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Data article

Leaf apoplastic proteome composition in UV-B treated *Arabidopsis thaliana* mutants impaired in extracellular glutathione degradation

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ABSTRACT

In plants, environmental perturbations often result in oxidative reactions in the apoplastic space, which are counteracted for by enzymatic and non-enzymatic antioxidative systems, including ascorbate and glutathione. However, the occurrence of the latter and its exact role in the extracellular space are not well documented. In *Arabidopsis thaliana*, the gamma-glutamyl transferase isoform GGT1 bound to the cell wall takes part in the so-called gamma-glutamyl cycle for extracellular glutathione degradation and recovery, and may be implicated in redox sensing and balance.

In this work, oxidative conditions were imposed with UV-B radiation and studied in redox altered *ggt1* mutants. Elevated UV-B has detrimental effects on plant metabolism, plasma membranes representing a major target for ROS generated by this harmful radiation. The response of *ggt1* knockout *Arabidopsis* leaves to UV-B radiation was assessed by investigating changes in apoplastic protein composition.

We then compared the expression changes resulting from the mutation and from the UV-B treatment. Rearrangements occurring in apoplastic protein composition suggest the involvement of hydrogen peroxide, which may ultimately act as a signal. Other important changes related to hormonal effects, cell wall remodeling, and redox activities are also reported. We argue that oxidative stress conditions imposed by UV-B and by disruption of the gamma-glutamyl cycle result in similar stress-induced responses, to some degree at least. Data shown here are associated with the

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article from Trentin et al. (2015) [1]; protein data have been deposited to the PRIDE database (Vizcaíno et al., 2014) [2] with identifier PXD001807.

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Specifications Table

Subject area	Plant Physiology and Biochemistry
More specific subject area	Glutathione metabolism
Type of data	MS data and annotations, spectrophotometric and chromatographic data
How data was acquired	i-TRAQ labelled peptides were analysed using mass spectrometry (LTQ Orbitrap, Thermo Scientific)
Data format	Analysed output data
Experimental factors	Apoplastic fluids (or ECWF, Extra-Cellular Washing Fluid) were obtained by the infiltration/centrifugation method
Experimental features	Depending on the purpose of analysis, different infiltration buffers were used for antioxidant measurements or proteome composition analysis.
Data source location	NOT APPLICABLE
Data accessibility	Proteomic data are stored and available in a public repository (PRIDE database, PXD001807, url: ⟨http://proteomecentral.proteomexchange.org/dataset/PXD001807⟩)

Value of the data

- Apoplastic proteomes from *A. thaliana* wt and ggt1- knockout mutants are compared for functional characterization of the cell-wall bound gamma-glutamyl transferase/transpeptidase GGT1 enzyme.
- Effects of UV-B radiation on the extracellular protein composition are also reported.
- Quantitative proteomics was performed by iTRAQ labelling.
- Results point to a role for apoplastic GGT1 in redox sensing/signaling.

1. Experimental design

A major aim of this analysis was to obtain information on the significance of the enzyme gamma-glutamyl transferase (GGT) in the response to oxidative conditions. Since the apoplastic isoform GGT1 is extracellular and cell-wall bound, we hypothesised that disrupting this enzyme's activity would result in altered redox conditions in the apoplast, that may affect the overall response to oxidative stress conditions starting from the apoplast. To this regard, UV-B radiation is known to induce oxidative damage to plasma membranes and originate ROS in the apoplast.

Therefore, we used a *ggt1* mutant line that had been previously characterized [3,4], and imposed a UV-B treatment. In this way, we generated four experimental conditions: 1) untreated, wildtype; 2) untreated, *ggt1* mutant; 3) UV-B treated, wildtype; and 4) UV-B treated, *ggt1* mutant.

Finally, we obtained the extracellular washing fluid (ECWF) with the aim to gain the following information: i) the effect of UV-B treatment on each genotype; ii) differential apoplastic protein composition in *ggt1 vs* . wildtype; iii) possible differences in the behavior of the *ggt1* mutant and the wildtype under UV-B.

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