Legends to Supplementary Figures

**Supplementary Figure 1** PCNA ubiquitination can be induced by DNA-damage in S-phase only. a, In G1 or G2/M-arrested cells, DNA damage does not induced PCNA ubiquitination. Cells arrested in G1 by α-factor or at the G2/M transition by nocodazole were treated with 0.02% MMS for 1 or 2 hours. Cell cycle arrest was controlled by microscopic inspection and anti-Clb2 Western blots. b, PCNA ubiquitination can be induced by hydroxyurea (HU), a drug that leads to replication fork stalling in S-phase. Cells were treated with 200mM HU added directly to the growth medium, and PCNA ubiquitination in these cells was visualized by anti-PCNA Western blots.

**Supplementary Figure 2** Survival rates after UV-light irradiation. a-b, DNA-damage sensitivity of rad5 and mms2 mutants deficient in error-free postreplicative lesion bypass is suppressed by siz1 mutation in dependency of RAD52. c, siz1 and rad52 have a non-epistatic relationship. d-e, Suppression of rad18 by deficiency in PCNA SUMOylation (siz1) is dependent on RAD52, RAD51, RAD54, RAD57, which make up the central components of homologous recombination, but not RAD50 or RAD59. f, Rescue of rad6 by siz1 does not require the nucleotide excision repair gene RAD2. g-h, Suppressive effects of mutation of K127 in the pol30 K164R background is visible only in the presence of functional RAD52. g, PCNA is not the only target of Rad6, as rad6 rad52 pol30K164R is more sensitive than rad52 pol30K164R. a-h, Shown are average values from independent experiment, error bars represent standard deviations.

**Supplementary Figure 3** PCNA SUMOylation levels are influenced by Srs2 a, Overexpression of the Srs2 fragment, which binds SUMOylated PCNA, increases the steady-state level of PCNA SUMOylation, without affecting its cell-cycle regulation. PCNA SUMOylation in WT and srs2 and in cells overexpressing
Srs2ΔN (Yiplac211::ADH-myc-Srs2ΔN), after α-factor arrest/release, was visualized by anti-PCNA Western blots. b, PCNA SUMOylation is not affected by overexpression of Srs2 mutants that lack the ability to bind SUMOylated PCNA. Myc-fusions of the Srs2ΔN fragment that binds PCNA and of different C-terminal deletions (of 8, 24 or 136 aa) of this fragment, that lack the SUMO-PCNA-binding site, were overexpressed and the PCNA SUMOylation was detected by anti-PCNA Western blots.

**Supplementary Figure 4** Survival rates in response to UV-light irradiation. a, siz1 and srs2 mutants rescue rad5 similarly and epistatically. In the triple mutant rad5 siz1 srs2 no additional rescue but even enhanced sensitivity compared to rad5 srs2 is observed. b, siz1 and srs2 have a non-epistatic relationship. c, Mutation of SUMOylation site K164 leads to a small, additional phenotype in a rad6 srs2 background. d-e, Deletions of the C-terminal tail of Srs2, which are not able to bind SUMOylated PCNA, rescue rad6 or rad18 damage sensitivity similarly to siz1. a-e, Average values from independent experiment are depicted with standard deviations as error bars.

**Supplementary Figure 5** The PCNA-SUMO-Srs2 check operates in diploid cells. a-b, siz1 and srs2ΔC mutants suppress the rad18 DNA-damage sensitivity in homozygous diploid cells. Specifically in diploids the srs2 deletion mutant shows a strong hypersensitivity, which probably arises through misregulated recombination between sister chromosomes. Shown are serial dilutions of cells on plates containing the indicated concentration of MMS (right) and the quantification of survival rates after UV-light irradiation (left) with standard deviations of independent experiments as error bars.