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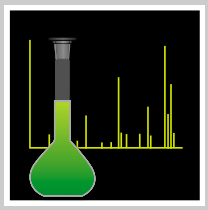

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Cell Growth Processes in *Arabidopsis thaliana* are Modified by Flavonols

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The phenylpropanoid pathway (PPP) is a biosynthetic process ubiquitously found in plants. A large array of compounds is produced by the PPP including lignin for wood formation, phytoalexins which serve as a line of defense against pathogens, or anthocyanins (a group of flavonoids) which protect the plant from UV-induced damage. Flavonols form a second subgroup of flavonoids and are attracting increasing attention due to their suggested health-beneficial effects.

Most of the flavonols accumulating in the model plant *Arabidopsis thaliana* are glycosylated with either glucose or rhamnose. The mutant plant *rol1-2* is affected in the synthesis of rhamnose, shows a modified flavonol glycoside accumulation, and exhibits a distorted cell growth phenotype. UHPLC-HR-MS/MS analysis was used to identify and quantify the exact flavonol glycoside composition. The biological significance of this change in the flavonol glycosylation profile was assessed by genetic means. Blocking flavonol biosynthesis in the *rol1-2* mutant, by introducing mutations in genes coding for enzymes active in the PPP pathway, leads to suppression of the *rol1-2* cell growth phenotype. Hence, flavonols accumulating in the *rol1-2* mutant interfere with proper cell development. One of several potential modes of action of flavonols is to modify the transport activity of auxin, a major plant hormone that affects numerous plant developmental processes. Auxin is not produced by all but only a restricted number of cells and must be transported to the target tissues. While the *rol1-2* mutant shows altered transport and accumulation of auxin in the plant shoot, this effect is reverted to wild-type levels by blocking flavonol biosynthesis.

Hence, the *rol1-2* mutant of *Arabidopsis thaliana* shows an alteration in the flavonol glycoside accumulation profile which was analyzed in detail by UHPLC-HR-MS/MS. The observed changes have a direct effect on cell growth processes in the *rol1-2* mutant, which thus can serve as a model system to investigate in detail the mode of action of flavonols on plant development.

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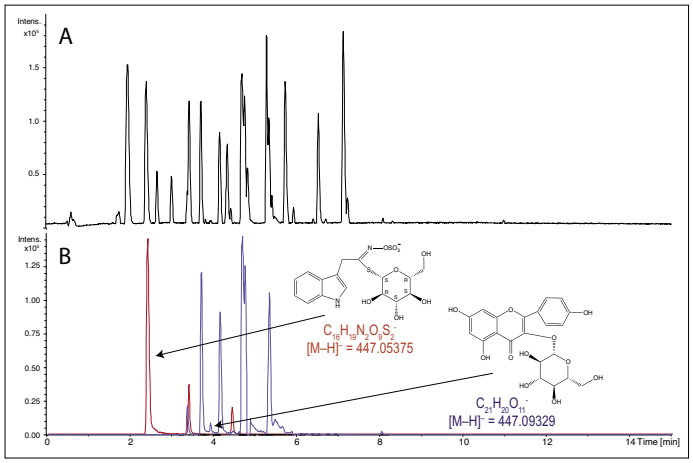
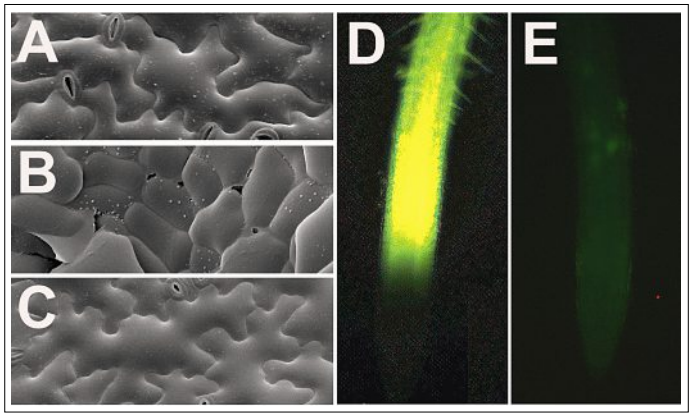


UHPLC-HR-ESI-MS of *A. thaliana* shoot extract. (A) Base peak chromatogram of the extract. (B) Selected high-resolution extracted ion chromatograms (± 0.005 m/z width) of glucosinolates (red, m/z 447.05375; 477.06431) and flavonoids (blue, m/z 431.09837; 447.09329; 463.08820; 477.10385; 489.10385; 577.15628; 593.15119; 609.14611; 623.16176; 739.20910; 755.20402; 771.19893) used for quantitative determination.



Flavonol-induced cell growth phenotype. Wild-type cotyledons of *A. thaliana* develop puzzle-shaped epidermal cells (A). In *rol1-2* mutants, they are brick-shaped (B), an effect that is suppressed in the *rol1-2 tt4* double mutant (C) which lacks chalcone synthase, one of the enzymes of the PPP essential for flavonoid biosynthesis. Flavonoids can be visualized in planta by staining with diphenylboric acid 2-aminoethyl ester in the root of the *rol1-2* (D) but not in the flavonoid-less *rol1-2 tt4* mutant (E).

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