

**Title of Project : Integrated understanding of interspecific-incompatibility and self-incompatibility in the Brassicaceae**

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Keyword : Plant, Reproduction, Signaling, Interspecific incompatibility, Self incompatibility

**【Purpose and Background of the Research】**

Many plants maintain their species and genetic diversity by selectively fertilizing only pollen of "same species" and "non-self" from among the miscellaneous pollen carried by insects and other pollinators. In the Brassicaceae, papillae cells at the tip of the pistil serve as the primary site of "interspecific-incompatibility", which involves the exclusion of "heterospecific" pollen, and "self-incompatibility", which involves the exclusion of "self" pollen (Fig. 1). The two incompatibility pathways are predicted to interfere with the pollen "acceptance pathway" in some way, but the actual mechanism of such interference is still unknown. The aim of this study is to deepen our understanding of the pathways involved in interspecific-incompatibility, self-incompatibility, and pollen acceptance, and to propose an integrated molecular model of pollen selection that includes interference between pathways.

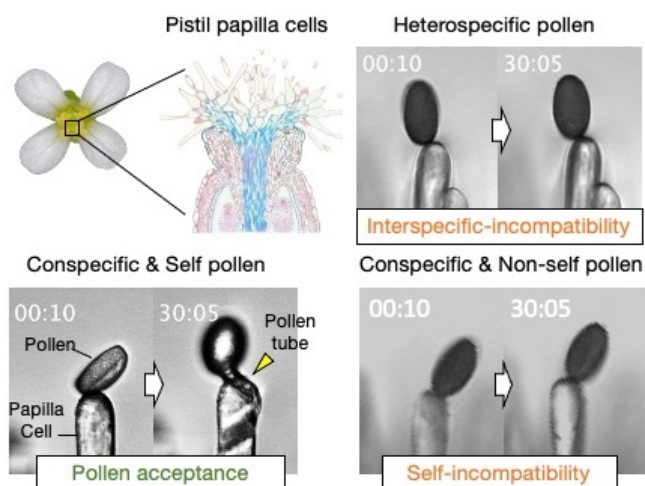


Fig. 1. Interspecific- and Self-incompatibilities in the Brassicaceae

**【Research Methods】**

To propose an integrated molecular model of pollen selection in the Brassicaceae, we will pursue the following three subjects.

**【Subject 1】** Investigation of the molecular mechanism of heterospecific pollen rejection

We have identified a stigmatic factor, SPRI1, which is involved in the active rejection of heterospecific pollen by the genome-wide association study (GWAS). Here we aim to understand the molecular function of SPRI1 including the identification of the putative pollen counterpart of this protein. In addition, we will continue to search for novel

interspecific-incompatibility factors using GWAS and clarify their relationship with SPRI1.

**【Subject 2】** Investigation of the molecular mechanism of self-pollen rejection

We have shown that the self-specific interaction between the *S* locus-encoded factors pollen ligand SP11 and the female receptor kinase SRK induces an incompatibility response with Ca<sup>2+</sup> influx into papilla cells. In this study, we will focus on the signal transduction pathway leading to the Ca<sup>2+</sup> influx downstream of SRK and try to identify the factors involved by biochemical or genetic methods.

**【Subject 3】** Investigation of the molecular mechanism of the pollen acceptance and interference

We have shown that conspecific pollen has a mechanism to induce papillae cells to respond to pollen acceptance. Using our original pollination bioassay system, we will search for pollen- and pistil-side factors involved in the pollen acceptance response by bioorganic chemistry and genetic approaches and clarify their physiological functions. Furthermore, we will elucidate the relationship between this acceptance pathway and the above two incompatibility pathways and propose an integrated molecular model of pollen selection.

**【Expected Research Achievements and Scientific Significance】**

Sexual reproduction has created genetic diversities by discriminating "species" at the stage of sexual reproduction, and also by promoting cross-fertilization within species. This study aims to elucidate the mechanisms that generate diversity, which is the foundation of biological prosperity. It is also expected to contribute to the understanding of the complex information processing system in plant cells. Furthermore, artificial control of this system is expected to lead to applications in agriculture, such as interspecific hybrid breeding and F<sub>1</sub> hybrid breeding.

**【Publications Relevant to the Project】**

- Fujii S, 10 co-authors, Takayama S. A stigmatic gene confers interspecies incompatibility in the Brassicaceae. *Nature Plants* 5, 731-741 (2019).
- Murase K, 16 co-authors, Takayama S. Mechanism of self/nonself-discrimination in *Brassica* self-incompatibility. *Nature Communications* 11, 4916 (2020).

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