

The role of homologous recombination mediators in centromere structure maintenance in *Schizosaccharomyces pombe*

Abstract

Homologous recombination is a repair pathway which together with heterochromatin proteins ensures proper replication and maintenance of centromere structure and function. Rrp1 and Rrp2 are proteins from *Schizosaccharomyces pombe* which belong to a SWI2/SNF2 family of DNA translocases and are involved in DNA repair by homologous recombination pathway. Data obtained in this doctoral thesis show that overproduction of Rrp1 and Rrp2 results in defective replication and chromosome segregation leading to genome instability and growth defects. Because centromeres consist of repetitive sequences they are potentially difficult to replicate regions. It was shown that Rrp1 and Rrp2 bind to DNA in centromere region. Moreover, Rrp1 and Rrp2 are able to increase transcriptional silencing of reporter gene inserted within a centromere region and they affect centromere structure. Rrp1 and Rrp2 putative DNA translocase and ubiquitin ligase activities are essential for proper centromere structure. Overproduction of Rrp1 and Rrp2 results in global decrease of histone levels. The role that Rrp1 and Rrp2 play in centromere stability maintenance is probably based on modulation of a proper balance between histone H3 and specific histone Cnp1 levels in the centromeric region.

Although data obtained so far suggest that Rrp1 and Rrp2 function as a complex, results shown in this thesis suggest that these proteins may possess separate roles in the cell. Rrp1, but not Rrp2, physically binds to PCNA and to maintain these interaction PIM motive of Rrp1 is essential. Overproduction of Rrp1 results in a reduction in PCNA level in cells, but at the same time it leads to increase in amount of ubiquitinated forms of PCNA. It is therefore possible that Rrp1 posses ligase activity and its function is to ubiquitinate PCNA which is a signal for proteasomal degradation. Rrp2 overproduction is more toxic to cells than Rrp1 overproduction. Rrp2, but not Rrp1, is involved in telomere stability maintenance. Rrp2 role in this region is possibly due to its protection of sumoylated proteins against degradation. Rrp1 may function in the cell in an opposite way and act as SUMO-targeted ubiquitin ligase.