SUBVERSION OF HOST TRANSCRIPTION BY MICROBIAL EFFECTORS

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Plant defense responses are often associated to the development of the so-called hypersensitive response (HR), a form of programmed cell death that prevents spreading of the pathogen beyond the inoculated zone. This defense-associated cell death is closely connected to plant physiological and developmental processes and needs to be tightly regulated to be not only efficient but also beneficial to the plant. Moreover, the sharp boundary of the HR suggests the existence of efficient mechanisms that control cell death and survival. Transcriptional regulation in host cells plays a crucial role in the establishment of plant disease resistance to pathogen attack [1]. The MYB transcription factor (TF) **MYB30** is a positive regulator of Arabidopsis defence and HR responses to bacterial pathogens [2]. MYB30 appears to modulate cell death-related lipid signaling by enhancing the synthesis of sphingolipid-containing very long chain fatty acids (VLCFAs) after bacterial inoculation [3]. Moreover, a second MYB TF of the MYB30 phylogenetic subgroup, **MYB96**, physically interacts and cooperates with MYB30 for the transcriptional activation of VLCFA production during the Arabidopsis defence response to bacteria (unpublished data).

In agreement with the finding that transcriptional activation of VLCFA-related genes by MYB30 is required to mount an efficient defence response during bacterial infection, we have demonstrated that MYB30 transcriptional activity is tightly controlled by the plant cell [4]. Indeed, MYB30 is able to induce partial nuclear relocalization of the secreted phospholipase ***At*sPLA2-a,** which is otherwise localized intracellularly in Golgi-associated vesicles before being secreted to the extracellular space. The physical interaction between MYB30 and *At*sPLA2-□□leads to repression of the MYB30-mediated transcriptional activity and negative regulation of plant HR and defence responses [5]. These data highlight the importance of dynamic nucleocytoplasmic protein trafficking for the regulation of defence-related transcription. An additional regulatory mechanism of MYB30 action was uncovered by the identification of the *Arabidopsis* RING-type E3-ubiquitin-ligase **MIEL1** (MYB30-INTERACTING E3 LIGASE1) as an MYB30 interactor in yeast [6]. MIEL1 is an active E3 ligase able to ubiquitinate MYB30 *in vitro*. In *Arabidopsis*, MIEL1 leads to MYB30 proteasomal degradation, downregulation of its transcriptional

activity and suppression of plant defence responses. Indeed, Arabidopsis *miel1* mutant plants displayed enhanced HR and resistance after inoculation with avirulent bacteria. These phenotypes are MYB30-dependent and correlate with down-regulation of MYB30 target genes related to VLCFA metabolism [6]. This work shows the important role played by ubiquitination during the transcriptional control of the HR and underlines the sophisticated fine-tuning of plant responses to pathogen attack. In addition, **SBT**, a serine-type endopeptidase of the subtilase family, has been recently identified as an additional MYB30 regulator. The *SBT* transcript is alternatively spliced giving rise to both a secreted (SBTa) and a nuclear (SBTb) protein. Interestingly, SBTb, but not SBTa, interacts with MYB30 blocking MYB30 DNA binding and transcriptional activation and this appears to be independent of SBT catalytic activity. *sbt* mutant plants, with no *SBTa* nor *SBTb* expression, display enhanced HR and defence and increased MYB30 target gene expression. These phenotypes are reverted by overexpression of SBTb, but not SBTa, in the *sbt* mutant background, underlining the specific repression of MYB30-mediated defence by SBTb (unpublished data). The coordinated action of these different regulators for the spatiotemporal control of MYB30 activity will be discussed.

Plant and animal pathogenic bacteria inject type III effectors (T3Es) into host cells to suppress host immunity and promote successful infection. XopD from *Xanthomonas campestris* is a modular T3E that is targeted to the nucleus of host cells where it is able to display a variety of biochemical activities. XopD exhibits small ubiquitin-like modifier (SUMO) protease activity thanks to the presence of a cysteine protease domain at its C-terminus. In addition, two tandemly repeated transcriptional repressor EAR (ERF-associated Amphiphillic Repression) motifs confer to XopD the ability to repress transcription of defence- and senescence-related plant genes. Finally, an intact helix- loop-helix domain (HLH) is necessary for XopD nuclear targeting and the ability to display non- specific DNA-binding. It has been suggested that a XopD N-terminal domain of unknown function may confer specificity for DNA-binding, but this hypothesis remains to be demonstrated [7]. Based on these biochemical properties, it was suggested that XopD mediates multiple protein-DNA and protein- protein interactions to modulate host transcription and that XopD may target plant TFs and/or regulators in the nucleus. Indeed, we showed that XopD from strain B100 of *X. campestris* pv. *campestris* is able to target MYB30 in *Arabidopsis*. XopD specifically interacts with MYB30 *via* its HLH domain, which is also necessary and sufficient for suppression of the transcriptional activation of MYB30 VLCFA-related target genes and therefore for inhibition of plant defence and HR responses [8]. Our work uncovered a new biochemical property of the HLH domain, beyond the previously identified activities related to nuclear targeting and DNA-binding.

References

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