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OKINAWA INSTITUTE OF SCIENCE AND TECHNOLOGY GRADUATE UNIVERSITY
沖縄科学技術大学院大学

rCBP-dependent regulation in rice innate immunity

Author	Nino Espinas
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Abstract

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Plant innate immunity against bacterial attacks is a two-tiered inducible system capable of defense responses at local and systemic areas. These systems are the PTI and ETI. During infection, PTI has the ability to recognize microbial signatures upon bacterial contact, while ETI recognizes microbial protein secretions called effectors delivered inside the cell. The activation of PTI and ETI confers systemic tissues of infected plants a broad-spectrum immunity against later pathogen attacks termed systemic acquired resistance (SAR). Defense priming is an adaptive component of SAR that regulates the molecular storage of defense memory for a more effective defense response.

The main aim of this work is finding a novel molecular defense signaling pathway that is controlled by acetylation at the infected (local defense) and systemic tissues (priming defense).

To investigate the role of histone acetyltransferase-dependent pathway in plant immunity, I have isolated transgenic and mutant lines of *rCBP*, [rice Cyclic adenosine monophosphate response element-binding protein (CREB) Binding Protein], under Nipponbare cultivar background using RNAi silencing and gRNA/Cas9-mediated genome editing. Animal CBP was initially described as both transcriptional coactivator and histone acetyltransferase. The *rCBP*-RNAi lines with mistargeting of the other members of CBP family are characterized by massive sterility and impairment of the number of effective grains. On the other hand, the CRISPR/Cas9 mutant lines have wild-type number of effective grains.

To profile the global acetylation of histone lysine-sites *via* rCBP, I performed mass spectrometry-based proteomics in data dependent acquisition (DDA) and parallel reaction monitoring (PRM) modes. My results showed that H3 lysine sites are possibly targeted by rCBP with very high acetylation specificity on H3K9.

To implicate the role of rCBP in rice innate immunity, I conducted a pathogenesis assay with bacterial pathogen, *Pseudomonas syringiae* pv. *oryzae* (*Pso*). Pathogenesis assay showed that *rCBP*^{-/-} mutants are resistant to *Pso* infection compared to segregated wild-type control.

I also performed transcriptome analysis on locally-infected tissues and on systemic tissues to investigate the genome-wide effects of *rCBP* mutation and to identify factors with roles in both local and systemic immune response. As a result, I have identified seven putative rCBP-dependent transcriptional repressors that possibly explain the resistance phenotype of *rCBP* mutant lines in locally-infected tissues. On the other hand, non-infected systemic tissues in mutant lines show diminishing number of genes with significant expression

Overall, these data preliminary indicate that *rCBP* is both a positive regulator of developmental processes and a negative regulator of rice immunity. These data also suggest that rCBP may execute this dual regulatory function either through H3K9ac and/or co-transcriptional activity on target gene loci. It is also tempting to hypothesize that *rCBP* might potentially regulate systemic defenses through an unknown mechanism at distal non-infected site in preparation for future infection episodes.