advantage of features of both pollen types, e.g. large size of *Lilium* pollen, transgenic *Arabidopsis* pollen expressing FRET nanosensors.

Expected Outcomes/Results. This study identifies and characterizes molecular components and processes, which are involved in pollen osmoregulation. Furthermore, establishing the combined pressure probe/FRET nanosensor method will allow investigating molecular details during pollen osmoregulation in particular but can be applied to other plant tissues, too. Finally, detailed knowledge on the molecular mechanisms during pollen osmoregulation may identify important markers for breeding drought-resistant crops to ensure sufficient crop yield for future generations.