CROSS-TALK IN LIGHT ACCLIMATION AND DISEASE RESISTANCE IN *ARABIDOPSIS THALIANA*

Saijaliisa Kangasjärvi[1], Eevi Rintamäki[1], Jose Manuel Perez-Perez[2], Jose Luis Micol[2], Engelbert Weis[3], Jaakko Kangasjärvi[4] and Eva-Mari Aro[1]

[1] Department of Biochemistry and Food Chemistry, University of Turku, 20014 Turku, Finland, [2] Department of Genetics, University of Miguel Hernandez, 03202 Elche, Spain [3] Institute for Botany, University of Münster, 48149 Münster, Germany,

Study of cross-talk in chloroplast signaling

Besides inducing short-term adjustments in photosynthetic complexes (1, 2), light modulates the developmental programs and stress resistance of plants through chloroplast signals that modulate transcriptional activity in the nucleus. Chloroplasts can act as environmental sensors, since their biosynthetic pathways are readily imbalanced upon environmental challenges (3, 4). The final acclimation responses are, however, influenced by cross-talk among signalling networks, including photoperiodic signals and hormonal regulation (5-7).

Goals of this work:

- to understand the role of chloroplast redox regulation in plant acclimation
- to identify mechanistic connections between chloroplast signaling and disease resistance pathways in plants
- to reveal how vascular tissues modulate stress tolerance in C3 plants

Antioxidant enzymes modulate chloroplast signals and contribute to developmental processes in plants

Chloroplast signaling is tightly connected with the redox balance of photosynthetic electron transfer components and formation of reactive oxygen species (ROS).

Transcript and metabolite profiling revealed that NADPH-thioredoxin reductase (NTRC) forms a connection between chloroplast thiol redox regulation and control of photoperiodic growth in Arabidopsis (4, 5).

Chloroplast ascorbate peroxidases (sAPX and tAPX) scavenge hydrogen peroxide, and are important for photoprotection of mesophyll cell chloroplasts during greening (8).

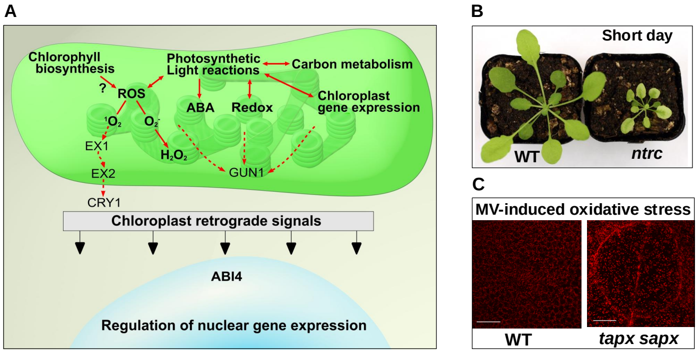

Figure 1.
A) Schematic overview of components that mediate chloroplast retrograde signals (3).
B) Retarded growth and variegated pigmentation of *ntrc* plants grown in short eight-hour photoperiod (5).
C) Photobleaching of mesophyll cell chloroplasts in *tapx sapx* double mutant leaves. Confocal laser scanning microscope images of chlorophyll autofluorescence from seedlings germinated in the presence of methylviologen (MV) to cause photo-oxidative stress in chloroplasts (8).

Bundle sheath cells have specialized roles in systemic light acclimation and regulation of leaf development

Perception of light stress initiates systemic signals that spread through the vasculature to confer stress resistance in non-exposed parts of the plant. Such long-distance signalling functions are related to unique characteristics of ROS and their detoxification in bundle sheath cells.

Biosynthesis of ABA and pronounced accumulation of hydrogen peroxide (H_2O_2) in bundle sheath cells promotes systemic signals under photo-oxidative stress.

Mutants deficient in RETICULATA (RE) exhibit reticulate leaf pigmentation and accumulate H2O2 in the vascular leaf tissue even in the absence of light stress (7).

RE is a chloroplast protein of unknown function. Transcript and metabolite profiling, analysis of ROS metabolism in single double mutants and identification of RE-interactors is underway to reveal the functional significance of RE (JM Perez-Perez and JL Micol; unpublished).

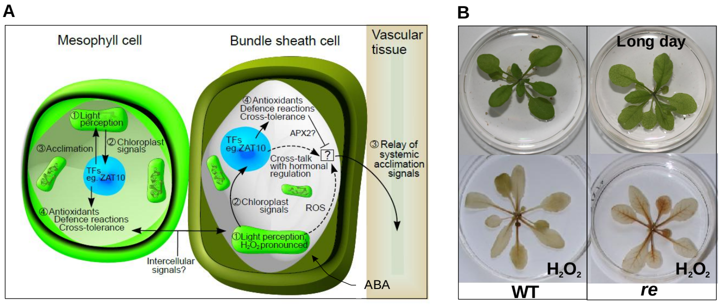

Figure 2.
A) Model depicting systemic signalling in high-light-exposed leaves (3).
B) The *re* mutant has malformed mesophyll tissue and accumulates H_2O_2 in the vascular leaf tissue even in the absence of light stress (7).

PP2A subunit B as a regulator of disease resistance and light acclimation in Arabidopsis

Knock-down mutants for a cytosolic protein phosphatase 2A (PP2A) regulatory subunit B' show constitutive disease resistance and cell death under moderate light intensity. Growth under high light rescues this phenotype (9).

Currently our work mainly focuses on the interaction partners and target processes of PP2A-B. A combination of biochemical pull-down assays and phenotypic analysis of Arabidopsis mutants is expected to specify the mechanisms of PP2A function in plants.

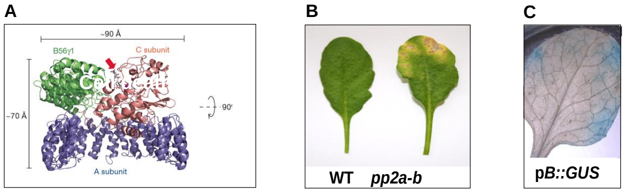

Figure 3.
A) Structure of the trimeric human PP2A holoenzyme consisting of a catalytic subunit C and regulatory subunits A and B.
B) Cell death lesions in *pp2a-b* leaves (9).
C) Promoter-driven GUS analysis demonstrating that the *PP2A-B* gene is active in patches, which resemble the yellowing lesions in *pp2a-b* mutant leaves.

Modulation of defence pathways through light-induced ROS formation in chloroplasts

ROS networks have vital signaling roles under adverse challenges, such as plant pathogens (6, 7, 9, 10).

Upon recognition of an invading pathogen, extra-cellular ROS induce defence gene expression, whereas chloroplastic ROS seems to promote the onset of cell death (6).

Light stress and chloroplast ROS were found to repress the expression of Cysteine-rich Receptor-like Kinases (CRKs), many of which elicit defence reactions upon infection. This inhibitory action presumably involves ABA and negative crosstalk with salicylic acid signaling (6).

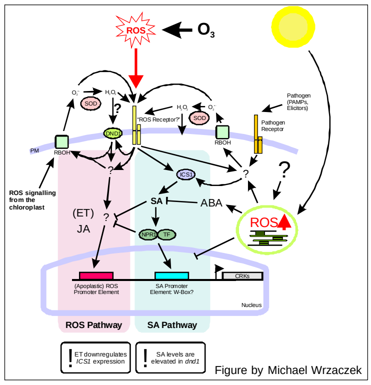

Figure 4. Scheme of ROS-dependent defence signaling pathways in Arabidopsis (6).

References to our work 2008-2010:

1. Tikkanen, M., Grieco, M., Kangasjärvi, S. and Aro, E.-M. (2010): **Thylakoid protein phosphorylation in higher plant chloroplasts optimises electron transfer under fluctuating light.** Plant Physiology, 152: 723-735.

2. Tikkanen, M., Nurmi, M., Kangasjärvi, S. and Aro, E.-M. (2008) **Core protein phosphorylation facilitates the repair of photodamaged photosystem II at high light.** Biochimical and Biophysical Acta. 1777:1432-1437.

3. Kangasjärvi, S., Nurmi, M., Tikkanen, M. and Aro, E.-M. (2009) **Cell-specific mechanisms and systemic signalling as emerging themes in light acclimation of C3 plants.** Plant Cell Environ 32:1230-1240.

4. Rintamäki, E., Lepistö, A. and Kangasjärvi, S. (2009) **Implication of chlorophyll biosynthesis on chloroplast-to-nucleus retrograde signaling.** Plant Signaling & Behavior 4: 545-547.

5. Lepistö A, Kangasjärvi S, Luomala EM, Brader G, Sipari N, Keranen M, Keinänen M, Rintamäki E (2009) **Chloroplast NADPH Thioredoxin Reductase Interacts with Photoperiodic Development in *Arabidopsis thaliana*.** Plant Physiol 149:1261-1276.

6. Wrzaczek, M. Brosché, M., Salojärvi, J., Kangasjärvi, S., Idänheimo, N., Mersmann, S., Robatzek, S., Karpinski, S., Karpinska, B.and Kangasjärvi, J. (2010) **Transcriptional regulation of the CRK/DUF26 group of Receptor-like protein kinases by ozone and plant hormones in Arabidopsis.** Submitted.

7. Overmyer, K., Kollist, H., Tuominen, H., Betz, C., Langebartels, C, Wingsle, G., Kangasjärvi, S., Brader, G., Mullineaux, P., and Kangasjärvi, J (2008) **Complex phenotypic profiles leading to ozone sensitivity in *Arabidopsis thaliana* mutants.** Plant Cell Environ31: 1237-1249.

8. Kangasjärvi, S, Lepistö, A, Hännikäinen, K., Luomala, E.-M., Aro, E.-M. and Rintamäki, E. (2008) **Dissection of the role of chloroplast stromal and thylakoid-bound ascorbate peroxidases in plant stress responses**. Biochem J 412:275-285.

9. Kangasjärvi, S. Tikkanen, M, Wrzaczek, M.,Rahikainen, M., Scharte, J., Holmström, M., Stauffer, J., Hiltunen, H.-M., Rips, S., Mulo, P., Weis, E., von Schaewen, A., and Aro, E-M. (2010) **Regulatory subunit B of protein phosphatase 2A is required for accurate leaf development and regulation of defence responses in Arabidopsis.** Manuscript.

10. Broche, M., Overmyer, K., Wrzaczek, M., Kangasjärvi, J and Kangasjärvi, S. (2010) **Stress signaling: reactive oxygen species.** In: Bohnert, H., Govindjee, Pareek, A. and Sopory, S. (Eds.): Abiotic Stress Adaptation in Plants: Physiological, Molecular and Genomic Foundation-. DOI: 10.1007/978-90-481-3112-9_5.