

233 *Etr1-1* receptor mutant gene reduces ethylene sensitivity in ornamental flowering plants

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Use of the dominant ethylene receptor mutant gene, *etr1-1*, from *Arabidopsis* has proved to be an effective way in creating ethylene insensitive ornamental flowering plants.

Effective regeneration and transformation systems for popular potted flowering plants *Campanula* (carpathian harebell) and *Kalanchoë* were established in order to introduce *etr1-1* mutant gene. The construct containing the *etr1-1* gene from *Arabidopsis thaliana* under the control of the flower specific *fbp1*-promoter from *Petunia* was used to obtain *Campanula carpatica* Jacq. and *Kalanchoë blossfeldiana* Poeln. potted plants with ethylene insensitive flowers. Flowering T0 lines were tested for their ethylene sensitivity by exposing them to 2 µl l⁻¹ ethylene. Wild type (non-transgenic) *Kalanchoë* and *Campanula* individual flowers wilted within 2 and 3 days, respectively. The best transgenic line of *Campanula* plant flowered up to 27 days in ethylene, while the best *Kalanchoë* line had only 1/3 wilted flowers after 10 days of continuous ethylene exposure. T1 progenies of both species, crosses between transgenic and wild type plant, showed stable dominant inheritance and expression of *etr1-1*, which makes the plants useful for breeding program.

235 A receptor-like kinase functions in a pathway with ROP GTPases to promote the polarization of asymmetric cell divisions in the maize leaf epidermis

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Asymmetric cell division is an important mechanism for generation of cellular diversity in plants as in other eukaryotes, but very little is known about the mechanisms governing division asymmetry or how division polarity is coordinated among cells in a tissue. Stomatal complexes in maize form through a sequence of coordinated, asymmetric divisions leading to the formation of a guard cell pair flanked by a pair of subsidiary cells. The asymmetric subsidiary mother cell (SMC) division is polarized toward the adjacent guard mother cell (GMC), apparently under the influence of a GMC-derived signal. We discovered an LRR-RLK, PAN1, that promotes the premitotic polarization of SMCs. PAN1 becomes localized in SMCs at sites of GMC contact before the appearance of other polarity markers. A second gene of as yet unknown identity, *pan2*, is implicated as a component of a PAN1 pathway, as are the nearly identical Rho-family GTPases ROP2 and ROP9. Mutations reducing ROP2/9 protein levels cause weak SMC polarity defects, and strongly enhance the *pan1* SMC polarity defect. ROP2/9 physically associates with PAN1 and localizes within premitotic SMCs at GMC contact sites in a PAN1-dependent manner, suggesting that these ROPs function downstream of PAN1. PAN1 has an inactive kinase domain, but is required for the accumulation of a membrane-associated phosphoprotein, suggesting a function for PAN1 in signal transduction. Proteomic strategies are underway to identify this phosphoprotein and other proteins that function together with PAN1 to promote SMC polarization. Together, our findings implicate PAN1, PAN2 and ROP GTPases as components of a pathway transmitting a GMC-derived cue that polarizes SMCs in preparation for their asymmetric division. Funded by NSF grants IOS-0843704 to LGS, IBN-0420226 to JEF, and DBI-0501862 to AWS.

234 A domain swap approach reveals the plant wall-associated kinase 1 as a receptor of oligogalacturonides

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Oligogalacturonides (OGs) released from the plant cell wall pectin are active as both damage-associated molecular patterns and regulators of plant growth. Members of the Wall-Associated Kinase (WAK) family bind these oligosaccharides *in vitro* and are candidate receptors. A chimeric receptor approach was used to define the role of WAK1. We first defined the appropriate design and demonstrated that the *Arabidopsis* pattern recognition receptor (PRR) EFR is amenable to the construction of functional chimeric receptors that carry the ectodomain of another PRR, FLS2. After, the analysis of EFR-derived chimeras, carrying the extracytoplasmic or the kinase domain of WAK1 revealed that WAK1 senses OGs *in vivo* and, respectively, triggers a defense response that mirrors that of OGs. Transgenic plants overexpressing WAK1 are more sensitive to OG signal.

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236 The diverse roles of NB-LRR proteins in plant innate immunity

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Most characterised plant nucleotide-binding and leucine rich repeat (NB-LRR) proteins function as immune receptors. Interestingly, we have identified a tomato NB-LRR protein that functions as a signalling component downstream of both extracellular and intracellular immune receptors. This protein was designated NB-LRR protein required for HR-associated cell death 1 (NRC1; Gabriëls et al., 2007, Plant J. 50). In a yeast two-hybrid screen a chloroplastic protein was identified as NRC1-binding protein 1 (NBP1). Both proteins were fused to fluorescent proteins and subjected to co-localisation studies by using confocal microscopy. The role of NBP1 in the signalling cascade of NRC1 is currently investigated. Tomato stably expressing *NRC1* RNAi constructs are being generated and will be tested for loss of resistance to a broad spectrum of plant pathogens in order to assess whether NRC1 is indeed a general key regulator of plant innate immunity.

The potato NB-LRR protein Rx is an immune receptor that confers immunity to potato virus X. Rx localises not only to the cytoplasm, but also to the nucleus despite the absence of a discernible nuclear localisation signal (NLS). In the cytoplasm Rx associates with a RanGTPase-activating protein 2 (RanGAP2), which is required for resistance to PVX (Tameling and Baulcombe 2007, Plant Cell 19; Sacco et al., 2007, Plant J. 52). RanGAPs are highly conserved in eukaryotes and are required for the regulation of nucleo-cytoplasmic trafficking. Co-expression studies in *Nicotiana benthamiana* revealed that overexpression of RanGAP2 (in the cytoplasm) attenuates the nuclear accumulation of Rx and enhances Rx-mediated defence. The opposite was observed when the Rx-interacting domain of RanGAP2 was fused to an NLS. Hence, co-expressed Rx hyperaccumulated in the nucleus, which resulted in an abolished immunity to PVX. Our data show that the ratio between the Rx pool in the cytoplasm and nucleoplasm dictates the initiation of defence signalling. This research is supported by NWO, EU 6th framework program BIO-EXPLOIT and TTI Green Genetics.