LABORATORY OF CELL RESPONSES

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Cells sense the environment around them, for example the amount of nutrients and hormones, as well as the temperature and pressure, and decide what kind of activities to undertake using this information. Germ cells, which produce sperm and eggs, begin halving their number of chromosomes during a special kind of cell division called meiosis, in response to the ambient conditions. In our laboratory, we use the fission yeast *Schizosaccharomyces pombe*, the simplest organism that performs meiosis (Figure 1), to research the mechanism by which cells switch from mitosis, the kind of cell division that divides cells equally to create two identical cells, to meiosis, which is essential for bringing forth genetically diverse progeny.

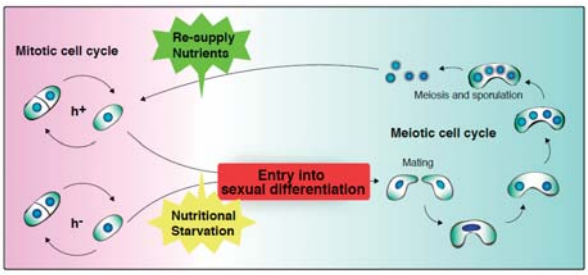

Figure 1. Life cycle of the fission yeast *S. pombe*. *S. pombe* cells proliferate by mitotic growth under nutrient-rich conditions. When starved of nutrients, especially nitrogen, *S. pombe* cells arrest the mitotic cell cycle and haploid cells conjugate with cells of the opposite mating type. Resulting diploid zygotes undergo meiosis and produce spores.

I. Signaling pathways that regulate the onset of sexual differentiation

We have been trying to elucidate how *S. pombe* cells switch their mode of cell cycle from mitotic to meiotic. We focus on a highly conserved kinase, namely Target of rapamycin (TOR) kinase, which plays key roles in the recognition of nutrition and the onset of sexual differentiation in *S. pombe*. TOR kinase forms two types of complexes, namely TORC1 and TORC2. TORC1 contains Tor2 as its catalytic subunit and is essential to suppress sexual differentiation in the presence of nitrogen. TORC2 contains Tor1 and, in contrast to TORC1, is required for onset of sexual differentiation under nitrogen starvation (Figure 2).

Temperature-sensitive *tor2* mutants initiate sexual differentiation even on rich medium at the restrictive temperature. To gain insights into the TORC1 signaling pathway, we have isolated mutants that initiate sexual differentiation ectopically under nutrient-rich conditions. In most mutants identified, TORC1 activity is downregulated and the mutated genes are involved in tRNA expression or modification. We are currently characterizing these mutants.

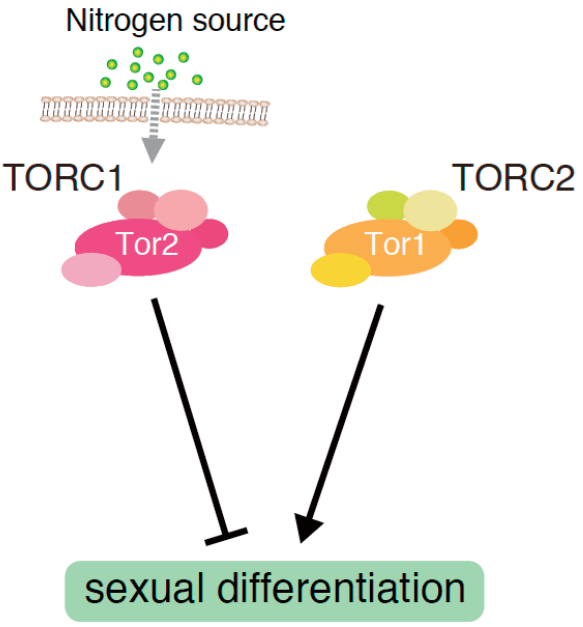

Figure 2. The two TOR complex pathways in *S. pombe*. TORC1, containing Tor2, and TORC2, containing Tor1, regulate sexual differentiation oppositely. TORC1 suppresses sexual differentiation in the presence of ample nitrogen.

II. The molecular mechanisms that establish the meiosis-specific gene expression profile

Expression of hundreds of genes are upregulated during meiosis. We have shown that specific control of the stability of meiotic transcripts, which is orchestrated by the interplay between RNA-binding proteins and a long non-coding RNA, contributes to the meiosis-specific gene expression in the fission yeast *S. pombe*. Understanding precise mechanisms of this control will shed light on the regulation of timely gene expression during meiosis.

A YTH-family RNA-binding protein Mmi1 plays a crucial role in the selective elimination system of meiosis-specific transcripts during the mitotic cell cycle. Mmi1 recognizes a region termed DSR (Determinant of Selective Removal) in meiotic transcripts, which is enriched with repeats of hexanucleotide motifs. Meiotic transcripts bound to Mmi1 are degraded by the RNA-degradation nuclear exosome machinery. Mmi1 also induces formation of facultative heterochromatin at a subset of its target genes.

During meiosis, a meiosis-specific nuclear body, called Mei2 dot, blocks the Mmi1-mediated elimination system. The Mei2 dot is composed of the RNA-binding protein Mei2 and a long non-coding RNA species termed meiRNA. Mei2 physically interacts with meiRNA and forms the dot structure at the chromosomal *sme2* locus, which encodes meiRNA. The Mei2 dot lures Mmi1 through numerous copies of the DSR motif on meiRNA and inhibits its function, so that meiotic transcripts harboring DSR are stably expressed (Figure 3).

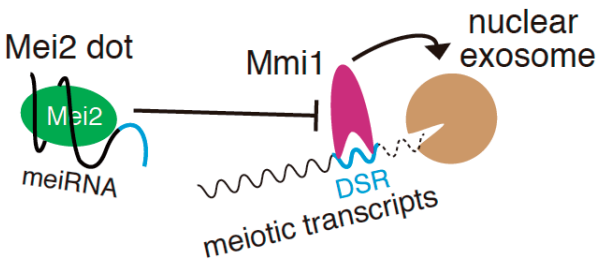

Figure 3. Selective elimination of meiosis-specific transcripts by the Mmi1/DSR system. Mmi1 binds to DSR in meiotic transcripts and induces their degradation by the nuclear exosome during the mitotic cell cycle. In meiotic cells, the Mei2 dot, composed of Mei2 and meiRNA, sequesters and inhibits Mmi1, so that DSR-harboring meiotic transcripts escape from Mmi1-mediated selective elimination.

We have shown that Mmi1 regulates the termination of transcription of its target genes. Mmi1-mediated termination of an upstream non-coding RNA ensures the expression of downstream genes, one of which encodes a mitogen-activated protein kinase kinase kinase (MAPKKK) essential for the initiation of sexual differentiation. We have also demonstrated that Mmi1 prevents untimely expression of meiotic proteins by tethering their mRNAs to nuclear foci. Multilayered suppression of meiotic genes by Mmi1 is vital for mitotic growth.

Publication List:

〔Original paper〕

● Touat-Todeschini, L., Shichino, Y., Dangin, M., Thierry-Mieg, N., Gliquin, B., Hiriart, E., Sachidanandam, R., Lambert, E., Brettschneider, J., Reuter, M., Kadlec, J., Pillai R., Yamashita, A., Yamamoto., M., and Verdel, A. (2017). Selective termination of lncRNA transcription promotes epigenetic silencing and cell differentiation. EMBO J. *36*, 2626-2641.

〔Review article〕

● Otsubo, Y., Yamamoto, M., and Yamashita, A. (2017). TORC1-dependent phosphorylation targets in fission yeast. Biomolecules *7*, 50.