Supplementary Information

Parental histone deposition on the replicated strands promotes error-free DNA damage tolerance and regulates drug resistance

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Table S1. *Saccharomyces cerevisiae* strains used in this study.

Strain	Relevant Genotype	Reference
FY1296	Mat a, ade2-1 trp1-1 leu2-3,112 his3-11,15 ura3 can1-100, Rad5+ (W303)	Lab collection
HY8755	W303 Mat a, ctf4delta::NAT	Lab collection
HY3373	W303 Mat a, dpb3delta::hphMX4	Lab collection
HY9391	W303 Mat a, dpb3delta::hphMX4, ctf4delta::NAT	This study
HY7220	W303 Mat a, dpb4delta::NAT	This study
HY7223	W303 Mat a, ctf4delta::TRP, dpb4delta::NAT	This study
HY7222	W303 Mat a, dpb3delta::hphMX4, dpb4delta::NAT	This study
HY7224	W303 Mat a, ctf4delta::TRP, dpb3delta::hphMX4, dpb4delta::NAT	This study
HY1788	W303 Mat a, his3-11,15::HIS3tetR-GFP (single integrant), ura3::3XURA3tetO112, omns	Lab collection
HY1854	W303 Mat a, his3-11,15::HIS3tetR-GFP (single integrant), ura3::3XURA3tetO112, omns, ctf4delta::TRP1	Lab collection
HY3927	W303 Mat a, his3-11,15::HIS3tetR-GFP (single integrant), ura3::3XURA3tetO112, omns, dpb3delta::hphMX4, ctf4delta::TRP1	Lab collection
HY3924	W303 Mat a, his3-11,15::HIS3tetR-GFP (single integrant), ura3::3XURA3tetO112, omns, dpb3delta::hphMX4	Lab collection
HY7607	W303 Mat a, dls1delta::KanMX	This study
HY7608	W303 Mat a, dls1delta::KanMX, dpb3delta::hphMX4	This study
HY11147	W303 Mat alpha, ctf4delta::NAT, dls1delta::KanMX, dpb3delta::hphMX4	This study
HY7695	W303 Mat a, ctf4delta::TRP, dls1delta::KanMX	This study
HY9374	W303 Mat a, Dpb3(K16A, K18D, K19A, K62A, K64A)-6xHIS-3FLAG::KANMX4	This study
HY10454	W303 Mat a, ctf4delta::NAT, Dpb3(K16A, K18D, K19A, K62A, K64A)-6xHIS-3FLAG::KANMX4	This study
HY2194	W303 Mat a, ctf4delta::TRP	Lab collection
HY7259	W303 Mat a, ctf4delta::TRP dpb3delta::hphMX4	Lab collection
HY4020	W303 Mat a, sgs1::pADH1-tc3-3xHA-Sgs1 (HPHMX4)	Lab collection
HY10415	W303 Mat a, sgs1::pADH1-tc3-3xHA-Sgs1 (KanMX), ctf4delta::NAT	This study

HY4213	W303 Mat a, sgs1::pADH1-tc3-3xHA-Sgs1 (NATMX), dpb3delta::hphMX4	Lab collection
HY11041	W303 Mat a, sgs1::pADH1-tc3-3xHA-Sgs1 (NATMX), dpb3delta::hphMX4, ctf4delta::KANMX	This study
HY11623	W303 Mat a, rev3delta:LEU2	This study
HY11715	W303 Mat a, ctf4delta::NAT, dpb3delta::hphMX4, rev3delta:LEU2	This study
HY11721	W303 Mat a, ctf4delta::NAT, Dpb3(K16A, K18D, K19A, K62A, K64A)-6xHIS-3FLAG::KANMX4, rev3delta:LEU2	This study
HY11055	W303 Mat a,rev1delta::KanMX4	This study
HY11073	W303 Mat a, ctf4delta::NAT, dpb3delta::hphMX4 rev1delta::KanMX4	This study
HY11366	W303 Mat a, ctf4delta::NAT, Dpb3(K16A, K18D, K19A, K62A, K64A)-6xHIS-3FLAG::KANMX4, rev1delta::KANMX4	This study
HY6848	W303 Mat a, sgs1delta::HIS3	Lab collection
HY3829	W303 Mat a, sgs1::AUR1, dpb3delta::hphMX4,	Lab collection
HY12065	W303 Mat a, sgs1::HIS3, dpb3delta::hphMX4,	Lab collection
HY11375	W303 Mat a, sgs1::HIS3, Dpb3(K16A, K18D, K19A, K62A, K64A)-6xHIS-3FLAG::KANMX4	This study
HY11468	W303 Mat a, pol1-2A2, sgs1::HIS3	This study
HY11425	W303 Mat a, mcm2-3A::hphNT, sgs1delta::HIS3	This study
HY11466	W303 Mat a, pol1-2A2, sgs1::pADH1-tc3-3xHA-Sgs1 (HPHMX4)	This study
HY11470	W303 Mat a, mcm2-3A::hphNT, sgs1::pADH1-tc3-3xHA-Sgs1 (NATMX)	This study
HY8323	W303 Mat a, ctf4-4E	K. Labib lab
HY7667	W303 Mat a, dpb3delta::hphMX4, ctf4-4E	This study
HY7902	W303 Mat a, ctf4-3E	K. Labib lab
HY7669	W303 Mat a, dpb3delta::hphMX4, ctf4-3E	This study
HY4160	W303 Mat a, mms22delta::HIS	This study
HY12186	W303 Mat a, ctf4delta::NAT, mms22delta::KAN	This study
HY12187	W303 Mat a, dpb3delta::hphMX4, mms22delta::KAN	This study
HY12188	W303 Mat a, dpb3delta::hphMX4, ctf4delta::NAT, mms22delta::KAN	This study

HY12306	W303 Mat a, mcm2-3A::hphMX4	Z. Zhang lab
HY11374	W303 Mat a, pol1-2A2	Z. Zhang lab
HY11799	W303 Mat a, mcm2-3A::hphMX4, rev3delta:LEU2	This study
HY9730	W303 Mat a, ctf4(460-927)-3FLAG(KANMX4)	This study
HY12411	W303 Mat a, ctf4(460-927)-3FLAG(KANMX), dpb3delta::hphMX4	This study
HY12412	W303 Mat alpha, ctf4delta::NAT, mcm2-3A:: hphMX4	This study
HY12413	W303 Mat alpha, ctf4delta::NAT, pol1-2A2	This study
FY0001	Mat alpha ade2-101 leu2-3,112 lys2-801 ura3-52 his3-delta200 (PY83)	Lab collection
HY3466	PY83 Mat alpha, ctf4delta::hphMX4	Lab collection
HY8684	PY83 Mat alpha, dpb3delta::NAT	This study
HY8683	PY83 Mat alpha, ctf4delta::hphMX4, dpb3delta::NAT	This study
HY8909	PY83 Mat alpha, ctf4delta::HPH, dpb3delta::NAT, rev3delta::KANMX	This study
FY2040	Mat alpha, can1delta::STE2pr-Sp_his5, lyp1delta, his3delta1, leu2delta0, ura3delta0, met15delta0, LYS2+	M.Foiani lab

Supplementary figures

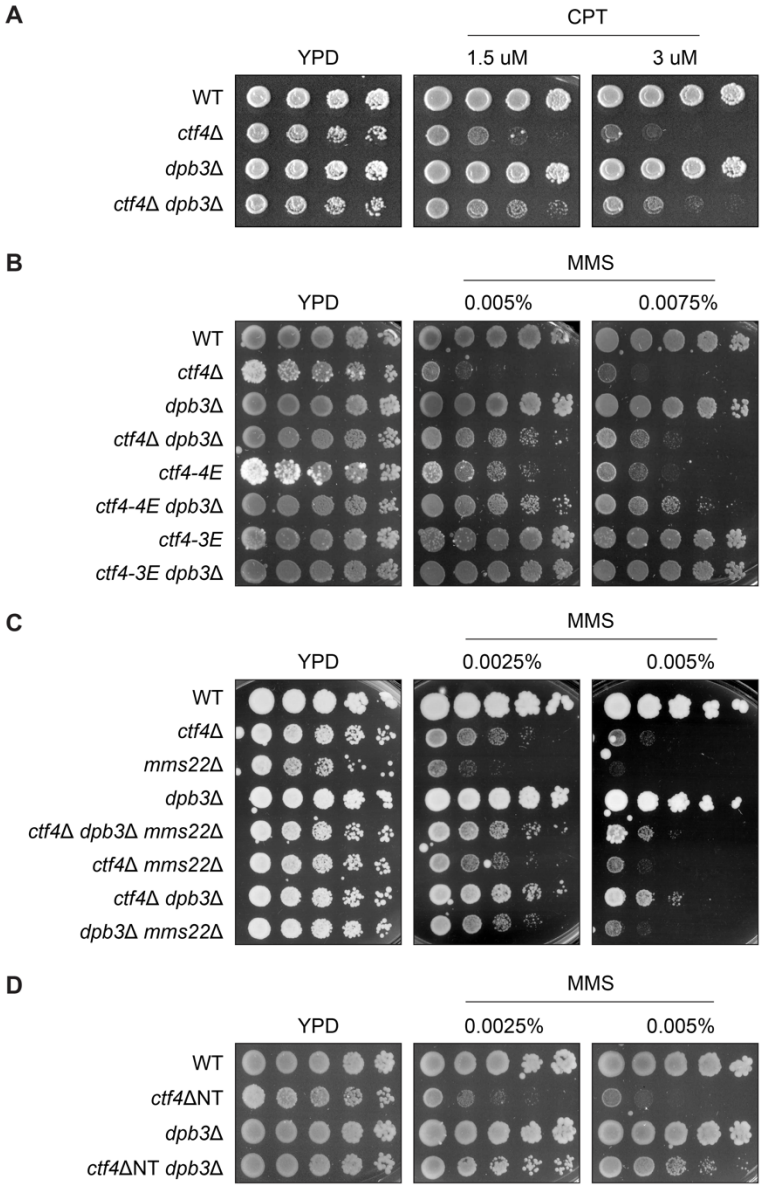

Figure S1. Ctf4 and Mms22 roles in the context of the replisome are being affected by Dpb3

(A-D) CPT and MMS sensitivity assay of cells with the indicated genotypes. Cells were grown overnight at 28°C, serially diluted and spotted on the indicated CPT and MMS concentration to test their sensitivity to the drug. Images were taken after 3 days of incubation at 28°C in two independent experiments.

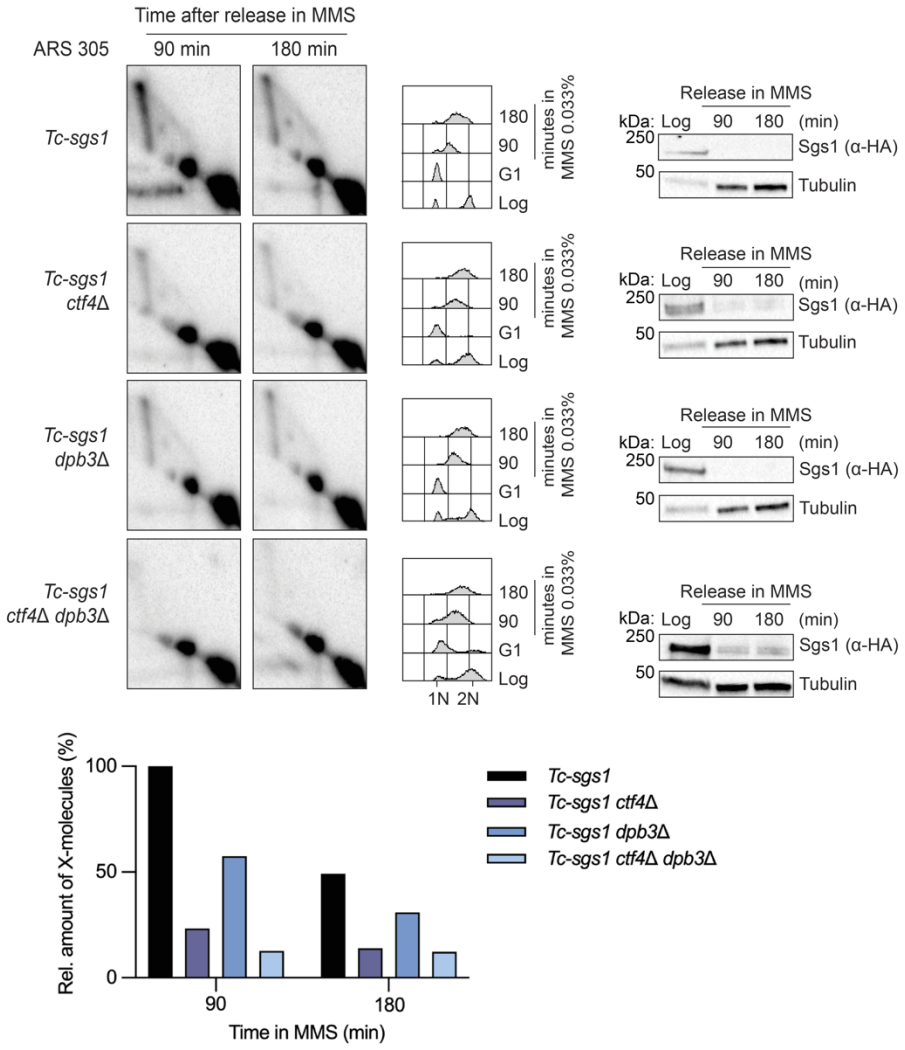

Figure S2. Error-free DNA damage tolerance via template switching is mediated by Ctf4 and Dpb3

2D gel analysis of DNA replication and recombination intermediates accumulating in the ARS305 region of the cells with the indicated genotypes. The strains with the indicated backgrounds were synchronized in G1 with alpha-factor at 25°C and released at 30°C in YPD supplemented with 0.033% MMS. Depletion of Sgs1 was achieved by adding Tetracycline 1 mM to YPD medium during G1 arrest and release. Cells were collected at the indicated time points, subjected to in vivo psoralen-mediated inter-strand DNA crosslinking and genomic DNA was extracted and digested with *Eco*RV and *Hind*III restriction enzymes for 2D gel

analysis of the DNA replication and recombination intermediates accumulating in the ARS305 region. Depletion of HA-Sgs1 was monitored by western blot, using tubulin as a loading control. Cell cycle progression was monitored by FACS analysis. 1N and 2N indicate G1 and G2/M cell cycle phases respectively. Quantification of the X-molecules signals is reported in the histogram. The intensity of the signals was normalized to the monomer spot and shown in the histograms relative to the highest signal assigned as 100%.

Strain	MMS-induced mutation rate *CAN1* (X 10^{-6})
WT	1.6 [-0.4/+0.4]
ctf4Δ	5.1 [-1.6/+1.6]
dpb3Δ	3.3 [-1.0/+1.0]
ctf4Δ dpb3Δ	16.7 [-4.8/+4.7]

Figure S3. Ctf4 and Dpb3 synergistically suppress mutagenesis

MMS-induced mutation rates at CAN1 locus (x10^{-6}) in cells with the indicated genotypes. Cells were treated with MMS 0.005% for 4 hours before proceeding with the mutagenesis assay. Mutation rates with 95% confidence intervals were calculated using the generating function (GF) estimator software bz-rates.

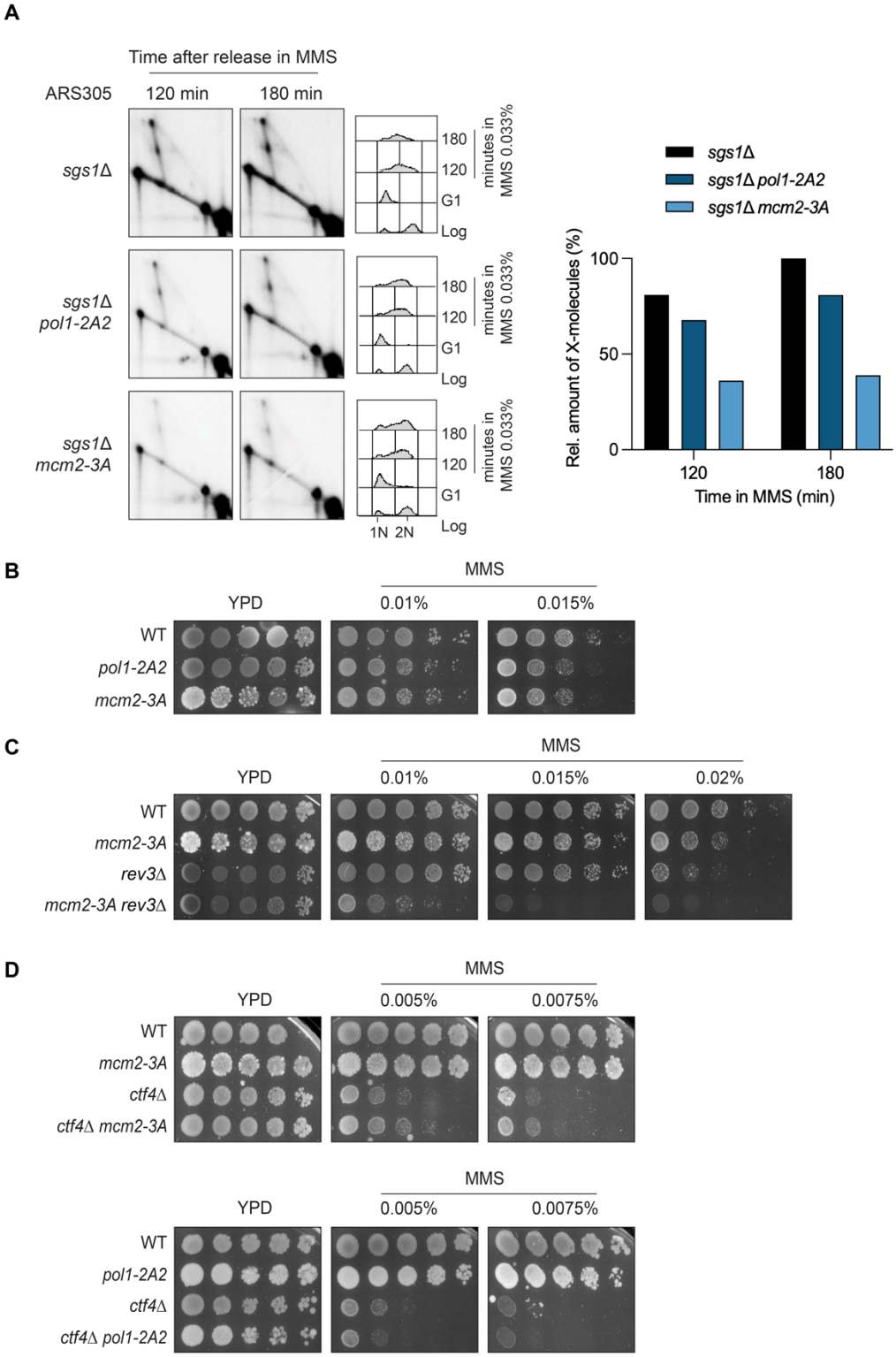

Figure S4. Replication-coupled recombination relies on Mcm2-Ctf4-Polα axis of parental nucleosome deposition that synergizes with loss of DNA polymerase zeta

(A) 2D gel analysis of DNA replication and recombination intermediates extracted from cells with the indicated genotypes. The strains with the indicated genetic backgrounds were synchronized in G1 with alpha-factor at 25°C and released at 30°C in YPD supplemented with 0.033% MMS. Cells were collected at the indicated time point, subjected to in vivo psoralen-mediated DNA inter-strand crosslinking and genomic DNA was extracted and was digested with *Nco*I restriction enzyme for 2D gel analysis of the DNA replication intermediates accumulating in the ARS305 region. Cell cycle progression was monitored by FACS analysis. 1N and 2N indicate G1 and G2/M cell cycle phases respectively. Quantification of the X-molecules signals is reported in the histogram. The intensity of the signals was normalized to the monomer spot and shown in the histograms relative to the highest signal assigned as 100%. **(B, C)** Synergistic effect of *REV3* deletion on the MMS sensitivity of *pol1-2A2* and *mcm2-3A* mutant cells. The cells with the indicated genotypes were grown overnight in liquid medium and serial dilutions of cells were spotted on YPD plates with and without the indicated concentrations of MMS. After 3 days of incubation at 28°C, the plates were photographed. Similar results were obtained in two independent experiments. **(D)** *mcm2-3A* and *pol1-2A2* effect on *ctf4*Δ cells MMS sensitivity. Cells of the indicated genotypes were grown overnight at 28°C, serially diluted and spotted on YPD plates containing MMS at the indicated concentrations. Cells were allowed to grow for three days at 28°C before images were taken. Three independent experiments were performed showing similar results.