Impact of sugars and sugar transporters on pollen tube growth in *Arabidopsis thaliana*

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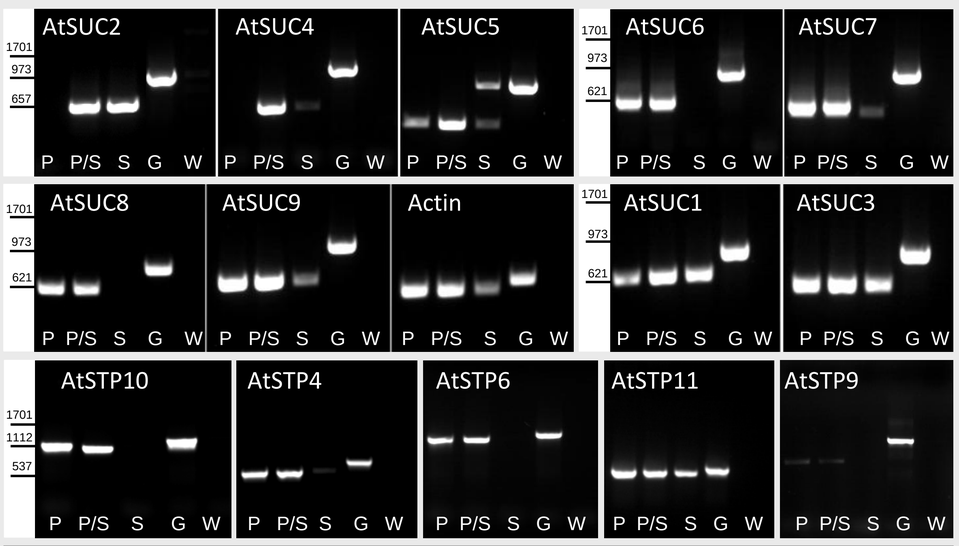
Research Question

In order to fertilize the egg cells pollen tubes have to cover distances ranging between some micrometers and 50 cm depending on the plant species. Elongating by tip growth they reach velocities up to 1 cm/h. This rapid growth requires a high amount of metabolic energy as well as a precise targeting towards the ovules. We studied the influence of extracellular sugars, which can serve both as nutrient and as signaling molecules, on the pollen tube growth of *Arabidopsis thaliana*.

Sugar transporters in pollen tubes

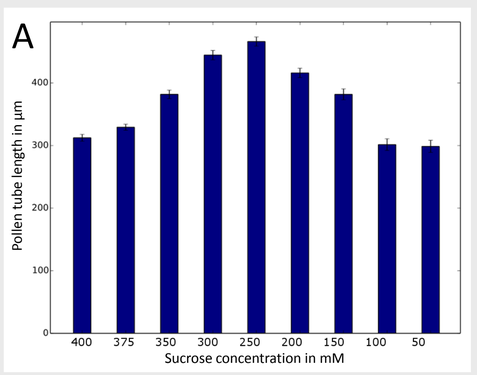
Via RT-PCR we could show that growing pollen tubes express several genes encoding sucrose and monosaccharide transporters. The expression of some of these genes seems to be enhanced in pollen tubes grown through the stigma compared to pollen germinated *in-vitro*.

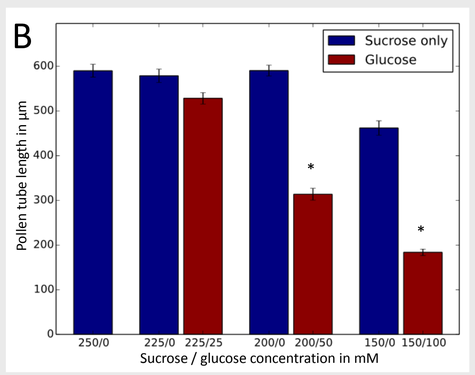


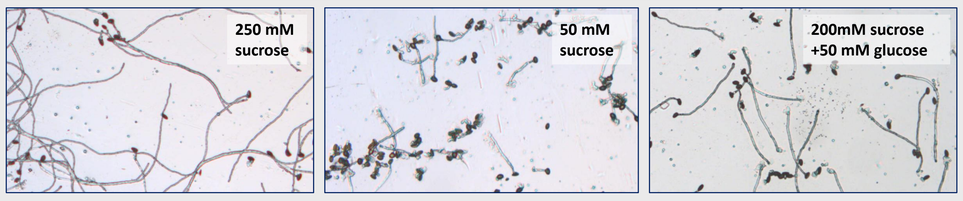
Expression analysis of *STP*s and *SUC*s in pollen tubes grown *in-vitro* (P), pollen germinated on stigmata (P/S) and stigmata without pollen (S). G: genomic DNA, W: water control

Sucrose and glucose show reverse effects

The expression of *SUC* and *STP* genes in pollen tubes suggests an important role for the substrates of the corresponding transport proteins during pollen tube growth. *In-vitro* pollen growth assays confirm that high concentrations of sucrose promote pollen tube growth **(A)**. However, the addition of glucose **(B)** to the germination medium leads to an inhibition of pollen tube elongation.

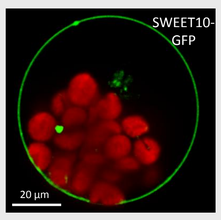






SWEETs may unload sugars into the style

Reporter gene analysis revealed the presence of at least three SWEETs in tissues of the style. In their function as sugar efflux carriers they might be responsible for unloading sugar into the transmitting tract of the style.

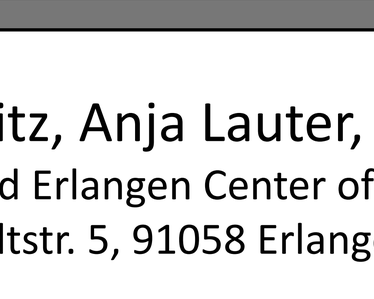


Glucose uptake measurements using FLIPs

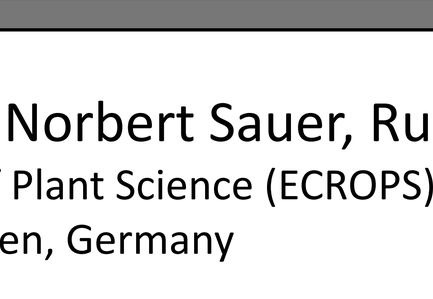
FLIPs are FRET based glucose nanosensors. Their change in the FRET ratio upon glucose binding can be used to analyse alterations in glucose concentrations in living cells. The expression of FLIPs in pollen tubes indicates that they take up glucose from the surrounding medium.

Low glucose → **530** / 480 nm = high FRET

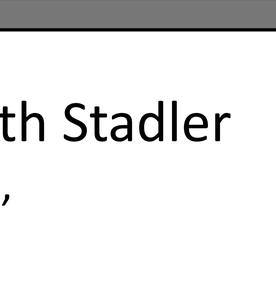
High glucose → 530 / **480** nm = low FRET



Emission spectrum of FLIPglu-600µ



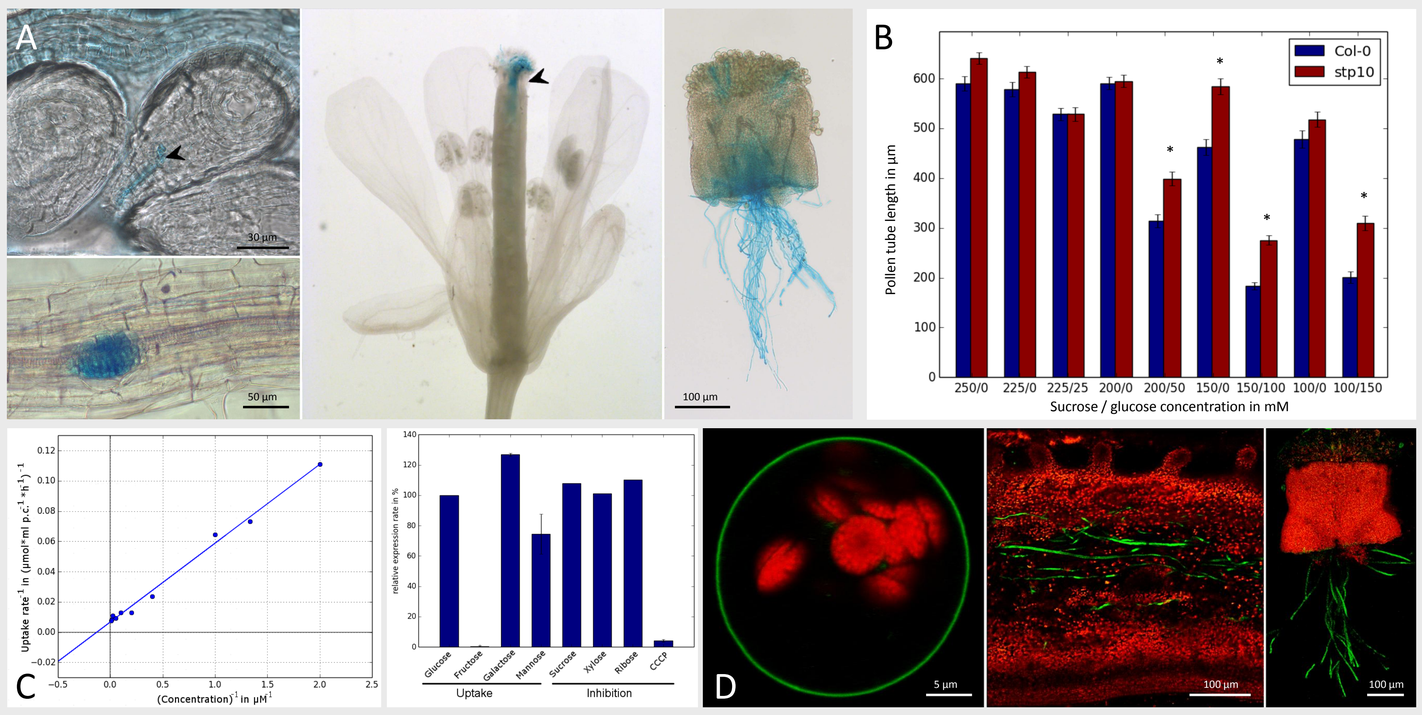
FRET ratio change upon glucose addition



FRET ratio mean values

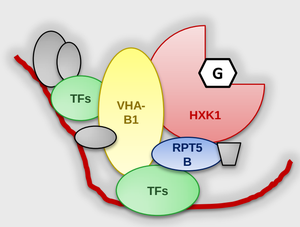
STP10 is involved in glucose uptake

RT-PCR data suggest the expression of *STP10*, a yet uncharacterized member of the STP-family, in pollen tubes. Reportergene analyses confirm the localization of STP10 at the PM of growing pollen tubes and also in side root buds **(A,D)**. Via expression in yeast we could show that STP10 is a high-affinity proton-coupled monosaccharide transporter **(C)**. *stp10* knock-out plants show a reduced sensitivity to glucose **(B)**.

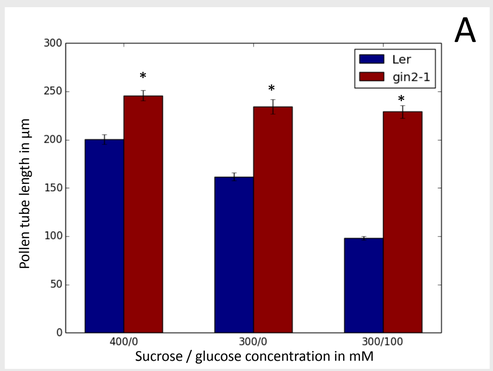


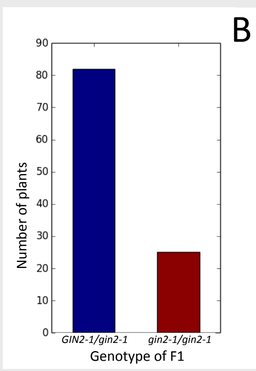
Glucose as signaling molecule

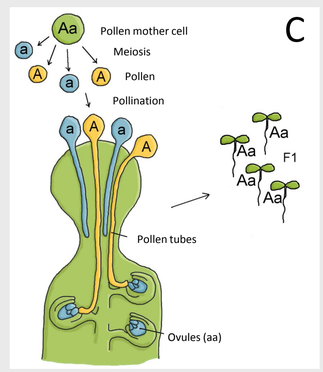
The inhibiting effect of glucose makes a function of this sugar as nutrient quite unlikely. As glucose does not interfere with pollen tube growth of glucose insensitive *gin2-1* mutant plants **(A)** it seems more probable that glucose is a signal for pollen tube growth or guidance. The reduced fertility of *gin2-1* mutant pollen **(B)** in segregation tests **(C)** indicates that glucose sensing is crucial for pollen tube function.



gin2-1 plants carry a mutation in the *Hexokinase1* gene







Summary

I. Several sugar transport proteins are localized in growing pollen tubes as well as in the surrounding maternal tissue.
II. FLIP-measurements and analysis of the *gin2-1* mutant indicate that glucose is taken up by growing pollen tubes, binds to HXK1 and serves as a signal molecule.
III. STP10 is an energy dependent high-affinity transporter for glucose, mannose and galactose, which is expressed in growing pollen tubes and side root buds.

Lit.: Büttner (2010), Rodriguez-Enriquez et al. (2012), Chen et al. (2010), Cho et al. (2006), Deuschle et al. (2006)