Stn1 supports Mec1 function in protecting stalled replication forks from degradation

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Abstract:

DNA replication is a highly conserved and fundamental molecular process that allows cells to duplicate their entire genome. This complex process can be disrupted by stress or DNA damage, compromising the replication fork, the structure where DNA replication takes place. Unprotected forks can collapse, leading to genome instability, a hallmark of cancer and other diseases. Replication stress threatens genome integrity by exposing replication forks to nucleolytic degradation. In both yeast and humans, the checkpoint kinases Mec1/ATR and Rad53/CHK2 limit deleterious single-stranded DNA (ssDNA), yet the protective mechanisms remain incompletely defined.

Here, we identify a role for the CST subunit Stn1 in cooperating with Mec1 to restrain ssDNA formation under nucleotide depletion. We discovered that Stn1 plays a critical role in protecting stalled replication forks from degradation, especially when Mec1 is not fully functional. Stn1 prevents the accumulation of ssDNA by limiting the nucleases Mre11, Exo1, Sgs1, the enzymes that degrade DNA, from excessively resecting replication forks. A gain-of-function allele (*stn1-L60F*) suppresses the sensitivity to replication stress of Mec1-deficient cells and reduces ssDNA at stalled replication forks, whereas a loss-of-function truncation (*stn1-ΔC*) exacerbates both phenotypes.

Our findings uncover a role for Stn1 in safeguarding genome stability by acting as a backup to checkpoint pathways to control DNA processing at stressed replication forks.