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Dissertation

Title	<i>MOLECULAR FUNCTIONS OF THE CHROMATIN REMODELER FUN30</i>
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Abstract	Many studies have identified conserved ATP-dependent chromatin remodeling complexes whose functions are to modulate DNA access by relieving chromatin-mediated repression. We have previously characterized Fun30 in <i>Saccharomyces cerevisiae</i> as a homodimer with ATP-dependent chromatin remodeling activity. Other studies have shown that Fun30 plays a role in maintaining the silenced state of subtelomeric and centromeric chromosomal regions. Fun30 has also been shown to play an important role in DNA damage repair by facilitating long range resection of DNA in Double Strand Breaks. This thesis focuses on understanding the mechanisms by which Fun30 is involved in DNA damage repair. Results presented here show that Fun30 can anneal complementary strands of DNA that is facilitated by ATP hydrolysis and a helicase activity in the presence of trap DNA. In addition, Fun30 was found to be able to relax both negatively and positively supercoiled DNA in an ATP-independent manner and cleave a 3' overhang in a forked DNA duplex or a duplex that has a protruding 3'. Annealing and 3' flap endonuclease activities of Fun30 suggest a mechanism by which Fun30 can facilitate double strand break repair by the Single Strand Annealing pathway, while a potential helicase activity can facilitate Synthesis Depended Strand Annealing and as a result reduce the generation of recombination intermediates. Moreover, employing in vivo approaches, we show that Fun30 genetically interacts with the Mus81 nuclease upon chronic treatment with chemicals that stall the replication fork, suggesting that Fun30 deletion might lead to the accumulation of toxic recombination intermediates that are difficult to resolve in the absence of Mus81. We also found that Fun30 deletion affects the cell cycle progression of cells lacking TopI, without affecting the viability of the cells. This might explain a function for Fun30 in facilitating the progression of the cell cycle in the presence of torsional stress which can be induced by TopI deletion. Moreover, we found that Fun30 is not involved in removing camptothecin induced TopI/DNA complexes since no genetic interaction between Tdp1 and Fun30 was observed. Furthermore, we show that Fun30 genetically interacts with Asf1 under DNA damaging conditions, suggesting that Fun30 is required in the absence of Asf1. Finally, couple of models are proposed that explain how Fun30 annealing and nuclease activities may be important in the Single Strand Annealing pathway and how Fun30 helicase activity might be used to reduce the level of toxic recombination intermediates and thus maintain genomic stability, which if compromised could lead to cancer or other diseases.