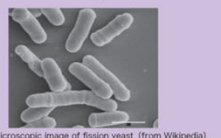
Iwasaki Lab

Homologous Recombination and Its Role in Genome Maintenance

Cell Biology Center http://www.iwasakilab.bio.titech.ac.jp

Fission yeast
Schizosaccharomyces pombe



Electron microscopic image of fission yeast (from Wikipedia)

Shape : cylindrical (3-4 μm in diameter; 7-15 μm long)
Fenome size : 13.8 Mb
Number of chronosomes : 3
chr I : 5.7 Mb, chr II : 4.6 Mb,
chr III : 3.5 Mb
(The whole sequence of the genome was determined by the Wellcome Sanger Institute in 2002)

Fig 1 Fission yeast

Molecular Genetics and Fission Yeast

In the field of molecular genetics, we aim to understand various biological phenomena at the molecular level. This research typically involves so-called model organisms such as E.coli (bacteria), yeast (lower eukaryotes), C.elegans (nematode worm), Arabidopsis (plants) and mice (mammals) (Fig 1). Two popular yeast model organisms are fission yeast and budding yeast, named after their mode of cell division. Fission yeast is thought to possess mechanisms more similar to those of animal cells than budding yeast. Using fission yeast, many important discoveries have been made in the fields of the cell cycle, chromosome separation, centromere, telomere, heterochromatin, and DNA repair. Dr. Paul Nurse was awarded a Nobel Prize in Physiology or Medicine in 2001 for the discovery of the Cdc2 kinase, a central regulator of the cell cycle.

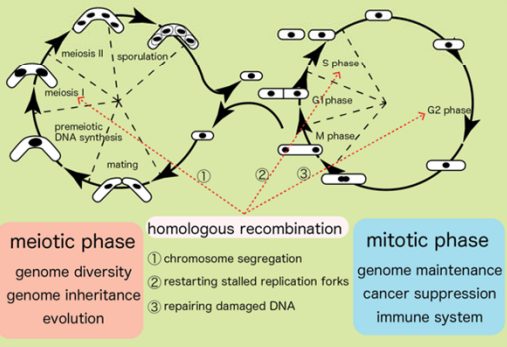


Fig 2 Life Cycle of Fission Yeast and Homologous Recombination

Homologous Recombination

Homologous recombination plays an important role not only in generating genetic diversity but also in the repair of damaged DNA and maintenance of genome integrity. It also plays a critical role in restarting a replication fork that has stalled upon encountering DNA damage. Homologous recombination is important for various other functions as well (Fig 2). We use fission yeast as a model to understand the molecular mechanisms of homologous recombination in maintaining genome integrity and controlling the genome rearrangements that diversify our genomes.

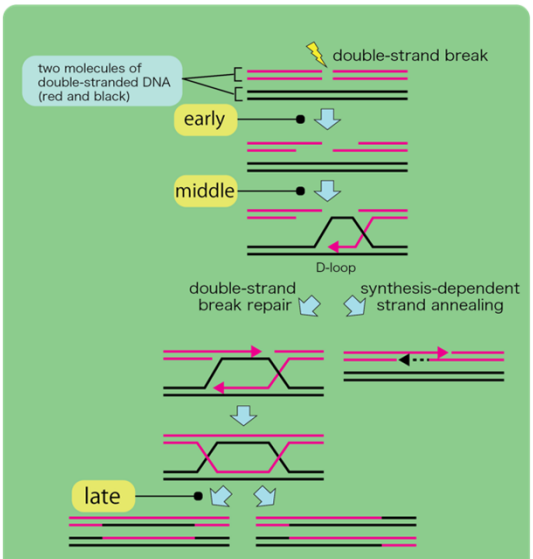


Fig 3 Homologous recombination and DNA double-stranded break repair

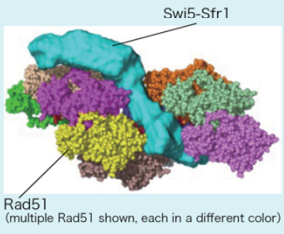


Fig 4 Rad51 and Swi5-Sfr1 in the form of the complex

Homologous Recombination in Genome Maintenance

Homologous recombination takes place in three steps (early, middle and late steps, Fig 3). In the early step, single-stranded DNA (ssDNA) is generated at the end of broken DNA. The Mre11-Rad50-Nbs1-Ctp1 complex plays a major role in this process. This ssDNA is bound by Rad51, a homologous recombinase. Rad51 forms a helical filament structure along ssDNA, which is essential for homology searching and strand exchange (middle step). Once strands are exchanged between homologous DNA molecules, a D-loop is formed, which is eventually processed through multiple enzymatic pathways to form mature recombinants (late step).

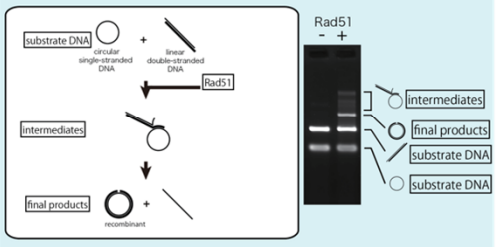


Fig 5 *in vitro* reconstituted homologous recombination

in vitro Reconstitution of Homologous Recombination

We have identified enzymes playing key roles at each step of homologous recombination, such as Nbs1, Ctp1, Swi5-Sfr1, Rad55-Rad57 and Fbh1. By characterizing these proteins, we aim to understand the whole mechanism of homologous recombination and its role in genome maintenance (Fig 4 and 5)

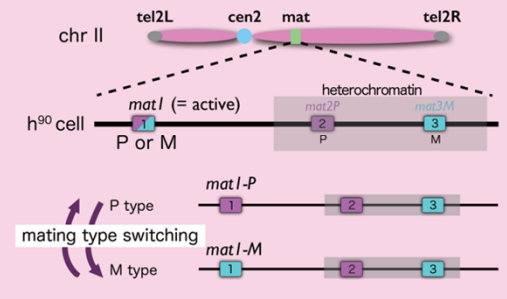


Fig 6 Mating type switching of fission yeast

Homologous Recombination in Genome Rearrangement

Homologous recombination that takes place during meiosis acts as a driving force for genome rearrangements. Meiotic recombination proceeds in a highly coordinated manner in which Dmc1, a meiosis-specific homologous recombinase, plays a key role along with Rad51. Also, during vegetative growth, mating type switching takes place through highly regulated genome rearrangement processes (Fig 6). We aim to understand how these genome rearrangement events are organized.