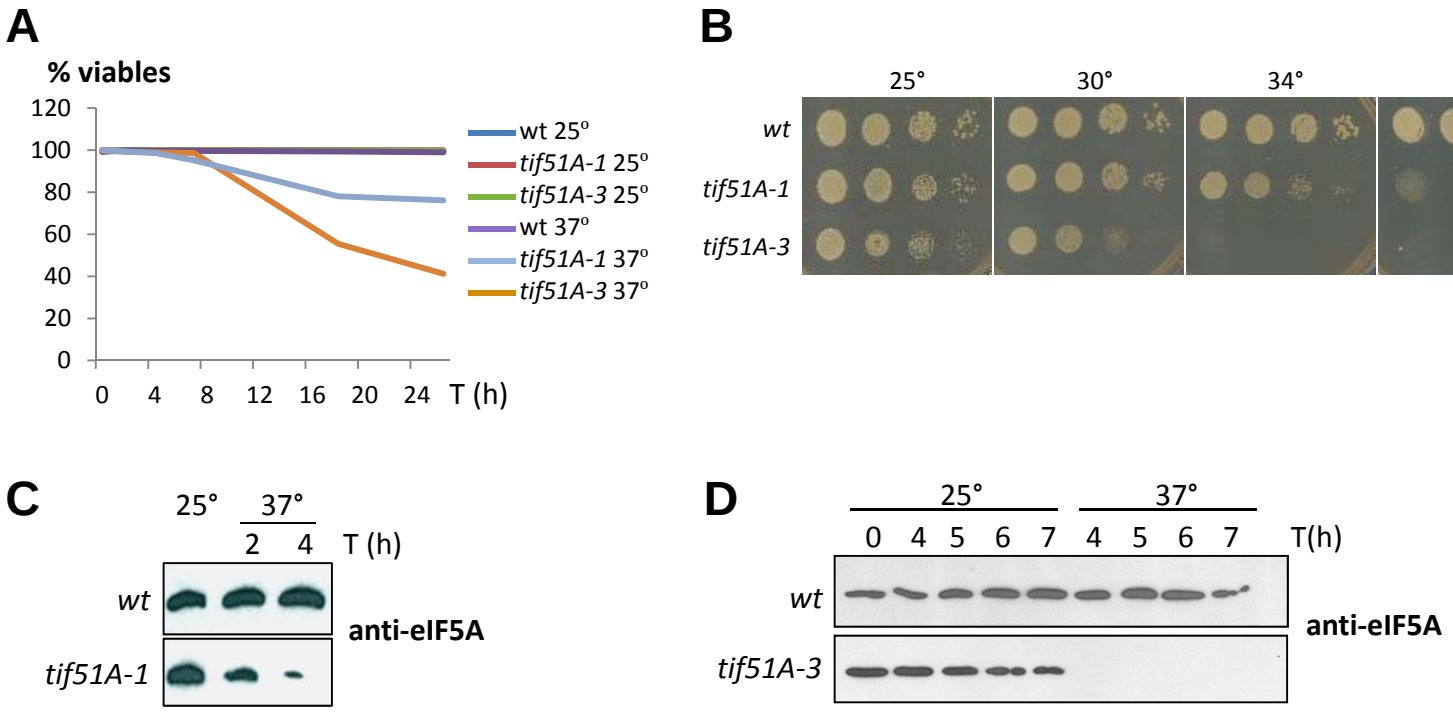

Figure S1 Depletion of the Tif51A protein results in lethality. (A) Percentage of viable cells in wild-type, *tif51A-1* and *tif51A-3* grown in synthetic media at 25° or after incubation at non-permissive (37°) at the times indicated. 3000 cells were counted for each sample. Cell viability was measured at the indicated times using the Muse Count & Viability Assay Kit and the Muse™ Cell Analyser (Millipore, Billerica, MA, USA) according to manufactures' instructions. (B) 10-fold serial dilutions of wild-type, *tif51A-1* and *tif51A-3* were plated onto YPD medium and incubated at the indicated temperatures. (C, D) Immunoblots showing eIF5A protein depletion in *tif51A-1* (C) and *tif51A-3* (D) after incubation at 37° for the indicated times. eIF5A protein expression was also visualized in wild-type under the same conditions.

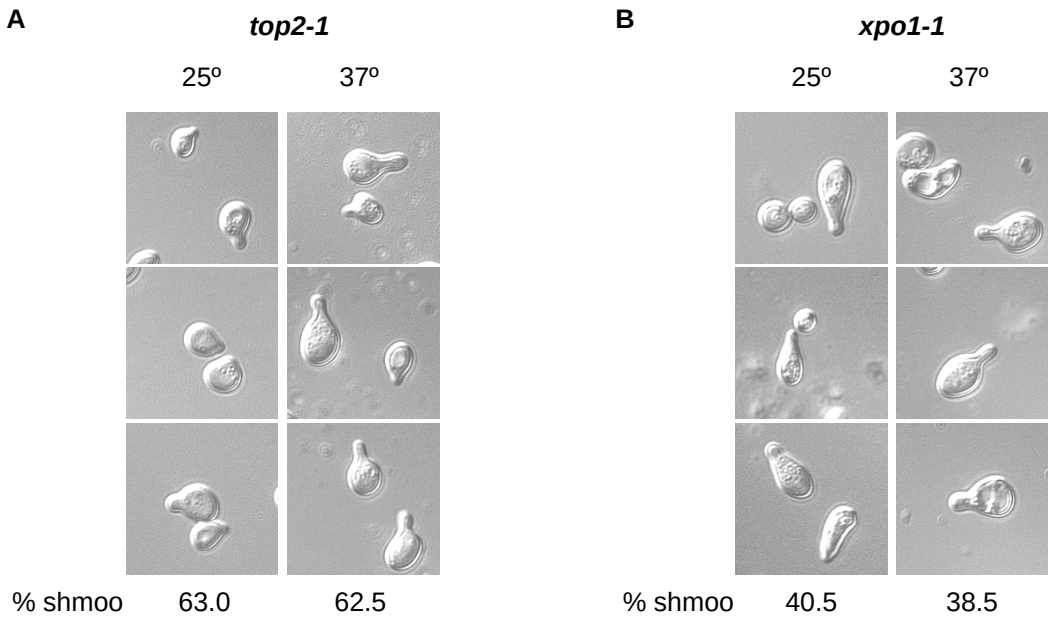

Figure S2 Yeast shmoo formation in temperature-sensitive mutants. (A, B) Representative DIC images of mutants containing temperature sensitive alleles in the essential proteins topoisomerase II (*top2-1*) (A) and karyopherine Crm1/Xpo1 (*xpo1-1*) (B). Mutants were grown at 25° until exponential phase and were maintained at 25° or transferred to 37° for 4h. Cells were then treated with 10 µg/ml α-factor for 2h. Percentage of cells containing shmoos in samples treated with α-factor are indicated. Approximately 300 cells were manually counted for each sample from at least two independent experiments.

Table S1 Yeast strains used in this study.

Strain	Genotype	Source
BY4741	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0*	Euroscarf
lia1Δ	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 lia1Δ::KanR*	Euroscarf
spe2Δ	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 spe2Δ::KanR*	Euroscarf
tif51BΔ	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 tif51BΔ::KanR*	Euroscarf
bnr1Δ	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 bnr1Δ::KanR*	Euroscarf
bni1Δ	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 bni1Δ::KanR*	Euroscarf
tif51A-1	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 tif51A-1::KanMX*	(Li *et al.* 2011)
tif51A-3	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 tif51A-3::KanMX*	(Li *et al.* 2011)
PAY723	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 BNI1-6HA::HIS3*	This study
PAY725	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 tif51A-1::KanMX BNI1-6HA::HIS3*	This study
PAY727	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 bni1Δ1240-1954-6HA::HIS3*	This study
PAY729	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 tif51A-1::KanMX bni1Δ1240-1954-6HA::HIS3*	This study
PAY739	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 bni1Δ1239-1307-6HA::HIS3*	This study
PAY741	MATa *his3Δ0 leu2Δ0 met15Δ0 ura3Δ0 tif51A-1::KanMX bni1Δ1239-1307-6HA::HIS3*	This study
top2-1	MATa *leu2-3,112 trp1-1 can1-100 ura3-1 ade2-1 his3-11,15 top2-1*	(Brill and Sternglanz 1988)
xpo1-1	MATa *leu2-3,112 trp1-1 can1-100 ura3-1 ade2-1 his3-11,15 xpo1::LEU2 pKW457 (xpo1-1 in pRS313)*	(Stade *et al.* 1997)

Table S2 List of plasmids used in this study.

Plasmid	Description	Source
PA171	GA2256 *HA6-HIS3*	Dr. G. Ammerer, University of Vienna, Vienna, Austria
PA283	pRS416 *SPA2-GFP, URA3*, 2μ	Dr. R. Arkowitz, CNRS, Nice, France
PA288	GA1815 *FUS1-LacZ, URA3*	Dr. G. Ammerer, University of Vienna, Vienna, Austria
PA159	Yep368 *STL1-LacZ, LEU2,* 2μ	Dr. E. de Nadal, University Pompeu Fabra, Barcelona, Spain
PA291	PB1028 *GAL-BNI1-4HA, URA3*, 2μ	Dr. D. Pellman (Dana-Farber Cancer Institute and Harvard Medical School, Boston, USA)

Table S3 List of primers used in this study.

Primer	Sequence	Use
BNI1-1	5′-AGATCCATAGGTGAGGCTAGCACAGGTAACAGGCTAAG TTTCAAATCCGGTTCTGCTGCTAG-3′	*BNI1* full length genomic tagging
BNI1-2	5′-GTTTGTTTTGGTATTACTGTTGTCATAATTTTTTGGTTTAA TATTCCTCGAGGCCAGAAGAC-3′	*BNI1* full length, C terminus truncated, and polyPro deletion genomic tagging
BNI1-3	5′-ACATGTGGAAAACGGAAAGC-3′	qPCR
BNI1-4	5′-AGATCTTCTGCGCCATCTGT-3′	qPCR
BNI1-5	5′-GCAGAAGATCTCTCTACTCAATCATCTGTACTCTCCTCAC AGCCGTCCGGTTCTGCTGCTAG-3′	*BNI1* C-truncated genomic taggin
BNI1-6	5′-GGCGCAGAAGATCTCTCTACTCAATCATCTGTACTCTCC TCACAGCTACCATCTGTATTATCT-3′	*BNI1* polyPro deletion genomic tagging
ACT1-1	5′-TCGTTCCAATTTACGCTGGTT-3′	qPCR
ACT1-2	5′-CGGCCAAATCGATTCTCAA-3′	qPCR
HXK2-1	5′-CAATTCCATTGGGTTTCACC-3′	qPCR
HXK2-2	5′-GCAACATTGGAACAACATCG-3′	qPCR

Supplementary literature cited

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