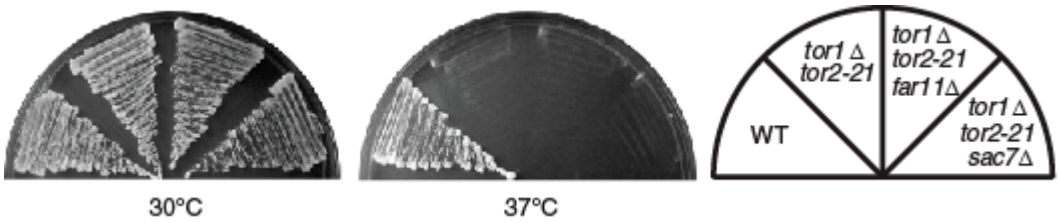

Figure S1 *sac7Δ* and *far11Δ* do not suppress the temperature-sensitive growth phenotype of a *tor1Δ tor2-21* double mutant. Wild-type (SH100), *tor1Δ tor2-21* (SH221), *tor1Δ tor2-21 sac7Δ* (TPY112), and *tor1Δ tor2-21 far11Δ* (TPY118) cells were grown on YPD plates at 30 °C and 37 °C.

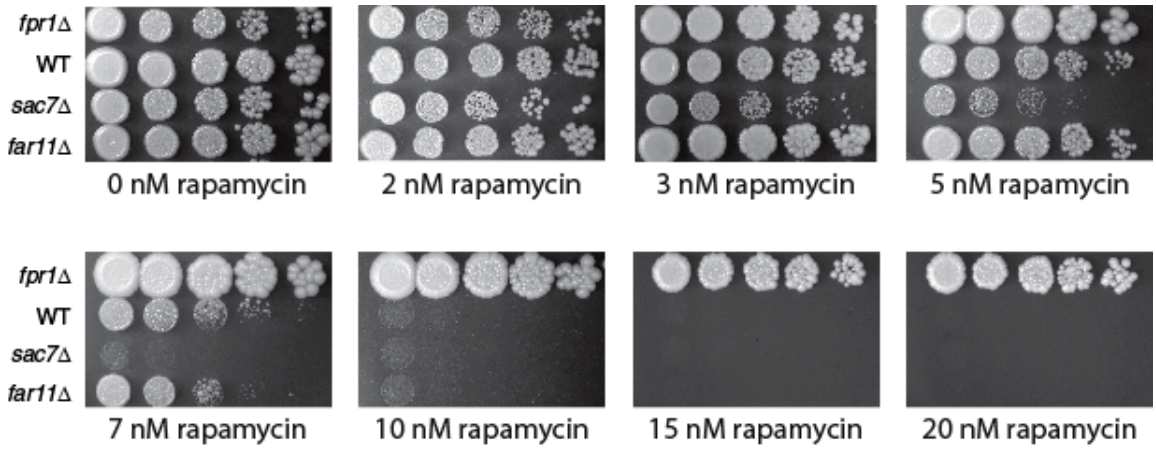

Figure S2 The effect of rapamycin on the growth of *sac7Δ* and *far11Δ* mutant cells. Cultures of wild-type (ZLY423), *fpr1Δ* (TPY122), *sac7Δ* (ZLY2404), and *far11Δ* mutant (TPY114) cells were serially diluted and spotted on YPD plates supplemented with different concentrations of rapamycin.

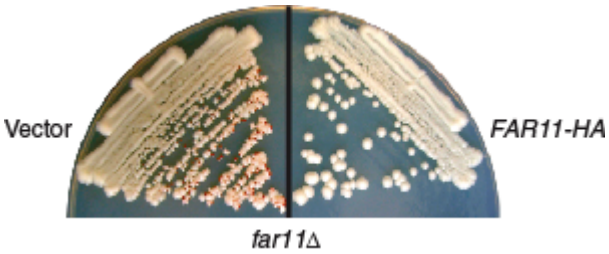

Figure S3 A *FAR11-HA* fusion construct is functional. *far11Δ* mutant cells (*lst8Δ ade2-1 far11Δ*, TPY114) carrying plasmid pRS412-LST8 and either pRS416 empty vector (Vector) or pRS416-FAR11-HA (*FAR11-HA*, pZL2762) were grown on YNBcasD medium supplemented with adenine.

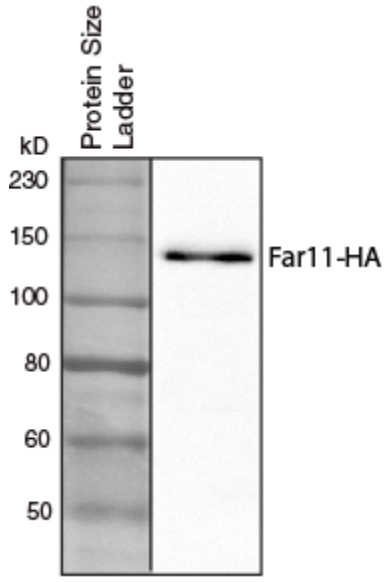

Figure S4 Far11-HA in total cellular proteins prepared by trichloroacetic acid precipitation exists as a single band on Western blots. Total cellular proteins were prepared from the yeast strain SY4078 carrying a centromeric plasmid encoding *FAR11-HA* (pZL2762) using the NaOH-β mercaptoethanol-trichloroacetic acid method as described (YAFFE and SCHATZ 1984) and separated by SDS-PAGE. Far11-HA was detected by immunoblotting with the high affinity rat monoclonal anti-HA antibody 3F10 (Roche).

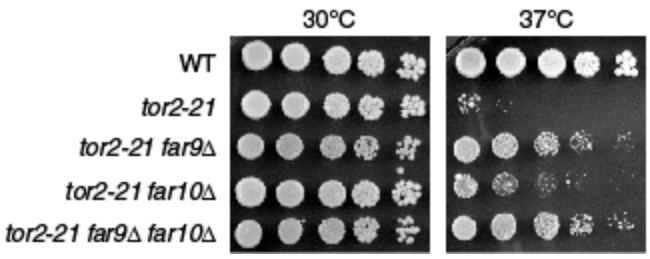

Figure S5 The effect of *far9Δ* and *far10Δ* on suppressing the temperature-sensitive growth phenotype of a *tor2-21* mutant. Wild-type (SH100), *tor2-21* (SH121), *tor2-21 far9Δ* (TPY207), *tor2-21 far10Δ* (TPY264) and *tor2-21 far9Δ far10Δ* mutant (TPY220) cells were serially diluted and spotted on YPD plates and grown at 30 °C and 37 °C.

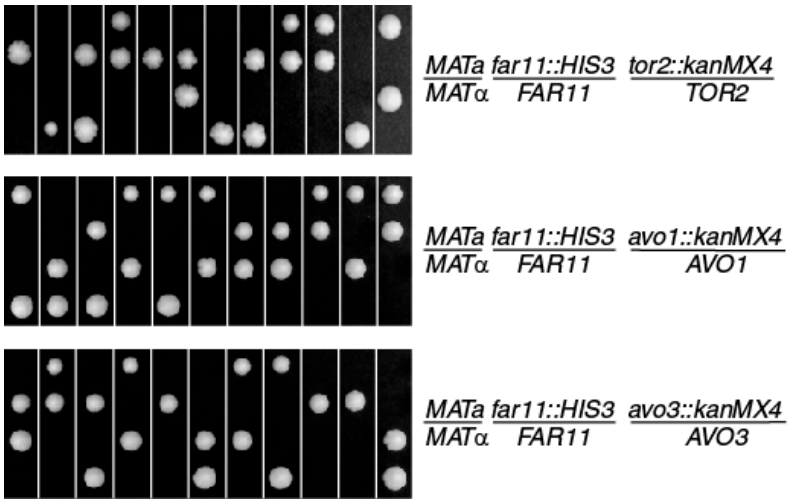

Figure S6 Tetrad analysis of sporulated diploid cells heterozygous for mutations in *FAR11* and *TOR2*, *AVO1* or *AVO3*. None of the colonies were geneticin (G418) resistant, indicating that no viable *tor2Δ*, *avo1Δ*, or *avo3Δ* mutant haploid cells were generated.

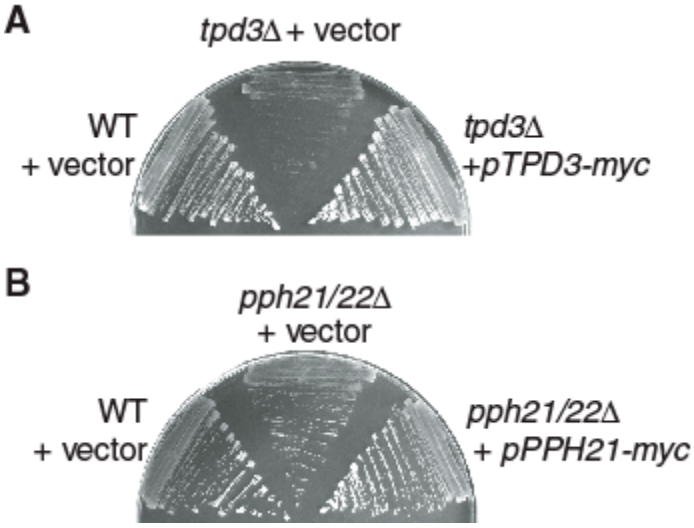

Figure S7 (A) Tpd3-myc is functional. Wild type (BY4741) and isogenic *tpd3Δ* mutant (BY4741 tpd3) cells carrying an empty vector (pRS415) or *TPD3-myc* plasmid (pTP242) as indicated were grown on leucine-dropout medium and the picture was taken after 3 days.
(B) Pph21-myc is functional. Wild type (BY4741) and isogenic *pph21/22Δ* mutant (BY4741 pph21/22) cells carrying an empty vector (pRS415) or *PPH21-myc* plasmid (pTP244) were analyzed for cell growth as described for panel A.

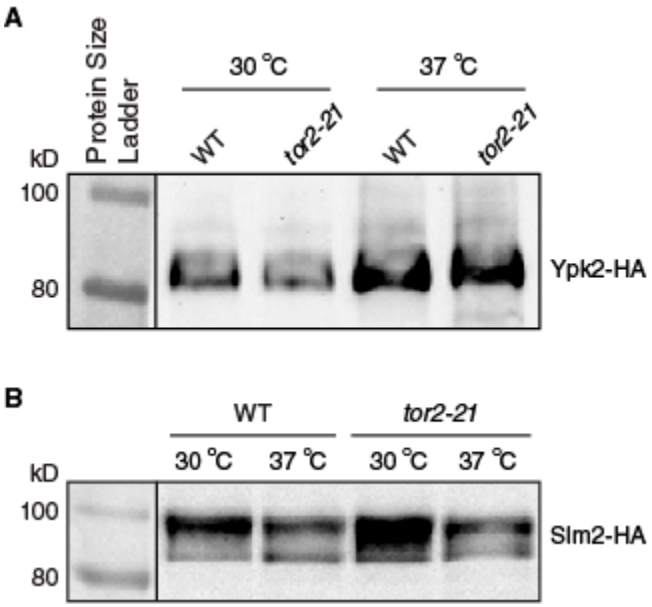

Figure S8 Immunoblot analysis of HA-tagged Ypk2 (panel A) and Slm2 (panel B). Wild-type (WT, SH100) and temperature-sensitive *tor2-21* mutant cells (SH121) expressing C-terminal 3xHA-tagged Ypk2 or Slm2 from a centromeric plasmid (*YPK2-HA*, pTP271; *SLM2-HA*, pTP377) were grown in YNBcasD medium at 30 °C to mid-log phase and switched to 37 °C for 3h before cellular proteins were processed for Western blotting.

Table S1 Quantitative analysis of polarization of the actin cytoskeleton in wild-type and isogenic *tor2-21* (SH121), *tor2-21 sac7Δ* (TPY110), *tor2-21 far11Δ* (TPY311), *tor2-21 sac7Δ far11Δ* (TPY301) mutant cells. Cells were grown in YPD medium at 30 °C to mid-log phase and then switched to 37 °C for 3 h before cells were fixed by formaldehyde and stained with rhodamine phalloidin. The percentage of cells with polarized actin cytoskeleton was tabulated.

Strain	# cells imaged	# polarized cells	% polarized cells
Wild-type (SH100)	474	357	75.3%
tor2-21 (SH121)	424	89	21.0%
tor2-21 sac7Δ (TPY110)	504	267	53.0%
tor2-21 far11Δ (TPY311)	461	240	52.1%
tor2-21 sac7Δ far11Δ (TPY301)	451	318	70.5%

Table S2 Strains used.

Strain	Genotype	Source/Reference
TPY114 (*far11*)	*MATa ade2-1 ura3 his3-11,15 leu2 lst8::LEU2 far11::kanMX4 [pRS412-LST8]*	This study
SY4078	SY2227 *FAR7-myc13-KAN <pSL2771>*	(KEMP and SPRAGUE 2003)
SH100 (WT)	*MATa leu2-3,112 ura3-52 rme1 trp1 his4 HMLa ade2 tor2::ADE2 [YCplac111::TOR2]*	(HELLIWELL *et al.* 1998)
SH121 (*tor2-21*)	*MATa leu2-3,112 ura3-52 rme1 trp1 his4 HMLa ade2 tor2::ADE2 [YCplac111::tor2-21]*	
SH221 (*tor1 tor2-21*)	*MATa leu2-3,112 ura3-52 rme1 trp1 his3 HMLa ade2 tor1::HIS3 tor2::ADE2 [YCplac111::tor2-21]*	
TPY110 (*tor2-21 sac7*)	SH121 *sac7::kanMX4*	This study
TPY311 (*tor2-21 far11*)	SH121 *far11::TRP1*	This study
TPY301 (*tor2-21 sac7 far11*)	SH121 *sac7::kanMX4 far11::TRP1*	This study
TPY112	SH221 *sac7::kanMX4*	This study
TPY118	SH221 *far11::kanMX4*	This study
TPY207 (*tor2-21 far9*)	SH121 *far9::kanMX4*	This study
TPY264 (*tor2-21 far10*)	SH121 *far10::URA3*	This study
TPY220 (*tor2-21 far9 far10*)	SH121 far9::kanMX4 *far10::URA3*	This study
ZLY423 (WT)	*MATa ade2-1 ura3 his3-11,15 leu2 lst8::LEU2 [pRS412-LST8]*	This study.
TPY122 (*fpr1*)	*MATa ade2-1 ura3 his3-11,15 leu2 lst8::LEU2 fpr1::kanMX4 [pRS412-LST8]*	This study
ZLY2404 (*sac7*)	*MATa ade2-1 ura3 his3-11,15 leu2 lst8::LEU2 sac7::kanMX4 [pRS412-LST8]*	This study
TPY114 (*far11*)	*MATa ade2-1 ura3 his3-11,15 leu2 lst8::LEU2 far11::kanMX4 [pRS412-LST8]*	This study
BY4741 (*pph21/22*)	*MATa ura3 leu2 his3 met15 pph21::kanMX4 pph22::kanMX4*	This study
BY4741	*MATa ura3 leu2 his3 met15*	Yeast genome
BY4741 *tpd3*	BY4741 *tpd3::kanMX4*	deletion project
BY4743	*MATa/MATalpha ura3/ura3 leu2/leu2 his3/his3 lys2/LYS2 met15/MET15*	
BY4743 *tor2/TOR2*	BY4743 *tor2::kanMX4/TOR2*	
BY4743 *avo1/AVO1*	BY4743 *avo1::kanMX4/AVO1*	
BY4743 *avo3/AVO3*	BY4743 *avo3::kanMX4/AVO3*	

TPY654	BY4743 *tor2::kanMX4/TOR2 far11::HIS3/FAR11*	This study
TPY652	BY4743 *avo1::kanMX4/AVO1 far11::HIS3/FAR11*	This study
TPY653	BY4743 *avo3::kanMX4/AVO3 far11::HIS3/FAR11*	This study

Table S3 Plasmids used.

Plasmid	Description	Source/Reference
pZL2762	pRS416-FAR11-HA, expressing Far11 from its own promoter with a 3xHA tag at the C-terminus.	This study
pTP377	pRS416-SLM2-HA, expressing Slm2 from its own promoter with a 3xHA tag at the C-terminus.	This study
pTP271	pRS416-YPK2-HA, expressing Ypk2 from its own promoter with a 3xHA tag at the C-terminus.	This study
pZL1255	pRS412-LST8	This study
pTP242	pRS415-ADH1-TPD3-myc, expressing Tpd3 from the *ADH1* promoter with a 3xmyc tag at the C-terminus.	This study
pTP244	pRS415-PPH21-myc, expressing Pph21 from its own promoter with a 3xmyc tag at the C-terminus.	This study

Supplemental References

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