

Supplementary Figure Legends

Figure S1. Cell viability was estimated by the number of colonies that were formed after a five-day incubation from 300 cells plated on rich media after IR (500 grays), and expressed as a proportion of the untreated sample. The experiment was repeated three times and survival represent the media of the three experiments with standard deviations.

Figure S2. Wild-type and *rgf1Δ* cells were cultured to mid-log phase in YES medium at 28°C, treated with PhI for 10min and then released into fresh medium without PhI at the same temperature. Tukey boxplot illustrating quantitative analysis of the size distribution of individual cells ($n > 100$) in a population of each strain and condition (right). One experiment representative of three is shown. Statistical significance was calculated using two-tailed unpaired Student's test. ** $p < 0.01$, *** $p < 0.001$; ns=non-significant.

Figure S3. Serial dilutions of the indicated strains (1, 0.5, 0.25, 0.025, 0.0025 and 0.00025) were spotted on rich YES plates containing the indicated DNA-damaging agents. Colony formation was analyzed after 3-4 days at 28°C, 2-3 days 32°C and 2 days 37°C. (bottom panels) *rgf1Δrho1-596*, *rgf1Δpck2Δ* and *rgf1Δpmk1Δ* mutants did not show synergism in their sensitivity to PhI as compared with the parental *rgf1Δ* deletion. The combination *rgf1Δpck1Δ* produced mutant cells more sensitive to PhI than any of the single mutations.

Figure S4. Wild-type cells containing an HA-tagged Cds1p were grown to log-phase and treated with 12.5 mM HU for 3h and with 10μg/ml PhI for 2h. Protein extracts were analyzed by western blot in phostag gels and the membranes were probed with anti-HA to visualize Cds1p.

Figure S5. (A) Biological repeats of the PFGE analysis shown in Figure 5A. Wild-type, *rgf1Δ* and *rad51Δ* cells were treated with 10μg/ml PhI for 30 min, and agarose plugs were prepared before treatment, immediately, at 6h (2.0 generations) and at 15h (3.5 generations) after treatment. The three chromosomes were fragmented by PhI treatment in the three strains, but re-assembled within 3.5 generations, only in the wild-type cells. (B) Wild-type cells were cultured to mid-log phase in YES medium and treated with PhI for the times indicated. The number of foci in 200 nuclei for each time was scored in three independent experiments.

Figure S6. Wild-type and *rgf1Δ* cells expressing Rad52p-YFP from the chromosome were cultured to mid-log phase in YES medium, treated with PhI for 10 min and then released into fresh medium without PhI in the presence of cycloheximide (CHX) 100μg/ml. Protein extracts from each of the indicated time-points were analyzed by western blot and the membrane was probed with anti-GFP to visualize Rad52p and anti-α-tubulin as a loading control. The graph shows the amount of Rad52p-YFP for each strain and time point starting 5h after CHX addition.

Figure S7. Serial dilutions of the indicated strains (1, 0.5, 0.25, 0.025, 0.0025 and 0.00025) were spotted on rich YES plates with or without PhI. Colony formation was analyzed after 3 days as indicated.

Fig. S1

% Survival		
Strain	no treatment	after 500 GI
wt	100	74,75 ± 4,99
<i>rgf1</i>	100	52,12 ± 5,69
<i>rho1-596</i>	100	56,59 ± 6,18
<i>rad51</i>	100	0

Fig. S2

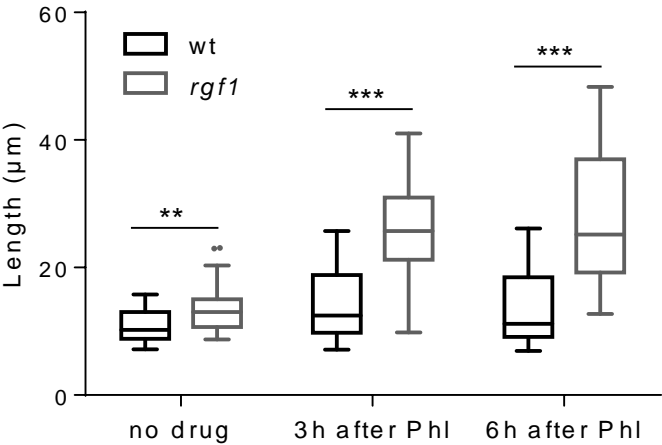


Fig. S3

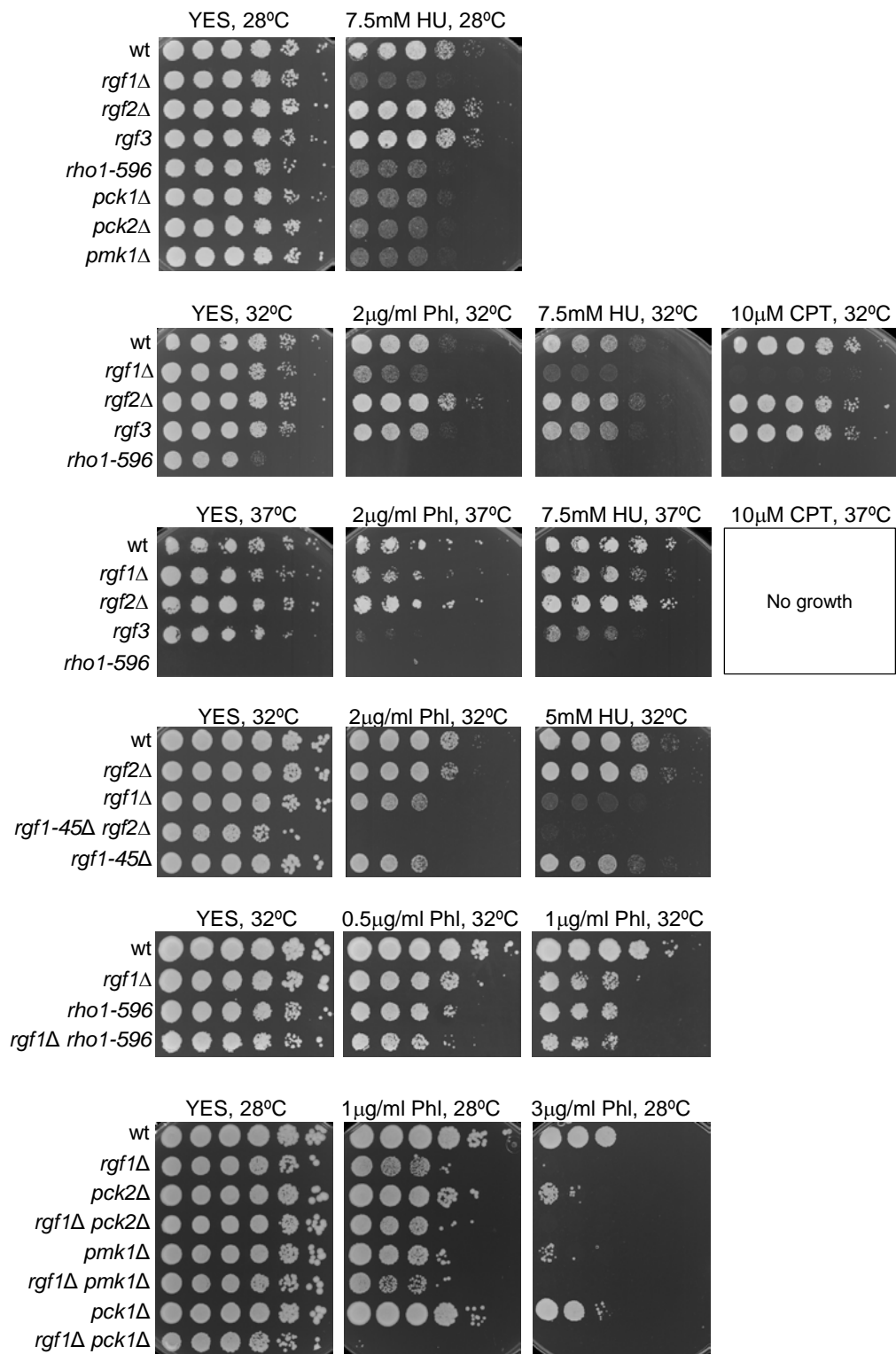


Fig. S4

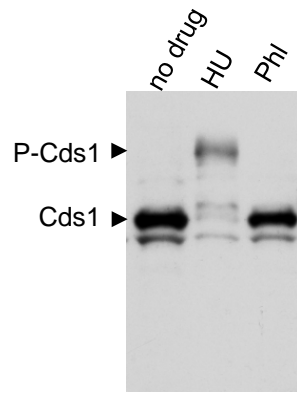
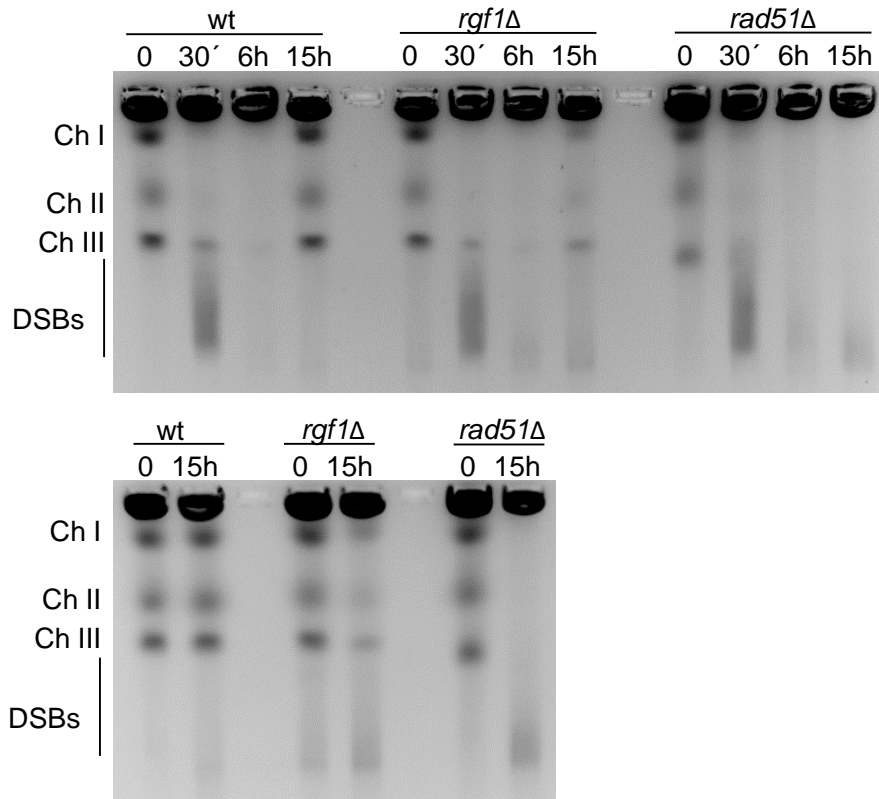


Fig. S5

A



B

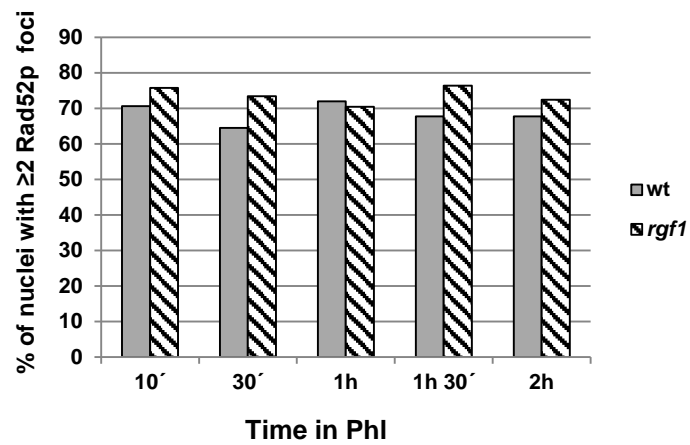


Fig. S6

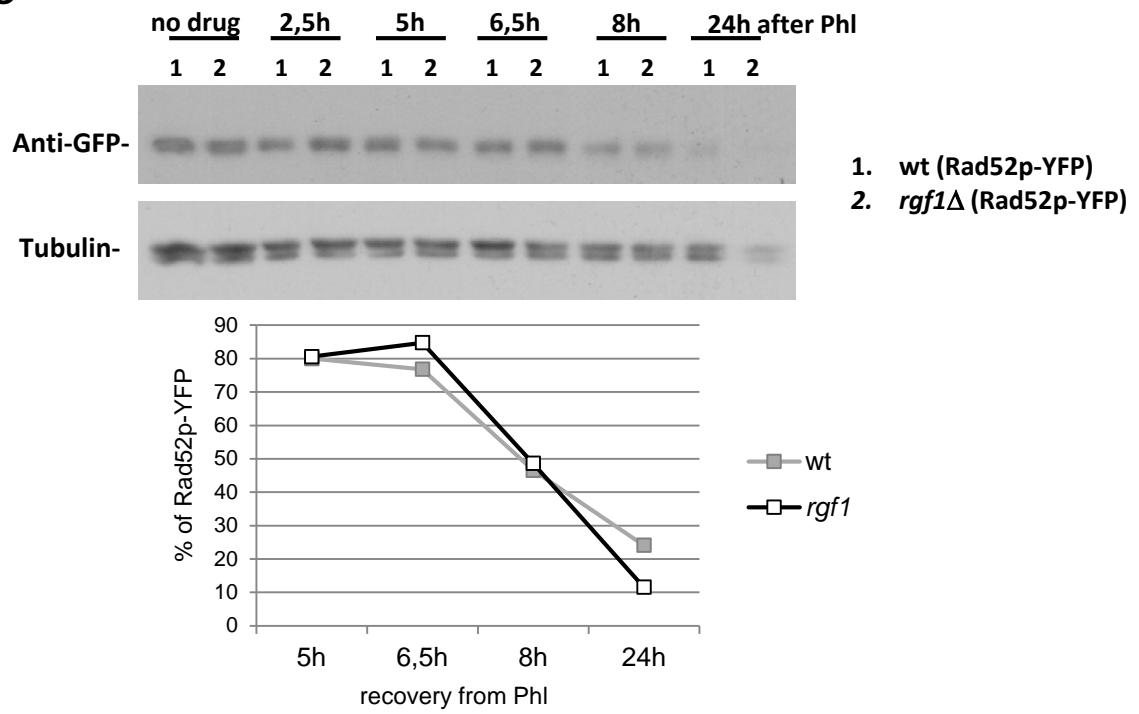
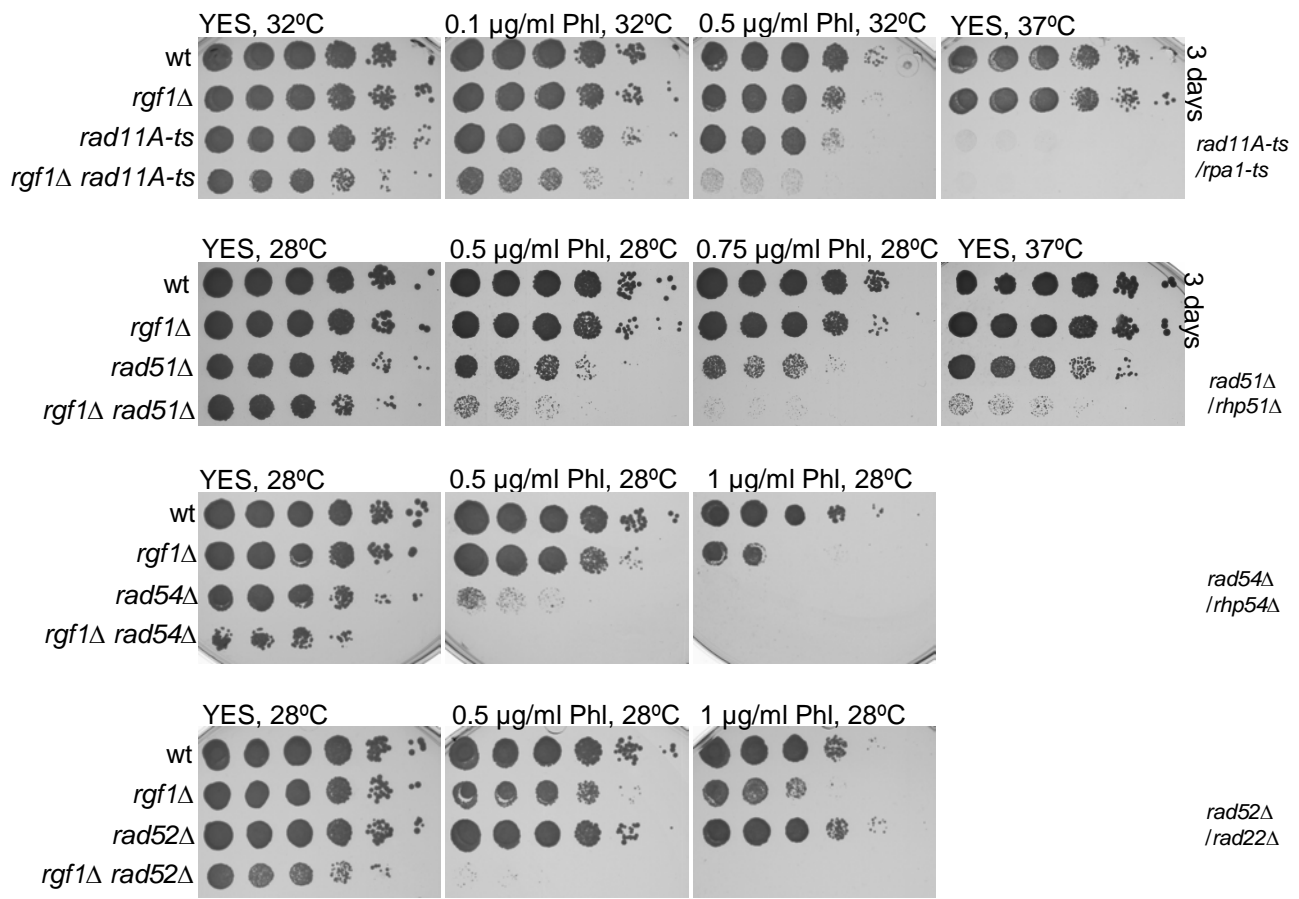


Fig. S7

Core repair proteins



Figures S1-S7, Manjón *et al*, 2016