Supplemental Information

Regulation of the Hsf1-dependent transcriptome via conserved bipartite contacts with Hsp70 promotes survival in yeast

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Figs. S1-S5

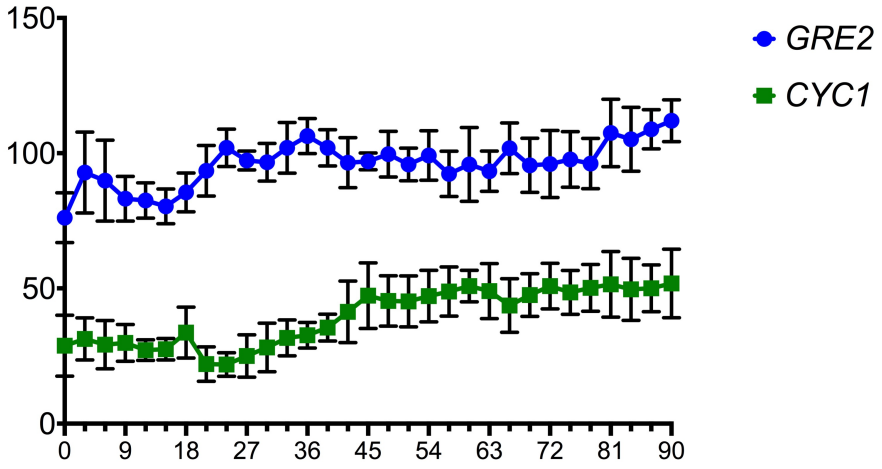

Fig. S1. LucCP+ activity at 37°C from unrelated non-stress induced *CYC1* promoter, and parent vector *GRE2* promoter. 90 min heat shock at 37°C heat shock of BY4741 carrying plasmids with lucCP+ under control of the unrelated *CYC1* promoter or the *GRE2* promoter vector used to create the HSE-lucCP+ reporter demonstrating lack of heat inducibility.

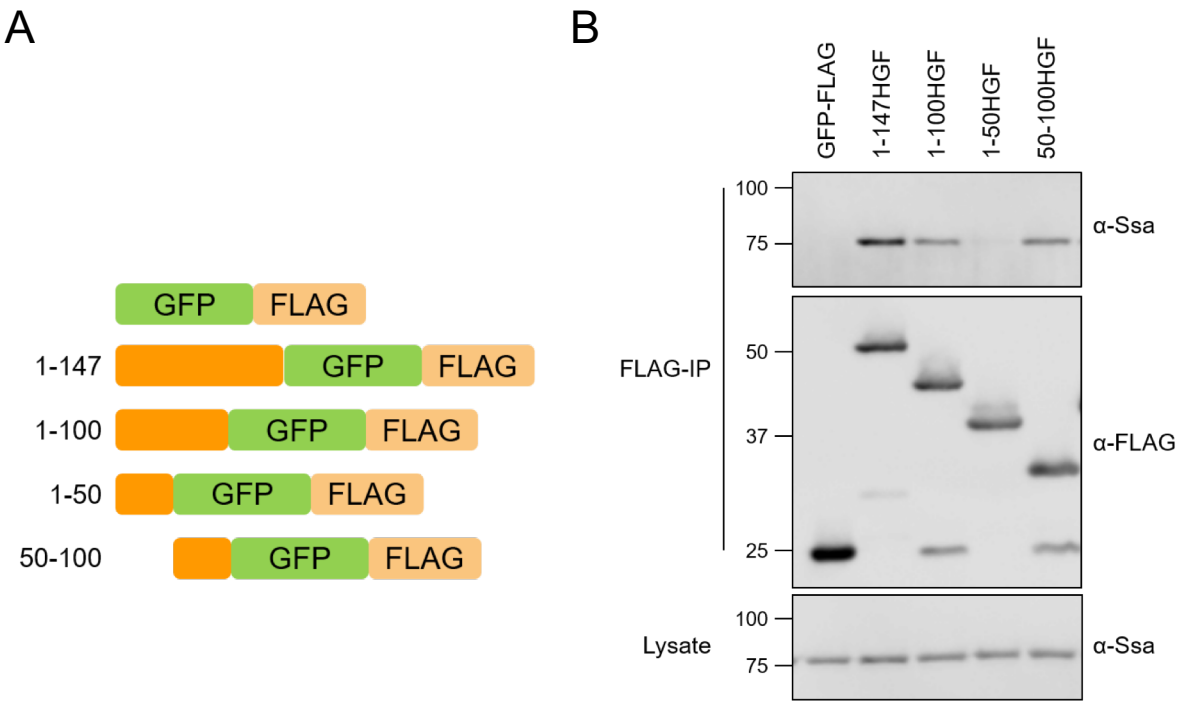

Fig. S2. Amino-terminal residues 50-100 are sufficient to interact with Ssa1. *a,* Hsf1 amino-terminal activation domain truncation constructs fused to GFP-FLAG. *b,* Western blots of lysates and FLAG immunoprecipitation of Hsf1-GFP-FLAG protein fusions, labeled 'HGF', with co-immunoprecipitation of Ssa.

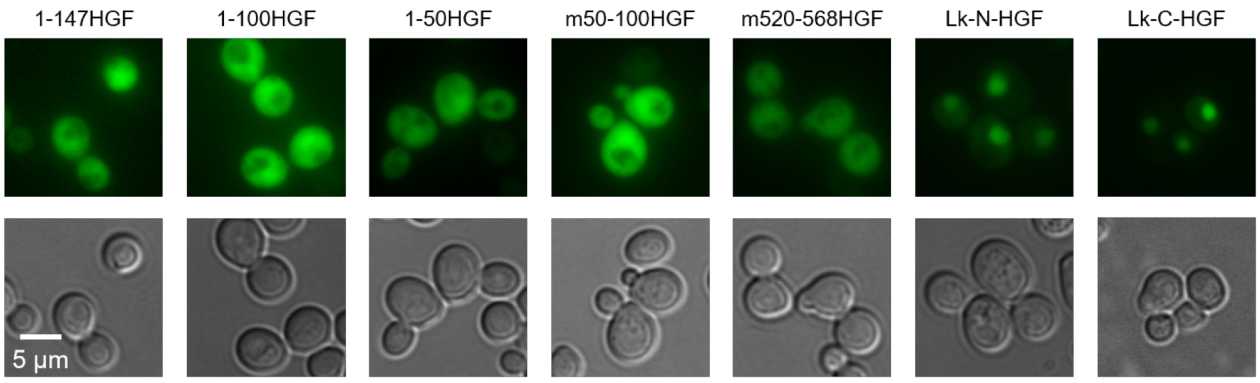

Fig. S3. Hsf1-GFP-FLAG fusion proteins are primarily diffuse. Microscopy of the indicated *S. cerevisiae* Hsf1-GFP-FLAG fusion constructs demonstrates diffuse localization throughout the cell. The *L. kluyveri* N-AD and C-AD constructs are likewise diffuse but localized within the nucleus, possibly by unidentified nuclear localization signals. GFP (top) and DIC (bottom) are representative images of at least twenty fluorescent cells.

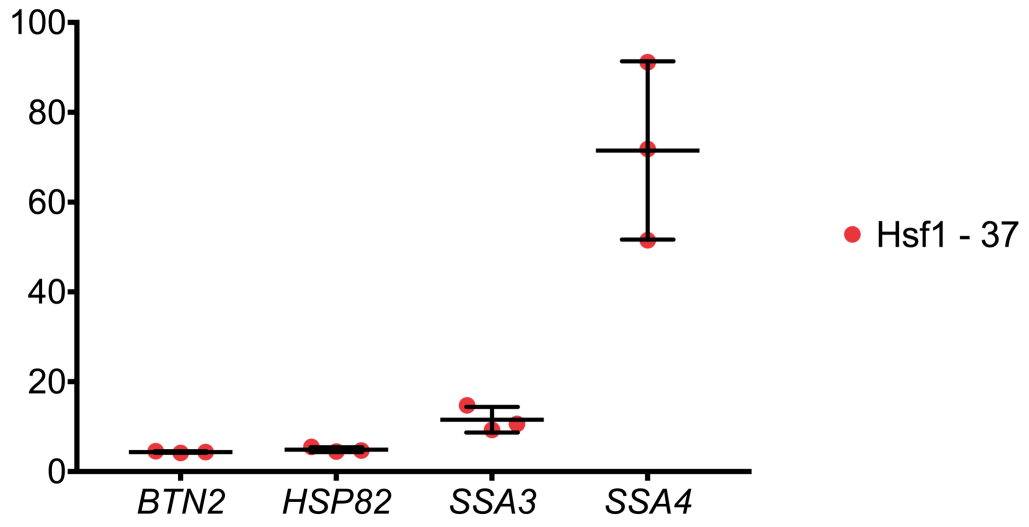

Fig. S4. qRT-PCR of Hsf1-target genes during heat shock. Quantitative RT-PCR of the Hsf1 target genes *BTN2*, *HSP82*, *SSA3* and *SSA4* is shown as fold increase in gene expression relative to the wild type strain after 15 min heat shock at 37˚ C. Results from three independent experiments are shown.

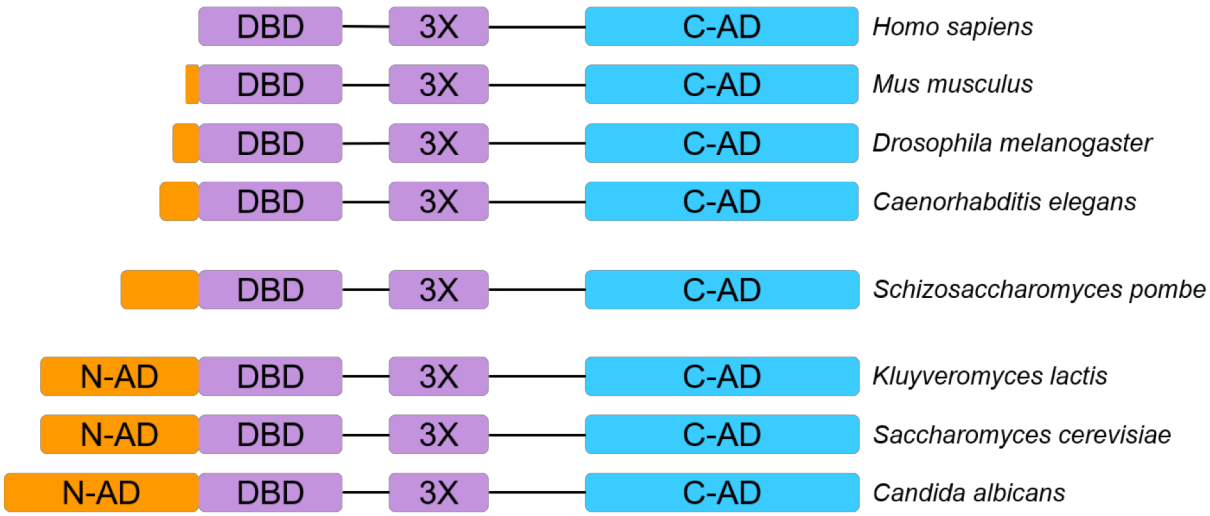

Fig. S5. HSF alignments from various eukaryotic species. Coding sequences for HSF1 proteins from the indicated species were obtained from GenBank and aligned at the level of domain architecture, with special attention paid to the presence of sequence amino-terminal to the highly conserved DNA-binding domain.