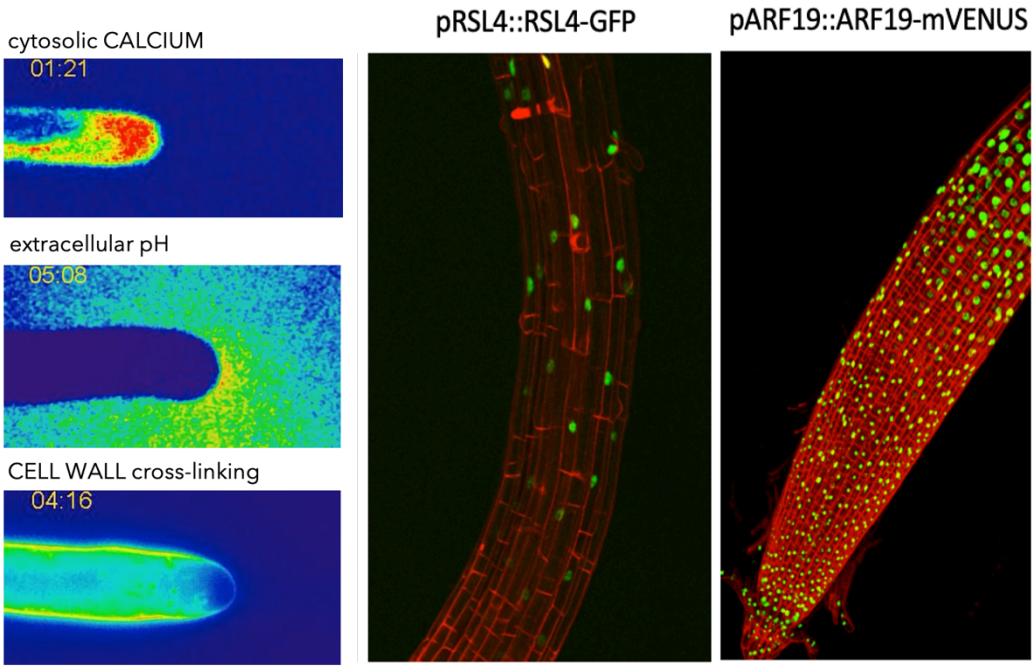
In the heath of the moment: study towards the mechanistic effect of temperature changes on root and root hair architecture and nutrient uptake

Description:

The main function of plant roots is the uptake of water and nutrients, the anchorage of the root in the soil and interaction with soil-living biota. Root hairs are tubular extensions of the root epidermis, and they greatly aid uptake processes by increasing the root's absorptive surface.
It is known that roots alter their growth when the root surrounding is changed (for example temperature), but far less mechanistic insight into the specific responses of the roots is available.

In this study we will verify the effect of changing temperature on the root system architecture, including the formation and morphology of root hairs. In addition, we will quantify the effect of altered temperature on the dynamics and responses of several cellular markers (intracellular calcium and pH, cell wall components, hormones, key transcription factors of root hair development) using genetically encoded fluorescent sensor lines and confocal microscopy (see image).



Methods:
- *In vitro* plant growth
- Plant growth analysis using microscopy and digital image analysis
- *Live cell imaging* using confocal microscopy of fluorescent markers

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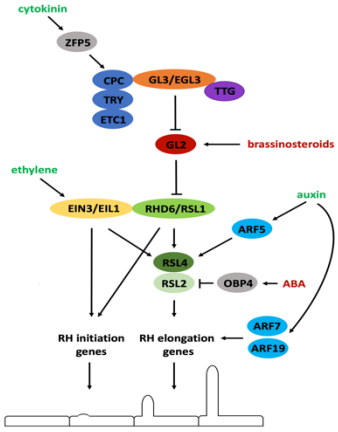
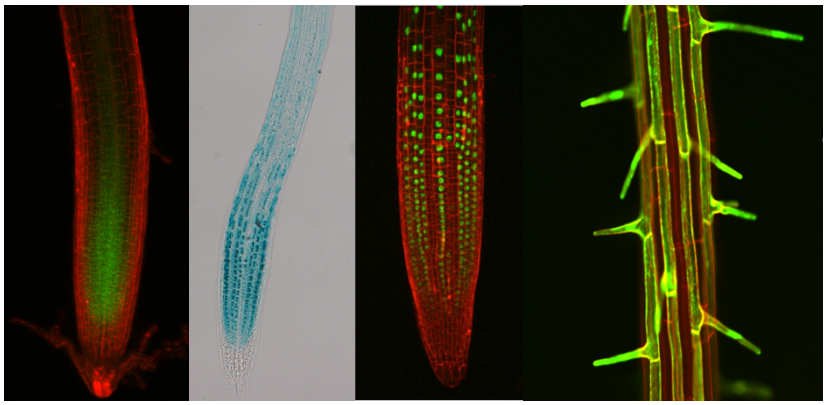
Regulation of root hair development through environmental changes

Description:

Root hairs are tubular extensions of the root epidermis, and they greatly aid the uptake of water and nutrients by largely increasing the root's absorptive surface. The underlying molecular pathway that determines which cells will form root hairs, how root hairs are initiated and how they are elongating mostly depends on the presence/absence of transcription factors that influence the expression of each other. In this pathway, hormones act as signalling molecules and as can be seen on the left scheme, they influence the expression of different genes within the developmental program.





Root hair development displays a high level of plasticity. Even subtle changes in the environment can be enough to lead to absent or extra root hairs, to increased root hair density or to changes in their length or morphology. Nevertheless, it is not known how changes in temperature (heat, cold), certain nutrient-deficiencies, presence of salt and the presence of rhizobacteria affect root hair development, nor what is the underlying genetic/molecular mechanism for these environmentally-induced growth changes.

In this study we will quantify the effect of changing environments on root hair development including growth rates, we will determine which hormones are involved in the response and we will identify which genes in the pathway have altered expression patterns using different marker lines coupled to state of the art microscopy (see right figures for the expression of 1 early, 2 middle and 1 late marker genes).

Methods:

- Plant growth analysis using digital image analysis
- Bright-field and confocal laser scanning microscopy

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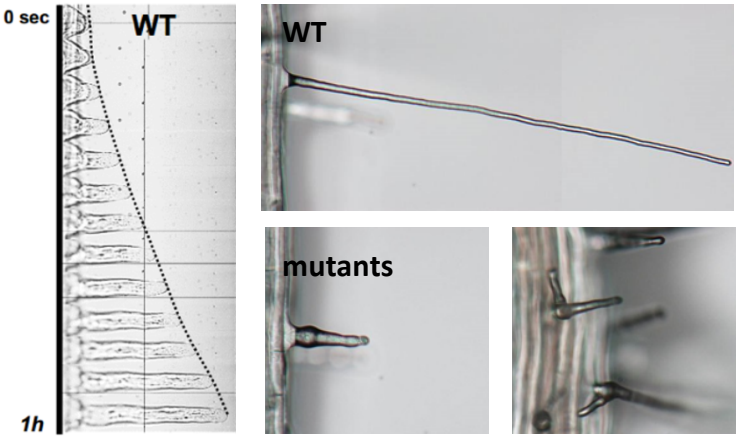
Characterization of protein targeting mutants and their involvement in root hair growth and environmental responses

Description:

Root hairs are tubular extensions of the root epidermis, and they greatly aid the uptake of water and nutrients by largely increasing the root's absorptive surface. Once epidermal cells have acquired root hair cell identity, they will form a small bulge in their cell wall which will subsequently elongate by very localised fusion of cell wall-rich vesicles with the plasma membrane at the root hair tip (see left figure). This specific called tipgrowth process occurs in a small number of plant cells, which includes root hairs and pollen tubes, and is highly controlled.

Based on root hair-specific expression of genes, we have recently isolated two mutants with shorter, bulged and even branched root hairs. Molecular analysis learned that they are not expressing two kinase proteins that are required for normal growth (see right figures wildtype (WT) and mutants). Proteome analysis identified several targets of both kinases that are involved in targeting of proteins to the root hair tip, and mutants in these genes show altered root hair growth.



In this study we will characterize the role of these kinase-targets in the highly controlled deposition of new cell wall material at the root hair tip. This will involve 1) detailed growth analysis using digital image analysis, 2) identification of their expression pattern, subcellular localisation and the effect of overexpression using molecular biology approaches coupled to state of the art microscopy, and 3) identification of their involvement in root hair growth responses to changes in the root's environment using digital image analysis.

Methods:

- Plant growth analysis using digital image analysis
- Molecular biology/cloning/transformation of bacteria and plants
- Bright-field and confocal laser scanning microscopy

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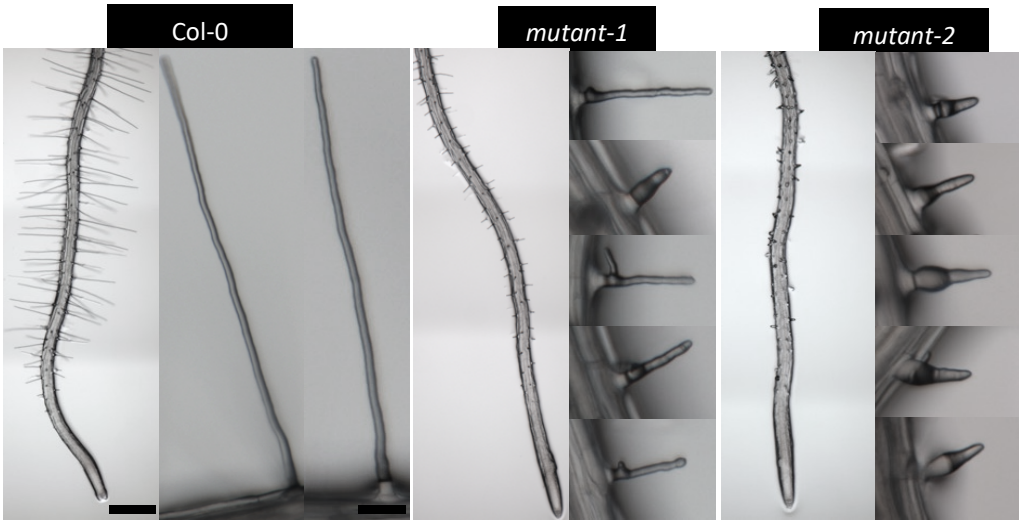
Regulation of root hair growth by RALF peptides and receptor-like kinases

Description:
Root hairs are tubular extensions of the root epidermis, and they greatly aid the uptake of water and nutrients by largely increasing the root's absorptive surface. Once epidermal cells have acquired root hair cell identity, they will form a small bulge in their cell wall which will subsequently elongate by very localised fusion of cell wall-rich vesicles with the plasma membrane at the root hair tip (see left figure). This specific called tipgrowth process occurs in a small number of plant cells, which includes root hairs and pollen tubes, and is highly controlled. Receptor-like kinases (RLKs) located in the apical plasma membrane monitor the status of the cell wall, and they do so by binding Rapid ALkalinization Factors (RALFs), small proteins that are secreted. Once bound by RLKs, they initiate a signaling cascade that triggers stiffening or loosening of the cell wall.

We have performed a transcriptome analysis of RALF-induced changes in gene expressions of wild type plants and several RLK-mutants (see image). This identified several genes/proteins with a putative direct controlling role during root hair growth.



In this study we will characterize the role of these genes/proteins in the highly controlled deposition of new cell wall material at the root hair tip. This will involve 1) detailed growth analysis using digital image analysis of their mutants compared to the wild type, 2) identification of their expression pattern, subcellular localisation and the effect of overexpression using molecular biology approaches (cloning) coupled to state of the art microscopy, and 3) identification of their involvement in root hair growth responses to changes in the root's environment using digital image analysis.

Methods:
- Plant growth analysis using digital image analysis
- Molecular biology/cloning/transformation of bacteria and plants
- Bright-field and confocal laser scanning microscopy

Research group: **IMPRES**

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