論文の要旨
（Thesis Summary）

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論文題目(Thesis Title) Understanding aspects behind recombination-dependent telomere maintenance and chromosome circularization using fission yeast
(分裂酵母を用いた環状染色体と組換え依存テロメア維持に関する性質の理解)

Telomere dysfunction or uncapped telomere can trigger series of genomic instability which can lead to cancer. Some population of cancer cells (10-15%) can escape the telomere dysfunction crisis through telomere maintenance by homologous recombination (HR). Alternatively, some cancer cells undergo chromosome circularization followed telomere dysfunction. Identifying the vulnerable points in these cells would allow me to find a possible target for cancer therapy. Therefore, I tried to explore the vulnerability in these two telomere dysfunctional situations, telomere recombination and circular chromosome using fission yeast (*S. pombe*) as a model organism. Deletion of fission yeast *pot1*$^{+}$ results in chromosome circularization. Rqh1 is a RecQ helicase that regulates HR events. Telomeres in *pot1*Δ *rqh1-hd* double mutant cells are maintained by HR and cells display long G2 and sensitive to microtubule inhibitors thiabendazole (TBZ). The reason behind the TBZ sensitivity phenotype of the *pot1*Δ *rqh1-hd* double mutant is unclear. One aim of my research is to understand the mechanism of the TBZ sensitivity phenotype of *pot1*Δ *rqh1-hd* double mutant which may allow me to find a specific vulnerability in cancer cells that maintain telomeres by HR. The second aim of my research is to identify the gene required for the maintenance of circular chromosomes which could enable development of cancer therapy that specifically targets cancer cells that have circular chromosomes. To study this, I used *S. pombe pot1*Δ cells that harbor circular chromosomes. Collectively, the data that I show in this thesis could contribute to the development of a therapeutic intervention to cure human cancers.

In the first study, I have investigated whether there is a link between the long G2 of *pot1*Δ *rqh1-hd* double mutant and its TBZ sensitivity. I found that shortening the G2 of the *pot1*Δ *rqh1-hd* double mutant by concomitant loss of function mutation of Chk1 downstream kinases, Wee1 and Mik1, or gain of function mutation of Cdc2 (*cdc2-3w*) suppressed the TBZ sensitivity and the accumulation of recombination intermediates at the telomeres of the double mutant. These results imply that the long G2 is the reason for the TBZ sensitivity of the *pot1*Δ *rqh1-hd* cells in the way that the activation of DNA

damage checkpoints and holding the *pot1*Δ *rqh1-hd* cells at long G2 provide cells with time to accumulate recombination structures at telomeres which disturb chromosomes segregation and render cells sensitive to TBZ. These results could further provide insight into possible roles of DNA damage signaling pathway in the regulation of HR events.

In the second study, I have investigated how circular chromosomes are maintained and how cells with circular chromosomes could survive using synthetic lethality approach. By using fission yeast *pot1*Δ and *trt1*Δ cells that have circular chromosomes, I found that the lack of function of CPC components is lethal to cells with circular chromosomes. The lack of functional CPC results in accumulation of elevated rates of chromosomes missegregation events and DNA damage. I further found that neither Shugoshin (Sgo2) nor heterochromatin protein (Swi6), which contribute to the centromeric localization of CPC, is lethal to *pot1*Δ cells with circular chromosomes. Both *pot1*Δ *sgo2*Δ and *pot1*Δ *swi6*Δ double mutants show a high percentage of DNA damage but a low percentage of chromosome missegregation, suggesting a link between chromosome missegregation and the lethality observed in *pot1*Δ *CPC* double mutants. These results demonstrate the importance of CPC for the survival of cells with circular chromosomes and shed light on the possible role of CPC in the maintenance of circular chromosomes.

Together, in this thesis, by using the fission yeast, I was able to simulate some phenotypes that occur in human cancer cells to understand biological questions which would be of clinical outcomes. I show a case in which the activation of DNA damage checkpoint could unexpectedly worsen the cells' viability by contributing to the accumulation of toxic recombination intermediates at the telomere. This finding implies that inhibition of human RecQ helicase and elongation of G2 phase by activating the G2 checkpoint may increase the sensitivity of cancer cells that maintain telomere by HR to anti-microtubule drugs. Furthermore, my results uncover the importance of CPC for the survival of cells with circular chromosomes. This finding implies that inhibition of CPC may specifically kill cancer cells that have circular chromosome. My work also proves that fission yeast is a good model organism to understand the mechanism of genome stability and cancer-related phenotypes that would directly contribute to cancer therapy and possibly other human diseases.