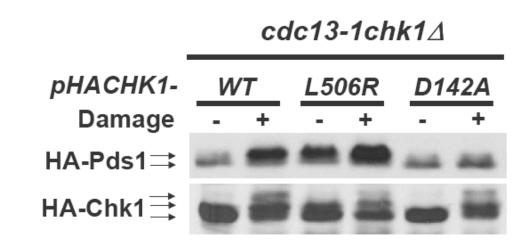
## Supplemental figure S1

A. Constructs encoding the indicated HA-tagged *CHK1* alleles were transformed into a *cdc13-1*  $chk1 \otimes HA-PDS1$  yeast strain and transformants were grown at the permissive temperature (no DNA damage) and restrictive temperature (DNA damage) for *cdc13-1*. Chk1 and Pds1 were visualized by Western analysis using anti-HA antibodies.

**B.** Constructs encoding the indicated HA-tagged *CHK1* alleles (used in A) were transformed into a  $chk1\otimes$  yeast strain and transformants were grown in SC media lacking Leucine at 30°C and collected for analyses when cells were in logarithmic growth phase (OD<sub>600</sub> 0.5). DNA content was visualized by staining cells with propidium iodide and the DNA content histograms were generated by FACS as in (5). The data from the histograms was analyzed with ModFit software to calculate cell cycle distribution.

## Supplemental figure S2

 $chk1 \otimes$  cells were co-transformed with a plasmid expressing untagged *CHK1*, *CHK1L506R* or empty vector and a plasmid carrying an HA-tagged *CHK1* allele. HA-tagged Chk1 proteins in the absence of DNA damage were analyzed by Western blotting.

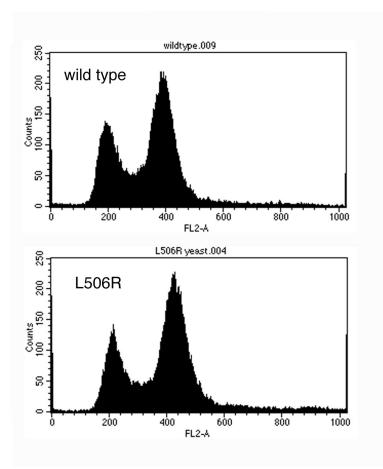


Β

Α

Wild type: G1 = 23.07 G2/M = 55.95 S = 20.98

Chk1L506R: G1 = 20.90 G2/M = 59.39 S = 19.71



## Supplemental Fig. 2S

