

1. 25%

Hypersensitive response (HR) is a form of programmed cell death (PCD) and the primary immune response that prevents pathogen invasion in plants. Here, we show that a microRNA *miR164* and its target gene *NAC4* (*At5g07680*), encoding a NAC transcription factor, play essential roles in the regulation of HR PCD in *Arabidopsis thaliana*. Cell death symptoms were noticeably enhanced in *NAC4*-overexpressing (*35S:NAC4*) and *mir164* mutant plants in response to avirulent bacterial pathogens. *NAC4* expression was induced by pathogen infection and negatively regulated by *miR164* expression. *NAC4*-binding DNA sequences were determined by *in vitro* binding site selection using random oligonucleotide sequences. Microarray, chromatin immunoprecipitation and quantitative real time polymerase chain reaction (qRT-PCR) analyses, followed by cell death assays in protoplasts, led to the identification of *NAC4* target genes *LURP1*, *WRKY40* and *WRKY54*, which act as negative regulators of cell death. Our results suggest that *NAC4* promotes hypersensitive cell death by suppressing its target genes and this immune process is fine-tuned by the negative action of *miR164*. (Lee et al., *New Phytologist* (2017) 214, 343-360)

- (1) 給此研究下一個英文標題
- (2) 以下除了專有名詞外，須用中文敘述
 - (a) 此研究在研究什麼樣的問題？
 - (b) 此研究用了哪些研究方法，這些研究得到什麼樣的結論？
 - (c) 此研究最主要的結論是什麼？

2. 25%

Isoprene is a well-studied volatile hemiterpene that protects plants from abiotic stress through mechanisms that are not fully understood. The antioxidant and membrane stabilizing potential of isoprene are the two most commonly invoked mechanisms. However, isoprene also affects phenylpropanoid metabolism, suggesting an additional role as a signalling molecule. In this study, microarray-based gene expression profiling reveals transcriptional reprogramming of *Arabidopsis thaliana* plants fumigated for 24 h with a physiologically relevant concentration of isoprene. Functional enrichment analysis of fumigated plants revealed enhanced heat- and light-stress-responsive processes in response to isoprene. Isoprene induced a network enriched in ERF and WRKY transcription factors, which may play a role in stress tolerance. The isoprene-induced up-regulation of phenylpropanoid biosynthetic genes was specifically confirmed using quantitative reverse transcription polymerase chain reaction. These results support a role for isoprene as a signalling molecule, in addition to its possible roles as an antioxidant and membrane thermoprotectant. (Harvey and Sharkey, *Plant, Cell and environments* (2016) 39, 1251-1263)

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見背面

3. 25%

The molecular mechanism that initiates the synthesis of starch granules is poorly understood. Here, we discovered two plastidial proteins involved in granule initiation in *Arabidopsis thaliana* leaves. Both contain coiled coils and a family-48 carbohydrate binding module (CBM48) and are homologs of the PROTEIN TARGETING TO STARCH (PTST) protein; thus, we named them PTST2 and PTST3. Chloroplasts in mesophyll cells typically contain five to seven granules, but remarkably, most chloroplasts in *ptst2* mutants contained zero or one large granule. Chloroplasts in *ptst3* had a slight reduction in granule number compared with the wild type, while those of the *ptst2 ptst3* double mutant contained even fewer granules than *ptst2*. The *ptst2* granules were larger but similar in morphology to wild-type granules, but those of the double mutant had an aberrant morphology. Immunoprecipitation showed that PTST2 interacts with STARCH SYNTHASE4 (SS4), which influences granule initiation and morphology. Overexpression of PTST2 resulted in chloroplasts containing many small granules, an effect that was dependent on the presence of SS4. Furthermore, isothermal titration calorimetry revealed that the CBM48 domain of PTST2, which is essential for its function, interacts with long maltooligosaccharides. We propose that PTST2 and PTST3 are critical during granule initiation, as they bind and deliver suitable maltooligosaccharide primers to SS4 (Seung et al. Plant Cell Jul 2017, 29 (7) 1657-1677).

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4. 25%

Proline (Pro) accumulation in plants is a well-documented physiological response to osmotic stress caused by drought or salinity. In *Arabidopsis* (*Arabidopsis thaliana*), the stress and ABA-induced $\Delta 1$ -PYRROLINE-5-CARBOXYLATE SYNTHETASE1 (*P5CS1*) gene was previously shown to control Pro biosynthesis in such adverse conditions. To identify regulatory factors that control the transcription of *P5CS1*, Y1H screens were performed with a genomic fragment of *P5CS1*, containing 1.2-kb promoter and 0.8-kb transcribed regions. The myeloblastosis (MYB)-type transcription factors PHOSPHATE STARVATION RESPONSE1 (PHR1) and PHR1-LIKE1 (PHL1) were identified to bind to *P5CS1* regulatory sequences in the first intron, which carries a conserved PHR1-binding site (P1BS) motif. Binding of PHR1 and PHL1 factors to P1BS was confirmed by Y1H, electrophoretic mobility assay and chromatin immunoprecipitation. Phosphate starvation led to gradual increase in Pro content in wild-type *Arabidopsis* plants as well as transcriptional activation of *P5CS1* and PRO DEHYDROGENASE2 genes. Induction of *P5CS1* transcription and Pro accumulation during phosphate deficiency was considerably reduced by *phr1* and *phl1* mutations and was impaired in the ABA-deficient *aba2-3* and ABA-insensitive *abi4-1* mutants. Growth and viability of *phr1phl1* double mutant was significantly reduced in phosphate-depleted medium,

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while growth was only marginally affected in the *aba2-3* mutants, suggesting that ABA is implicated in growth retardation in such nutritional stress. Our results reveal a previously unknown link between Pro metabolism and phosphate nutrition and show that Pro biosynthesis is target of cross talk between ABA signaling and regulation of phosphate homeostasis through PHR1- and PHL1-mediated transcriptional activation of the *P5CSI* gene (Aleksza et al., Plant Physiology, 2017, 175 (1) 555-567).

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試題隨卷繳回