

LABORATORY OF BIOLOGICAL DIVERSITY

JOHZUKA Group

Assistant Professor: JOHZUKA, Katsuki
 Technical Assistant: ISHINE Naomi

Chromosome condensation is a basic cellular process that ensures the faithful segregation of chromosomes in both mitosis and meiosis. This process is required not only for shrinking the length of chromosome arms, but also for resolving entanglements between sister-chromatids that are created during DNA replication. Any abnormality in this process leads to segregation errors or aneuploidy, resulting in cell lethality. Chromosome condensation is mainly achieved by condensin, a hetero-pentameric protein complex, widely conserved from yeast to humans. Despite its conservation and importance for chromosome dynamics, how condensin works is not well understood. Recent studies reveal that condensin functions are not restricted to chromosome condensation and segregation during cell divisions. It is required for diverse DNA metabolism such as genome stability, transcriptional regulation, and cell differentiation.

Our research interest is to understand the mechanism and regulation of chromosome condensation. We have been studying the role of condensin in the budding yeast *Saccharomyces cerevisiae*. Microscopic observation indicated the nucleolar localization of condensin. Consistent with this, the ribosomal RNA gene (rDNA) repeat is the most condensed region in the genome during mitosis. We have found that condensin specifically binds to the RFB site located within the rDNA repeat. To date, the best characterized condensin binding region is the rDNA repeat on the right arm of chromosome XII in budding yeast. We further discovered the multiple protein network required to recruit condensin to the RFB site.

I. Dynamic relocalization of condensin during meiosis

Our genetic screening indicated that two proteins, Csm1 and Lrs4, were required for condensin recruitment to the RFB site. Physical interactions between Csm1/Lrs4 and subunits of condensin are important for recruitment of condensin to the RFB site. These proteins are known as components of monopolin complex that are required for faithful segregation of homologous chromosomes during meiotic division I. During meiosis I, monopolin complex re-localizes from rDNA repeat to the centromere and acts for ensuring sister-chromatid co-orientation. Re-localization of Csm1/Lrs4 proteins suggested the re-localization of condensin from rDNA repeat to centromere. As expected, chromatin-IP experiments indicated that condensin re-localizes to the centromere during meiosis I. Condensin might clamp sister-chromatids together during meiosis I.

II. Condensin-dependent chromatin folding

The RFB site, which consists of a ~150bp DNA sequence, is functioning as a cis-element for recruitment of condensin to chromatin in the yeast genome. If the RFB site is inserted into an ectopic chromosomal locus, condensin can associate

with the ectopic RFB site. To explore the role of condensin in chromosome organization, we have constructed a strain in which two RFB sites are inserted on an ectopic chromosome arm with an interval of 15kb distance in the cell with complete deletion of chromosomal rDNA repeat. Using this strain, condensin-dependent chromatin interaction between two RFBs was examined by chromosome conformation capture (3C) assay. We found the condensin-dependent chromatin interaction between the two RFB sites on the chromosome arm. This result indicates that condensin plays a role in chromatin interaction between condensin binding sites, and this interaction leads to creation of a chromatin loop between those sites (Figure 1). It is thought that condensin-dependent chromatin folding is one of the basic molecular processes of chromosome condensation. During the cell cycle stages, the RFB - RFB interaction signal increases in metaphase and reaches its maximum level in anaphase. In addition to the RFB - RFB interaction, the chromatin interactions between internal regions of two RFBs increases in anaphase. Thus, the configuration of chromatin fiber changes from a simple loop into a complicated twisted shape as the cell cycle progresses from metaphase to anaphase.

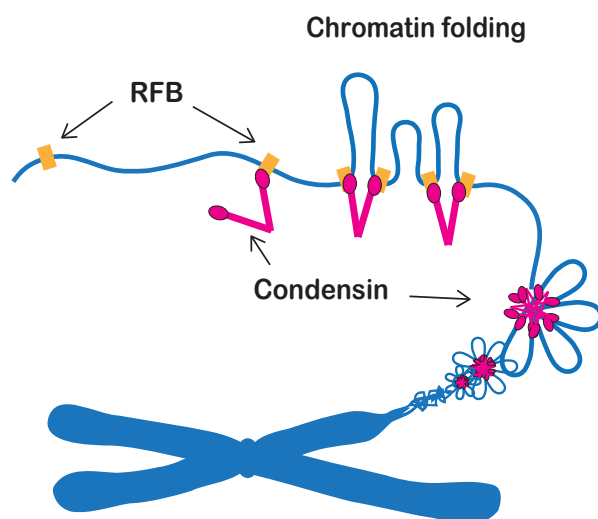


Figure 1. A Schematic model of chromosome condensation. Condensin makes chromatin interactions between adjacent binding sites (RFB, for example). This leads to a folding of chromatin fibers between the sites, as a basic process of chromosome condensation.