

THE RECQ HELICASE SGS1 AND EVENTS AT THE REPLICATION FORK

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The SGS1 gene in *Saccharomyces cerevisiae* is a member of the RecQ family of DNA helicases. In humans disruptions in three of the five homologues have been linked to the autosomal recessive disorders: Bloom's syndrome (BLM), Werner's syndrome (WRN), and Rothmund-Thomson's syndrome (RECQL4). It has been proposed these RecQ helicases help maintain genomic integrity during S-phase and play an important role in the S-phase checkpoint response, however the molecular mechanism for how this is achieved remains unclear. During S-phase by chromatin immunoprecipitation assays both DNA polymerases and can be seen at replication forks stalled by hydroxyurea treatment for 40-60 min. This stabilization requires the activities of both the ATM-related kinase Mec1 and Sgs1, a member of the RecQ family of DNA helicases. The Mec1-Ddc2 complex was shown to be recruited to stalled replication forks only after damage, presumably through an interaction between Ddc2 and Replication Protein A (RPA). In contrast, Sgs1 is present at forks during normal replication. It remains bound during genotoxic stress and new data suggests that Sgs1 partially contributes to Mec1-Ddc2 recruitment after HU treatment. However, an additive effect on polymerase stability and cell viability is observed in *sgs1/mec1-100* double mutant cells suggesting that the extreme synergy exhibited by mutations in these two proteins for genomic rearrangement and their differential effects on late origin firing in the presence of HU might result from Sgs1 and Mec1 contributing sequentially or independently to polymerase stabilization and replication fork recovery. Indeed, Sgs1's role in polymerase stabilization can be separated from its ability to bind and contribute to Rad53 (Chk2 kinase) activation. Since RecQ helicases act preferentially on four-way junction DNA substrates, we propose that Sgs1p acts at stalled replication forks to resolve inappropriate strand exchange structures which could contribute to the destabilization and ultimate collapse of a stalled replication forks. Consistently, its action is at least in part Top3- and Rad51-dependent.