



(0) Research field

CPR Subcommittee: Biology

Keywords: epigenome regulation, histone modification, histone variant, non-coding RNA, plant

(1) Long-term goal of laboratory and research background

Our laboratory aims to elucidate the molecular mechanisms of epigenome regulation networks in plant life cycle. To understand this topic involving histone modification and non-coding RNAs, we perform cutting-edge multidisciplinary approaches using model plants, including molecular biology, chemical biology and integrated omics analysis.

(2) Current research activities (FY2019) and plan (until Mar. 2025)

1. Unveiling regulatory network for salinity stress response controlled by HDACs by transcriptome analysis in Arabidopsis.

Acetylation in histone and non-histone proteins is balanced by histone acetyltransferase and histone deacetylase (HDAC) enzymatic activity, an essential aspect of fine-tuning plant response to environmental stresses. A previous study indicated that class I (HDA19) and class II (HDA5/14/15/18) RPD3-like family HDACs control positive and negative responses to salinity stress, respectively. Furthermore, quintuple *hda5/14/15/18/19* mutants (*quint*) exhibit salinity stress tolerance, suggesting that *hda19* suppresses the sensitivity to salinity stress present in quadruple *hda5/14/15/18* mutants (*quad*). In the present study, transcriptome analysis of the *quint* mutant was conducted to elucidate the hierarchical control of salinity stress response operated by RPD3-like family HDACs (HDA5/14/15/18/19). The analysis indicates that deficiency in HDA19 has a bigger impact on salinity stress response than in class II HDACs. Furthermore, the expression pattern of genes encoding enzymes that metabolize phytohormones raises the possibility that a drastic change in the homeostasis of phytohormones. Abscisic acid accumulation actually increased in *hda19-3* and *quint* plants, and decreased in *quad*, compared with wild-type plants. Notably, 7.8% of the salt-responsive genes in *quint* plants exhibited a similar expression pattern in *quad* plants, suggesting that some gene sets are regulated in an HDA5/14/15/18-dependent manner (Ueda *et al.*, 2019).

To date, we have identified histone deacetylases involved in stress response in Arabidopsis. For the purpose of the manipulation of stress response, we will develop the screening system to find chemical compounds which alter these HDACs activity. The transcriptome and phytohormone data information would be useful for the evaluation for the effect of newly identified compounds having HDAC inhibitory or activating effect. The methods would apply for HDAC enzymes in land plants to understand the role of HDACs in not only stress adaptation but also development and so on.

2. Intracellular localization of histone deacetylase HDA6 in plants

The observation of intracellular localization of HDACs provides information on where each HDAC participates in biological process because HDACs are located to different cytoplasmic compartments such as the nucleus, mitochondria, and plastids. Although HDA6 plays an important role in chromatin control and response to drought stress, its intracellular localization has not been observed in detail. In this paper, we generated transformants expressing HDA6-GFP in the model plant, *Arabidopsis thaliana*, and the crops, rice, and cassava. We observed the localization of the fusion protein and showed that HDA6-GFP was expressed in the whole root and localized at the nucleus in Arabidopsis, rice, and cassava. Remarkably, HDA6-GFP clearly formed speckles that were actively colocalized with chromocenters in Arabidopsis root meristem. In contrast, such speckles were unlikely to be formed in rice or cassava. Because HDA6 directly binds to the acetate synthesis genes, which function in drought tolerance, we performed live imaging analyses to examine the cellular dynamics of pH in roots and the subnuclear dynamics of HDA6 responding to acetic acid treatment. The number of HDA6 speckles increased during drought stress, suggesting a role in contributing to drought stress tolerance (Kurita *et al.*, J. Plant Res.).

The study suggests that at least HDA6 homologues in plants have diversified their roles during the evolution of plants. The roles of HDACs in stress adaptation and development still remain unknown in crops. Further study will uncover unique roles of HDACs in stress adaptation and development in crops.

Future plan

Besides the above, we are elucidating novel epigenome regulation factor networks and their functions in plant life cycles. These factors will include histone modification enzymes, histone variants, DNA methyltransferases, and non-coding RNAs. We will use integrated omics, protein-protein and protein-RNA interaction analysis, and imaging analysis. Antagonistic or synergistic interactions that fine-tune biological processes, such as between histone modifications (e.g. acetylation vs methylation), between histone variants (e.g. H2B vs H3), and between histone modifications and non-coding RNAs, will be elucidated.

(3) Members

as of March, 2020

(Chief Scientist)

Motoaki Seki

(Research scientist)

Akihiro Matsui, Minoru Ueda

(Technical Staff)

Junko Ishida, Satoshi Takahashi, Maho Tanaka

(4) Representative research achievements

1. “Transcriptome analysis of the hierarchical response of HDAC proteins that respond in an antagonistic manner to salinity stress” Ueda, M., Matsui, A., Watanabe, S., Kobayashi, M., Saito, K., Tanaka, M., Ishida, J., Kusano, M., Seo, M. and Seki, M. *Front. Plant Sci.* (2019) 10: 1323.
2. “Intracellular localization of histone deacetylase HDA6 in plants” Kurita, K. #, Sakamoto, Y. #, Naruse, S., Matsunaga, T.M., Arata, H., Higashiyama, T., Habu, Y., Utsumi, Y., Utsumi, C., Tanaka, M., Takahashi, S., Kim, J.M., Seki, M., Sakamoto, T. and Matsunaga, S. *J. Plant Res.* (2019) 132:629-640.
3. “Long noncoding RNAs in *Brassica rapa* L. following vernalization” Shea, D., Nishida, N., Takada, S., Itabashi, E., Takahashi, S., Akter, A., Miyaji, N., Osabe, K., Mehraj, H., Shimizu, M., Seki, M., Kakizaki, T., Okazaki, K., Dennis, E. and Fujimoto, R. *Sci. Rep.* (2019) 9:9302.

Laboratory Homepage

https://www.riken.jp/en/research/labs/chief/plant_epigen_reg/index.html

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