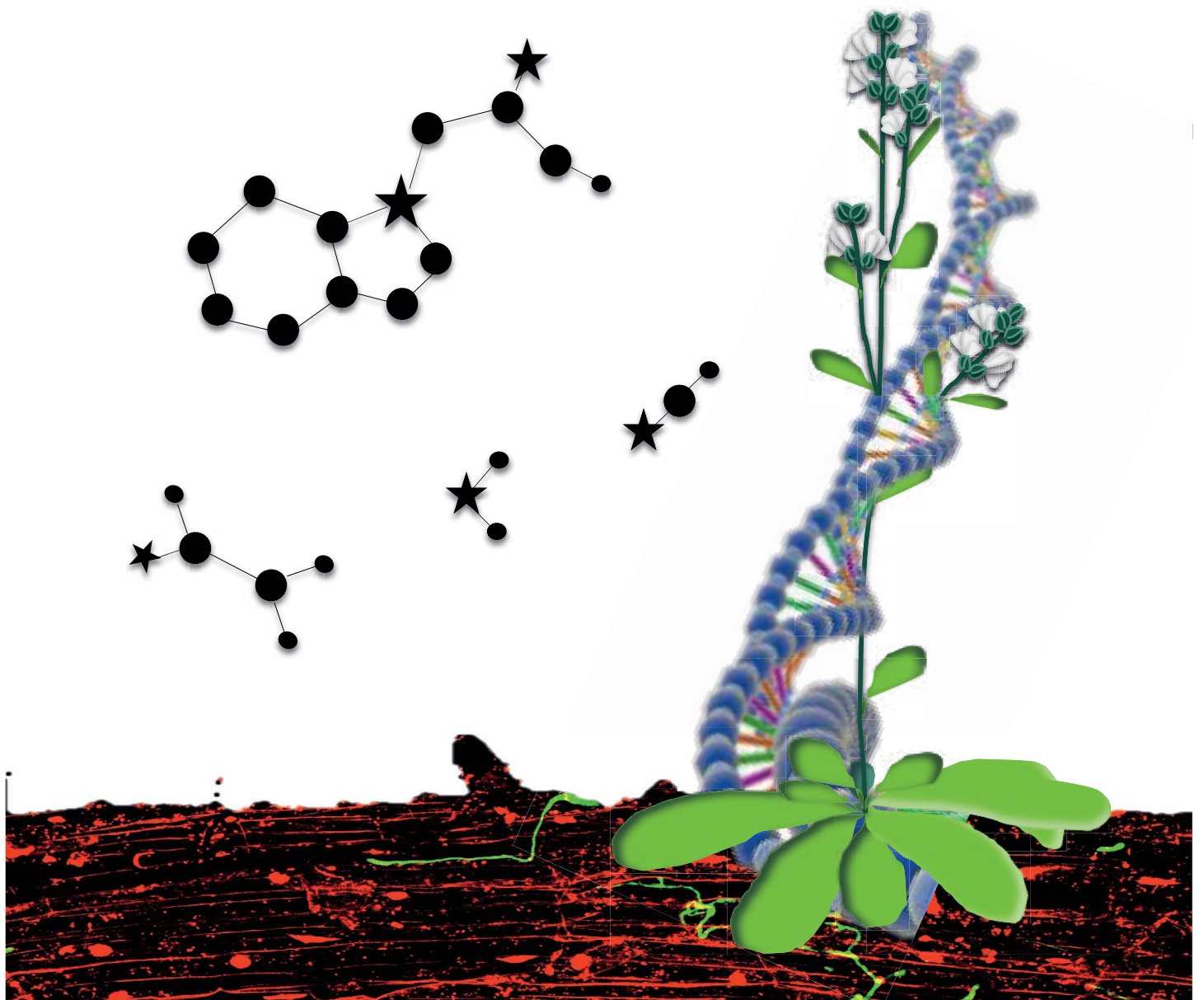


XVI MEETING OF PLANT MOLECULAR BIOLOGY



ABSTRACT BOOK



THE WHEN AND WHERE: HOW DEVELOPMENT MODULATES IMMUNITY THROUGH A MIRNA-NLR-PHASICRNA NETWORK

Ángel Del-Espino¹, Diego López-Márquez^{1,2}, Nieves López-Pagán¹, Ignacio Rubio-Somoza³, Javier Ruiz-Albert¹, Eduardo R. Bejarano¹, Carmen R. Beuzón¹.

¹Instituto de Hortofruticultura Subtropical y Mediterránea "La Mayora", Universidad de Málaga-Consejo Superior de Investigaciones Científicas (IHSM-UMA-CSIC), Depto. Biología Celular, Genética y Fisiología, Málaga, Spain; ²Present address: University of Copenhagen, Department of Biology, Copenhagen, Denmark. ³Molecular Reprogramming and Evolution (MoRE) Lab, Centre for Research in Agricultural Genomics (CRAG), Carrer Vall Moronta Edifici CRAG, 08193, Barcelona, Spain.

Corresponding authors: Eduardo Rodríguez Bejarano, edu_rodri@uma.es and Carmen R. Beuzón López, cbl@uma.es



Plants possess a battery of dedicated intracellular immune receptors known as nucleotide-binding leucine-rich repeat proteins (NLRs) involved in recognition of pathogen-derived effectors and activation of effector-triggered immunity (ETI). In a recent work, we report a two-tiered regulatory network in *Arabidopsis* mediated by a microRNA (miR825-5p) and a wave of secondary NLR-derived phased small RNAs (phasi-NLRs) involved in silencing dozens of NLR genes encoding Toll/interleukin-1 NLRs (TNLs). In our model, targeting of *MIST1* (microRNA-silenced TNL1) transcripts by miR825-5p, triggers the generation of phasi-NLRs that subsequently silence a wide network of TNL-encoding genes and reinforce the silencing of *MIST1* (López-Márquez *et al.*, 2021).

Although miRNA and phasi-NLR mediated regulation of NLRs has been characterized to some extent, the current knowledge regarding how these miRNA-target-phasiRNA networks are modulated and their contribution to the immune response during different developmental stages is still scarce (Deng *et al.*, 2018; Cui *et al.*, 2019). Thus, through generation of GUS-based *A. thaliana* reporter lines we have characterized the expression pattern of both genes (*MIR825* and *MIST1*) as well as the domain activity of miR825-5p. Moreover, a comprehensive analysis of RNA and small RNA-seq datasets, as well as the use of different orthogonal approaches, unravelled a hitherto unknown connection between miR825-5p/phasi-NLR mediated regulation of NLRs and plant development. Further analysis also supports the notion that the action of this regulatory mechanism is restricted to control NLR expression in an organ specific way, mainly operating in leaves of the model plant. At present, we are working to understand how the changes on miRNA/phasi-NLR levels during developmental stages modulate the strength of the immune response and the biological impact of this developmental-dependent regulation on plant health.

In summary, our results point to a developmentally regulated sRNA-based control of NLR expression in *Arabidopsis* and shed light on how levels of these regulatory molecules (miRNAs and phasi-NLRs) are modulated during development or in an organ specific manner to fine-tune NLR expression.

Cui, C., Wang, J.-J., Zhao, J.-H., Fang, Y.-Y., He, X.-F., Guo, H.-S., *et al.* (2019). A Brassica miRNA Regulates Plant Growth and Immunity through Distinct Modes of Action. *Molecular Plant*, 1–38. doi:10.1016/j.molp.2019.11.010.

Deng, Y., Wang, J., Tung, J., Liu, D., Zhou, Y., He, S., *et al.* (2018). A role for small RNA in regulating innate immunity during plant growth. *PLoS Pathog* 14, e1006756–22. doi:10.1371/journal.ppat.1006756.

López-Márquez, D., Del-Espino, Á., López-Pagán, N., Rodríguez-Negrete, E.A., Rubio-Somoza, I., Ruiz-Albert, J., Bejarano, E.R., and Beuzón, C.R. (2021). miR825-5p targets the TIR-NBS-LRR gene *MIST1* and down-regulates basal immunity against *Pseudomonas syringae* in *Arabidopsis*. *Journal of Experimental Botany* 72, 7316–7334. doi:10.1093/jxb/erab354.

Acknowledgements & Funding

This work was supported by Project Grant RTI2018-095069-B-I00 financed by MCIN/AEI/10.13039/501100011033/ and FEDER, and by Project Grant PY18-2398 financed by Junta de Andalucía. ADE was funded by a FPU Grant (Predoctoral Fellowship from the Spanish Ministerio de Ciencia, Innovación y Universidades; FPU17/03520).