Expanded View Figures

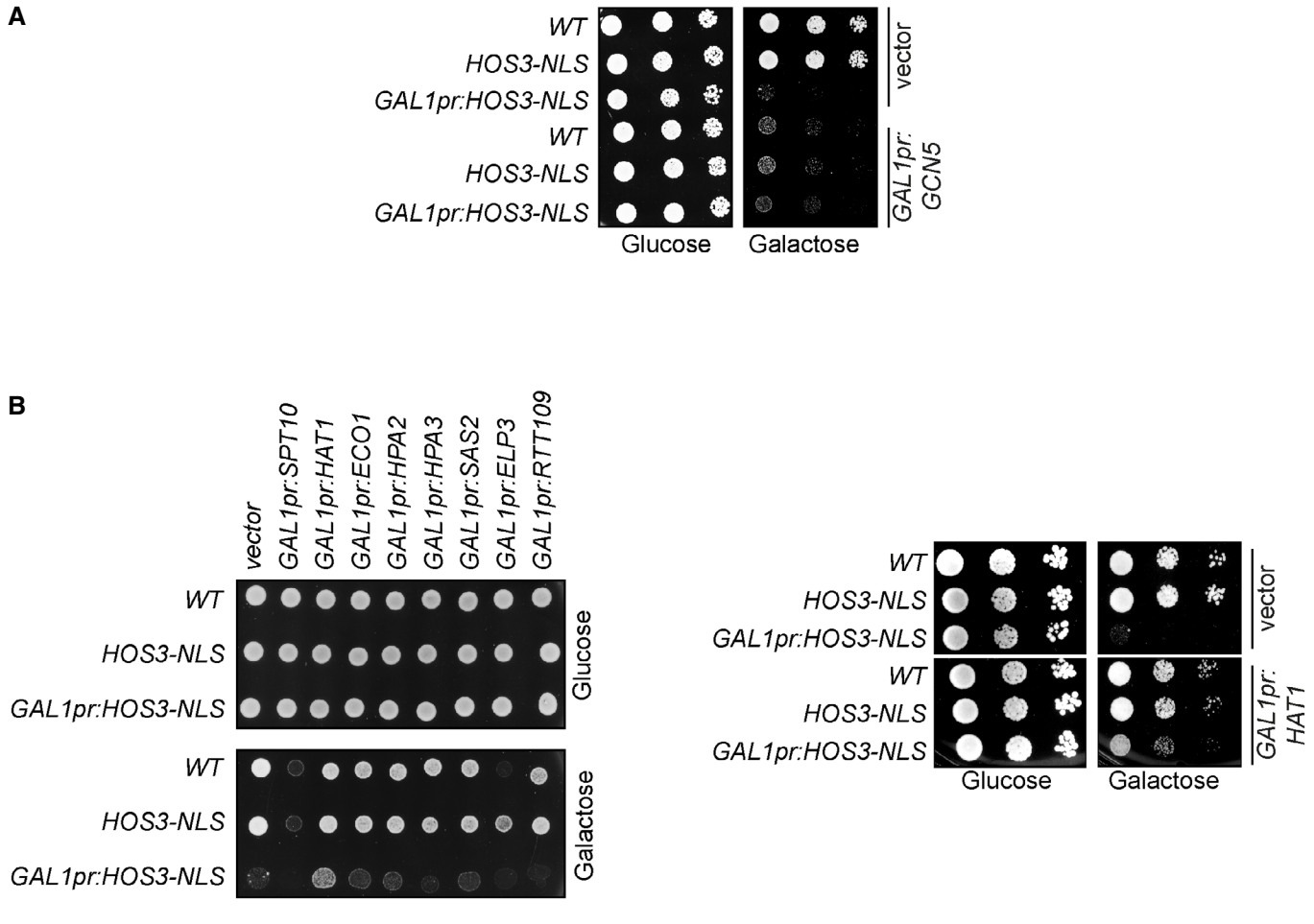

Figure EV1. Effect of KAT overexpression in Hos3-NLS-dependent growth inhibition.

A Overexpression of the KAT Gcn5 is toxic. 10-fold serial dilutions of wild-type (*WT*), *HOS3-NLS-GFP* and *GAL1pr:HOS3-NLS-GFP* (single copy at the endogenous locus) transformed with an empty vector or the *GAL1pr:GCN5-HA* plasmid were spotted onto SC-Glu and SC-Gal medium and incubated at 25°C for 3 days.

B Role of KAT overexpression in cell viability and ability to rescue growth in the presence of overexpressed *HOS3-NLS*. (*Left*) Exponential cultures of the indicated strains transformed with an empty vector or plasmids overexpressing the KATs Spt10, Eco1, Hpa2, Hpa3, Sas2, Elp3 or Rtt109 were spotted onto SC-Glu and SC-Gal medium and incubated at 25°C for 3 days. (*Right*) The effect of *HAT1* overexpression was also assessed in 10-fold serial dilutions as in (A).

Figure EV2. Role of *ESA1*, *GCN5* and *HAT1* in Start.

A *esa1-ts* and *gcn5Δ esa1-ts* mutants have bud emergence defects. Cells of the indicated strains were arrested in G1 by treatment with α-factor for 2.5 h at 25°C, shifted to 37°C for 1 h and released from the G1 arrest at 37°C. Cells were fixed at the indicated times, and the presence of buds (arrowheads) was assessed by microscopy.

B *HAT1* is not involved in Start. Bright-field images of cells of the indicated strains, treated as in (A) and scored at the indicated times after α-factor washout. The left subpanel shows the fraction of budded cells in one of two independent experiments with similar results. At least 200 cells were scored for each strain and time point. Arrowheads point to cell buds. Scale bars in (A and B), 5 μm.

Source data are available online for this figure.

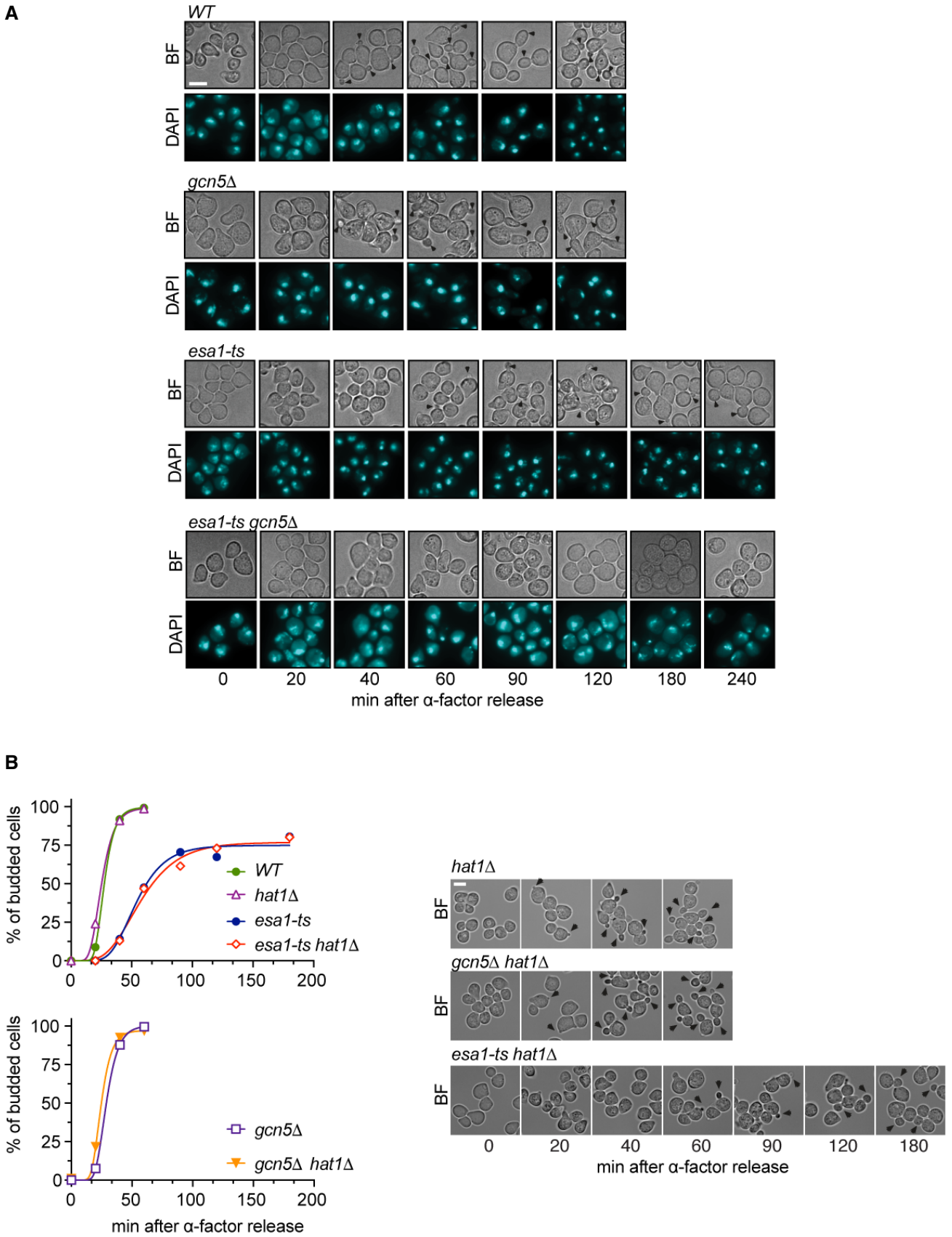

Figure EV2.

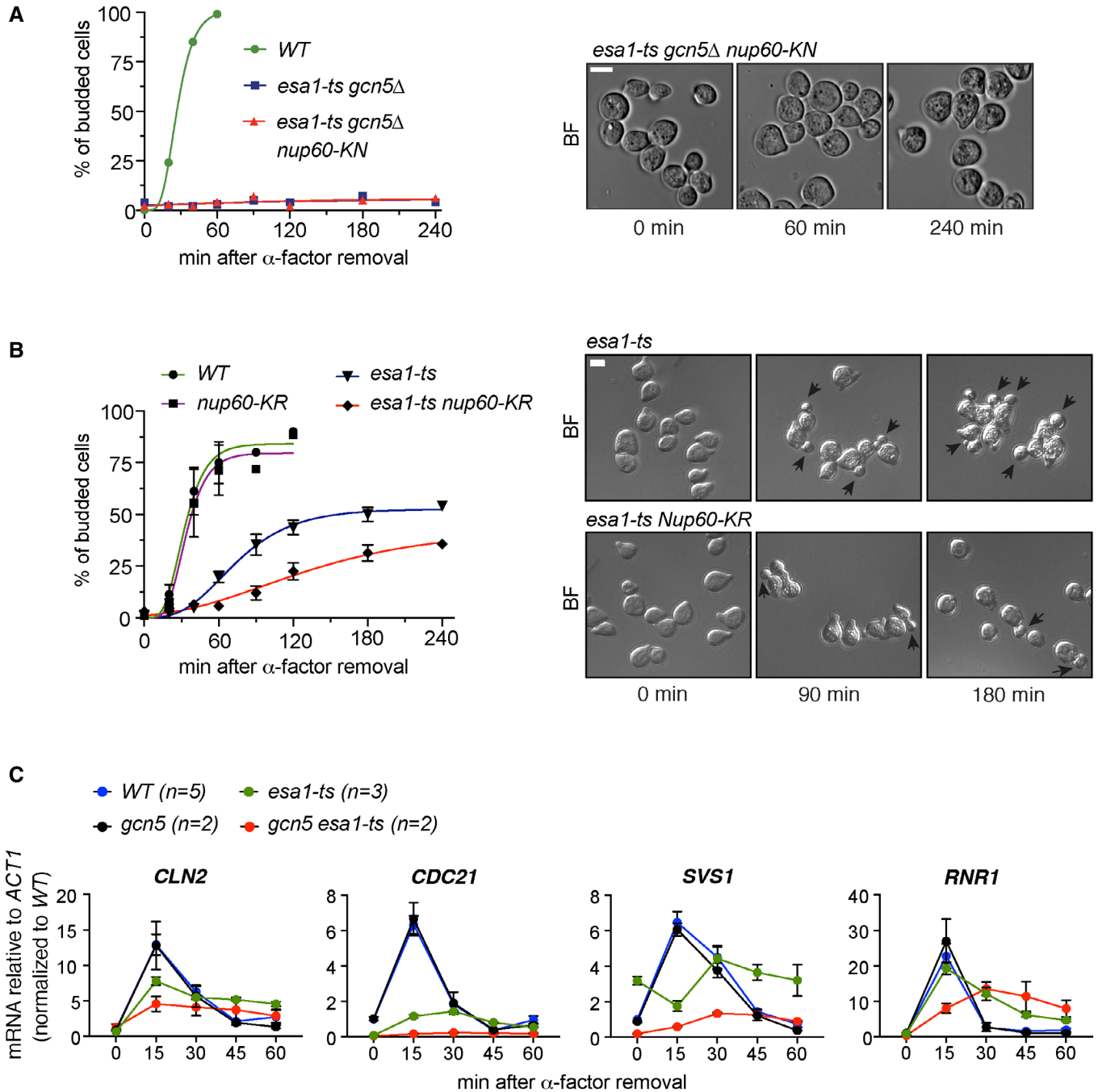

Figure EV3. G1/S transition in KAT and Nup60 KN/KR mutants.

A Acetyl-mimic Nup60-KN does not rescue the budding defect of the double mutant *esa1-ts gcn5*Δ. Cells were arrested in G1 by treatment with α-factor for 2.5 h at 25°C, shifted to 37°C for 1 h and released from the G1 arrest at 37°C. Cells were fixed at the indicated times, and the presence of buds was assessed by microscopy. The left subpanel shows the fraction of budded cells in one of two independent experiments with similar results.

B Non-acetylatable Nup60-KR increases the budding defect of *esa1-ts* cells. In (A and B), at least 200 cells were scored for each strain and time point. In (B), data from three independent experiments are represented as mean and SEM. In bright-field images of cells at the indicated times after α-factor washout, arrowheads point to cell buds.

C mRNA levels of *CLN2*, *CDC21*, *SVS1* and *RNR1* were determined for cells of the indicated strains after G1 arrest and release at restrictive temperature, with samples collected at indicated times. Data from $n > 2$ independent experiments are represented as mean and SEM (*WT*, *esa1-ts*) and data from $n = 2$ independent experiments (*gcn5*Δ, *gcn5*Δ *esa1-ts*) as mean and range. The $2^{\Delta Ct}$ values were then normalised relative to the wild-type value at 0 min.

Data information: (A, B) Scale bar, 4 μm.
Source data are available online for this figure.

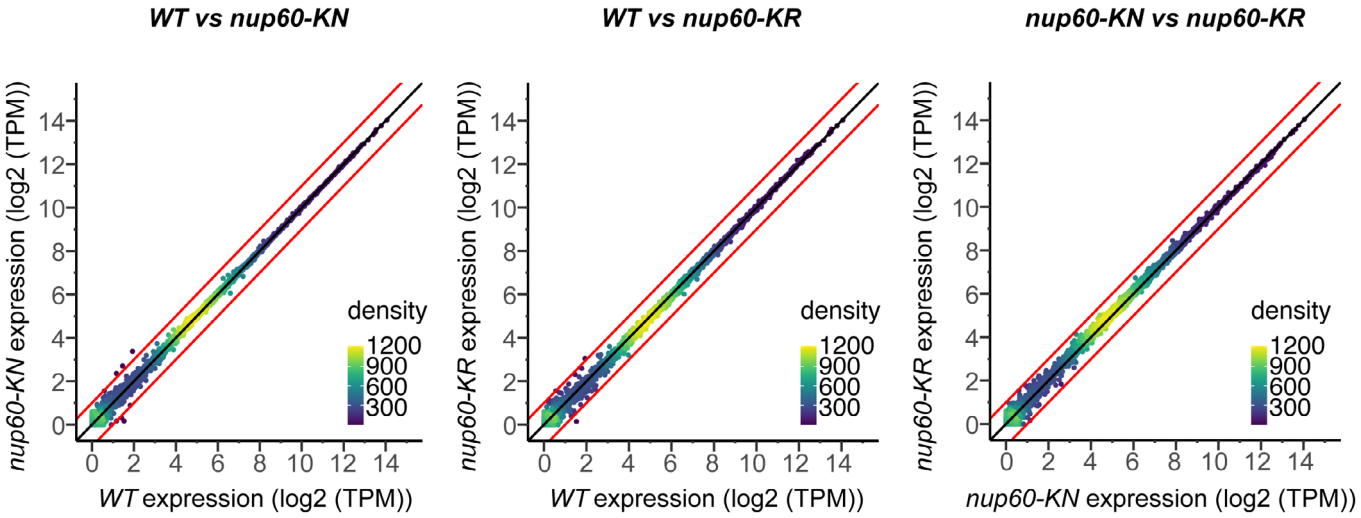

Figure EV4. Nup60-KN and Nup60-KR alleles do not alter mRNA levels.

Expression levels in *nup60-KN* and *nup60-KR* strains compared to expression in the wild-type strain (*WT*) and to each other. Red lines show a fold change of 1. Genes with FC > 1 are *YCR107W*, *YCR106W* and *YGL263W* (subtelomeric); genes with FC < −1 are *YFL065C* (subtelomeric), *YLR124W* and *tF(GAA)F* (phenylalanine tRNA).

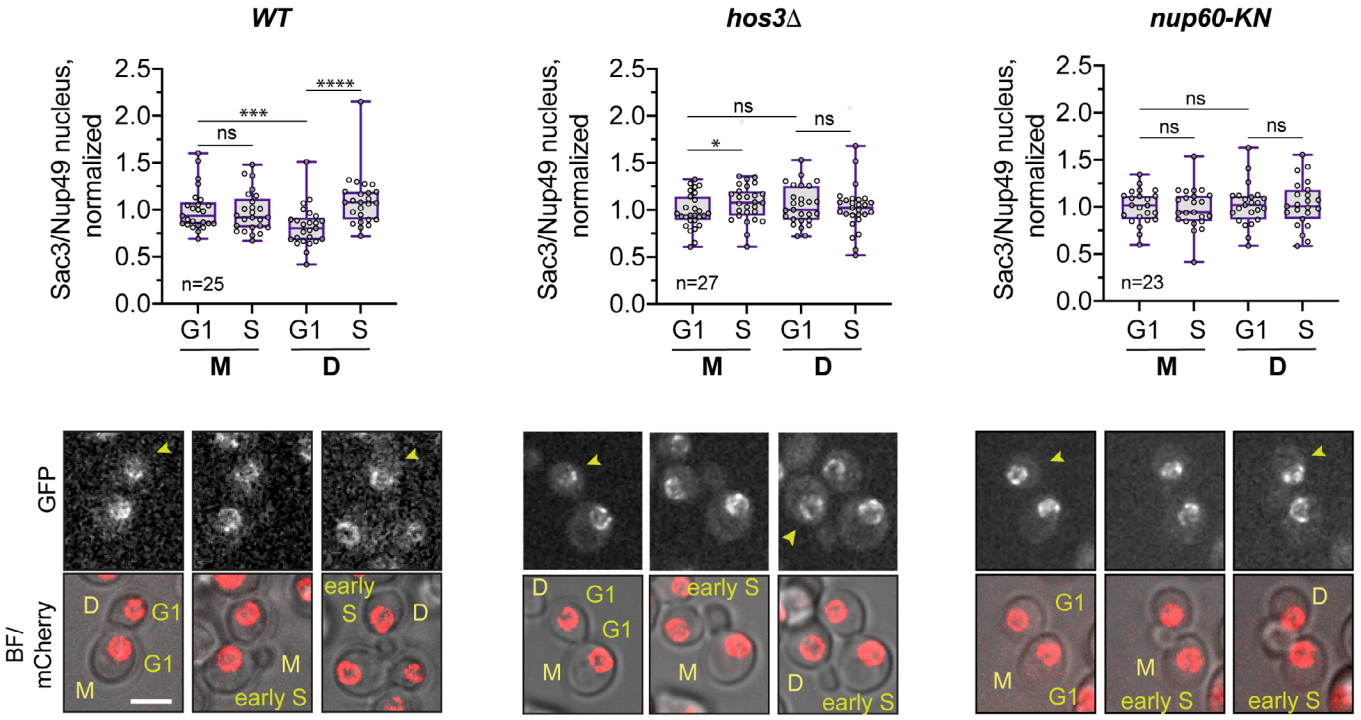

Figure EV5. Sac3 recruitment to daughter cell nuclei in G1 and S phase.

Cells of the indicated strains were imaged by time-lapse microscopy, and the fluorescence levels of Sac3-GFP and Nup49-mCherry were determined in G1 (unbudded cells after cytokinesis) and S phase (cells with small buds). The NPC component Nup49 was used as a control for nuclear pore complex protein levels. Fluorescence intensity was measured in sum projections of whole-cell Z-stacks, by segmentation of the nuclear area in the mCherry channel. The ratio of Sac3 to Nup49 intensities was then normalised relative to the mean intensity of wild-type mothers. Boxes include 50% of data points, the line represents the median, and whiskers extend to maximum and minimum values. ****, $P \leq 0.0001$; ***, $P < 0.001$, *, $P \leq 0.05$; and ns, $P > 0.05$, two-tailed paired *t*-test. Scale bar, 4 μm. *n* = number of cells, pooled from three independent experiments with similar results. Arrows point to daughter cells.

Source data are available online for this figure.