

Table S1

Strains used in this study

Strain	Genotype	Source
SN152	<i>arg4Δ/arg4Δ leu2Δ/leu2 Δ his1Δ/his1Δ ura3Δ::imm⁴³⁴/URA3 iro1Δ::imm⁴³⁴/IRO1</i>	(1)
DK318 (WT)	<i>arg4Δ::ARG4 /arg4Δ his1Δ::HIS1/his1Δ ura3Δ::imm⁴³⁴/URA3 iro1Δ::imm⁴³⁴/IRO1</i>	(2)
MAY7 (<i>ppg1Δ/+</i>)	<i>ppg1Δ::C.m.HIS1/PPG1 arg4Δ::ARG4/arg4Δ leu2Δ::LEU2/leu2Δ his1Δ/his1Δ ura3Δ ::imm⁴³⁴/URA3 iro1Δ ::imm⁴³⁴/IRO1</i>	This study
MAY34 (<i>ppg1Δ/Δ</i>)	<i>ppg1Δ::C.d.HIS1/ppg1Δ::C.m.LEU2/ arg4Δ::ARG4/arg4Δ leu2Δ/leu2Δ his1Δ/his1Δ ura3Δ::imm⁴³⁴/URA3 iro1Δ::imm⁴³⁴/IRO1</i>	This study
MAY50 (<i>ppg1Δ/Δ::PPG1</i>)	<i>ppg1Δ::PPG1::SAT1/ppg1Δ::C.d.HIS1 arg4Δ::ARG4/arg4Δ leu2Δ/leu2Δ his1Δ/his1Δ ura3Δ::imm⁴³⁴/URA3 iro1Δ::imm⁴³⁴/IRO1</i>	This study
MAY55 (<i>ppg1Δ/Δ::ppg1_{H248A H173A D90L}</i>)	<i>ppg1Δ::PPG1_{H248A H173A D90L} ::SAT1/ppg1Δ::C.d.HIS1 arg4Δ::ARG4/arg4Δ leu2Δ/leu2Δ his1Δ/his1Δ ura3Δ::imm⁴³⁴/URA3 iro1Δ::imm⁴³⁴/IRO1</i>	This study
MAY60 (<i>tpk1Δ/Δ</i>)	<i>tpk1Δ::frt/tpk1Δ::SAT1 arg4Δ::ARG4 /arg4Δ his1Δ::HIS1/his1Δ ura3Δ::imm⁴³⁴/URA3 iro1Δ::imm⁴³⁴/IRO1</i>	This study
HLC52 (<i>efg1Δ/Δ</i>)	<i>efg1Δ::hisG/efg1Δ::hisG-URA3-hisG ura3Δ::imm⁴³⁴/ura3Δ::imm⁴³⁴</i>	(3)
MAY65 (<i>ppg1Δ/Δ tpk1Δ/Δ</i>)	<i>ppg1Δ::C.d.HIS1/ppg1Δ::C.m.LEU2 tpk1Δ::frt/tpk1Δ::SAT1 arg4Δ::ARG4/arg4Δ leu2Δ/leu2Δ his1Δ/his1Δ ura3Δ::imm⁴³⁴/URA3 iro1Δ::imm⁴³⁴/IRO1</i>	This study
MAY70 (<i>ppg1Δ/Δ efg1Δ/Δ</i>)	<i>ppg1Δ::C.d.HIS1/ppg1Δ::C.m.LEU2 efg1Δ::frt/efg1Δ::SAT1 arg4Δ::ARG4/arg4Δ leu2Δ/leu2Δ his1Δ/his1Δ ura3Δ::imm⁴³⁴/URA3 iro1Δ::imm⁴³⁴/IRO1</i>	This study
MAY73 (<i>tetO-PPG1</i>)	<i>tetR tetO-PPG1::FLP-SAT1/PPG1 arg4Δ::ARG4 /arg4Δ his1Δ::HIS1/his1Δ ura3Δ::imm⁴³⁴/URA3 iro1Δ::imm⁴³⁴/IRO1</i>	This study

C.d. = *dublinsiensis*; C.m. = *C. maltosa*

References

1. **Noble SM, Johnson AD.** 2005. Strains and strategies for large-scale gene deletion studies of the diploid human fungal pathogen *Candida albicans*. *Eukaryot Cell* **4**:298-309.
2. **Banerjee M, Thompson DS, Lazzell A, Carlisle PL, Pierce C, Monteagudo C, Lopez-Ribot JL, Kadosh D.** 2008. *UME6*, a novel filament-specific regulator of *Candida albicans* hyphal extension and virulence *Molecular biology of the cell* **19**:1354-1365.
3. **Lo HJ, Kohler JR, DiDomenico B, Loebenberg D, Cacciapuoti A, Fink GR.** 1997. Nonfilamentous *C. albicans* mutants are avirulent. *Cell* **90**:939-949.

Table S2**Primers used in this study**

#	Primer Name	Sequence	Description
1	MAO5	GTATGCGCCCTAACTACCT	<i>PPG1</i> 5' flank upstream for fusion PCR
2	MAO6	cacggcgcgctagcagcggCCCACATATAATGAACGA CAGC	<i>PPG1</i> 5' flank downstream for fusion PCR
3	MAO7	gtcagcggccgcatccctgcGAACCCATCATGTATTTC	<i>PPG1</i> 3' flank upstream for fusion PCR
4	MAO8	GAAGGGTTTTCAATTTGC	<i>PPG1</i> 3' flank downstream for fusion PCR
5	RZO37	ccgetgctagcgcgcccgtgACCAGTGTGATGGAT ATCTGC	5' vector primer for <i>LEU2</i> , <i>ARG4</i> , <i>HIS1</i> markers (designed by R. Zordan)
6	RZO38	gcagggatcggcccgtgacAGCTCGGATCCACTA GTAACG	3' vector primer for <i>LEU2</i> , <i>ARG4</i> , <i>HIS1</i> markers (designed by R. Zordan)
7	MAO155	ATCACTAAAGCTCGACCAGTT	5' <i>PPG1</i> flank upstream primer for split marker add-back of <i>PPG1</i>
8	MAO156	ACTTCTTCTTTCTTCAGCTCTCTT	5' <i>PPG1</i> flank upstream nested primer for split marker add-back of <i>PPG1</i>
9	MAO157	ggtaccGTCTTGAAACTGATACAATTGGAGA	5' <i>PPG1</i> flank downstream primer for split marker add-back of <i>PPG1</i>
10	MAO158	ccgcggCACACACGTACACACACATTC	3' <i>PPG1</i> flank upstream primer for split marker add-back of <i>PPG1</i>
11	MAO159	GTAAAGCTAAGAGTAACTCGGT	3' <i>PPG1</i> flank downstream primer for split marker add-back of <i>PPG1</i>
12	MAO160	GGCGTATATGATGATTTAAGTTGGG	3' <i>PPG1</i> flank downstream nested primer for split marker add-back of <i>PPG1</i>
13	MAO77	aggcgcgccatggccATGACGGTGCCGTTCAAATAC C	Upstream non-mutagenic primer to clone <i>PPG1</i> into pAG10H vector using NcoI restriction enzyme. Also used as a non-mutagenic primer to generate D90L mutation.
14	MAO78	gcagccggatccTTACAAGAAATACTCAACTTGTT GAC	Downstream non-mutagenic primer to clone <i>PPG1</i> into pAG10H vector using BamHI restriction enzyme. Also used as a non-mutagenic primer to generate H248A mutation.
15	MAO133	CACTGAATAATAACCACGATCAACGTACA <u>AAA</u> CCCAAGAATAAATAATTAGTTTGAGGCG	<i>PPG1</i> downstream internal mutagenic primer to

16	MAO150	CGTGGTTATTATTCAGTGGAACTATATC	generate D90L mutation <i>PPG1</i> upstream internal non-mutagenic primer to generate H173A mutation
17	MAO151	CCAATTGTTGACAACCTGGGGGATAACCCACC <u>GGCACATGCAAATATCTTGCCATCTATAG</u>	<i>PPG1</i> downstream internal mutagenic primer to generate H173A mutation
18	MAO152	CCAGTTGTCAACAATTGGATCAAATACGAGC AGTCG	<i>PPG1</i> upstream internal non-mutagenic primer to generate H248A mutation
19	MAO153	CTCGTATACCCTTCATTACACAATTGGGCAGC TCTAATCATTTGCACC	<i>PPG1</i> downstream internal mutagenic primer to generate H248A mutation
20	MAO154	GGTGCAAATGATTAGAGCTGCCCAATTGTGT AATGAAGGGTATACGAG	<i>PPG1</i> upstream internal mutagenic primer to generate H248A mutation
21	MAO138	GAGGTAGTTGGTGTGGAAAGA	5' <i>TPK1</i> flank upstream primer for split marker deletion of <i>TPK1</i>
22	MAO148	ACTGATGTCAATTTGTTGGCG	5' <i>TPK1</i> flank upstream nested primer for split marker deletion of <i>TPK1</i>
23	MAO139	ggtaccGAAAGGAGAAGGAGGAGGATAAC	5' <i>TPK1</i> flank downstream primer for split marker deletion of <i>TPK1</i>
24	MAO140	ccgcggCACTATATGAATTGGCTTCCTTGTT	3' <i>TPK1</i> flank upstream primer for split marker deletion of <i>TPK1</i>
25	MAO141	CTCAACTATTGTCTGAATGTGTGTATC	3' <i>TPK1</i> flank downstream primer for split marker deletion of <i>TPK1</i>
26	MAO149	GGAAAGACCTCTAATTCGAT	3' <i>TPK1</i> flank downstream nested primer for split marker deletion of <i>TPK1</i>
27	MAO126	gatcataccgaacagaggtaccTACCTGGCCCTGGCAAA C	5' primer for -98 to -566 region of <i>PPG1</i> promoter used for cloning into pEL9
28	MAO128	gatcataccgaacagaccgcggGTGGGTACACATTCAAC AGA	3' primer for -98 to -566 region of <i>PPG1</i> promoter used for cloning into pEL9
29	MAO127	gatcataccgaacagaggtaccATGGATTGGGTAGTGGG TGAT	5' primer for -33 to +446 region of <i>PPG1</i> used for cloning into pEL9
30	MAO129	gatcataccgaacagaccgcggGGCTGACCCTTGATATT TGTTTAAG	3' primer for -33 to +446 region of <i>PPG1</i> used for cloning into pEL9
31	DKO400	GACATATTGACCGACATAAT	5' WT <i>ARG4</i> add-back primer
32	DKO401	CTAATGACTGAATTTGATGTA	3' WT <i>ARG4</i> add-back primer

33	DKO404	GTCGACTATGTCATTCAAGC	5' WT <i>LEU2</i> add-back primer
34	DKO405	CAGTTGTGTGCATATTTAGA	3' WT <i>LEU2</i> add-back primer
35	GSO127	ATGGACGGTGGTATGTTTTAG	Split marker upstream primer for <i>SAT</i> flipper
36	GSO128	TGTTGAATCAATTGCCTTGC	
37	GSO129	ATAGGATCCGAAGTTCCTATACTTTCTAGAG	Split marker downstream primer for <i>SAT</i> flipper
38	GSO130	GTCAGATCTGAAGTTCCTATTCTC	Forward primer for full length <i>SAT</i> flipper
39	DKO523	GTTGACCGAAGCTCCAATGAATCC	Reverse primer for full length <i>SAT</i> flipper
40	DKO526	CAGCAATACCTGGGAACATGG	5' primer for <i>ACT1</i> probe
41	DKO526	CAGCAATACCTGGGAACATGG	3' primer for <i>ACT1</i> probe
42	DKO526	CAGCAATACCTGGGAACATGG	5' primer for <i>ACT1</i> probe
43	DKO526	CAGCAATACCTGGGAACATGG	3' primer for <i>ACT1</i> probe
44	DKO526	CAGCAATACCTGGGAACATGG	5' primer for <i>ECE1</i> probe
45	DKO526	CAGCAATACCTGGGAACATGG	3' primer for <i>ECE1</i> probe
46	DKO526	CAGCAATACCTGGGAACATGG	5' primer for <i>HWPI</i> probe
47	DKO526	CAGCAATACCTGGGAACATGG	3' primer for <i>HWPI</i> probe
48	DKO526	CAGCAATACCTGGGAACATGG	5' primer for <i>NRG1</i> probe
49	DKO526	CAGCAATACCTGGGAACATGG	3' primer for <i>NRG1</i> probe
50	DKO526	CAGCAATACCTGGGAACATGG	5' primer for <i>ALS3</i> probe
51	DKO526	CAGCAATACCTGGGAACATGG	3' primer for <i>ALS3</i> probe

Note: Lowercase bases indicate flanking sequences which contain restriction sites for cloning.

*In order to generate a *ALS3*-specific probe, a 1017 bp fragment was generated using these primers which was cleaved with *HincII* to yield a 758 bp product, as previously described¹.

Note: Underlined bases were mutated.

Reference

1. Hoyer, L.L., Payne, T.L., Bell, M., Myers, A.M. and S. Scherer. 1998. *Candida albicans ALS3* and insights into the nature of the *ALS* gene family. *Curr Genet* 33:451-459.

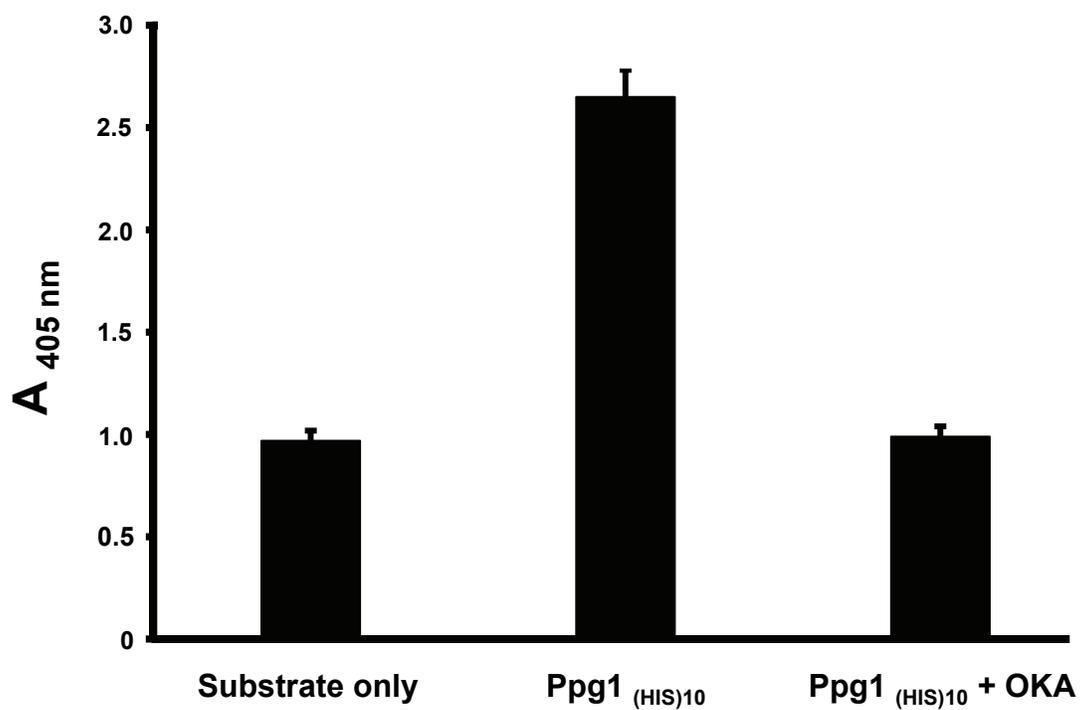


FIG S1. Ppg1 phosphatase activity is strongly inhibited by okadaic acid. Phosphatase assays were performed in the presence and absence of 1 μ M okadaic acid (OKA) using the Ppg1_{(HIS)10} recombinant protein.

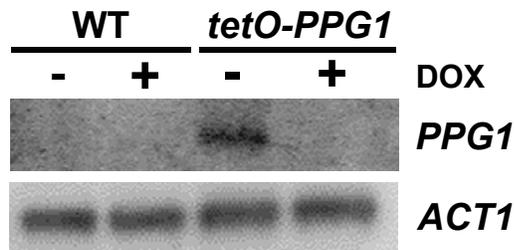
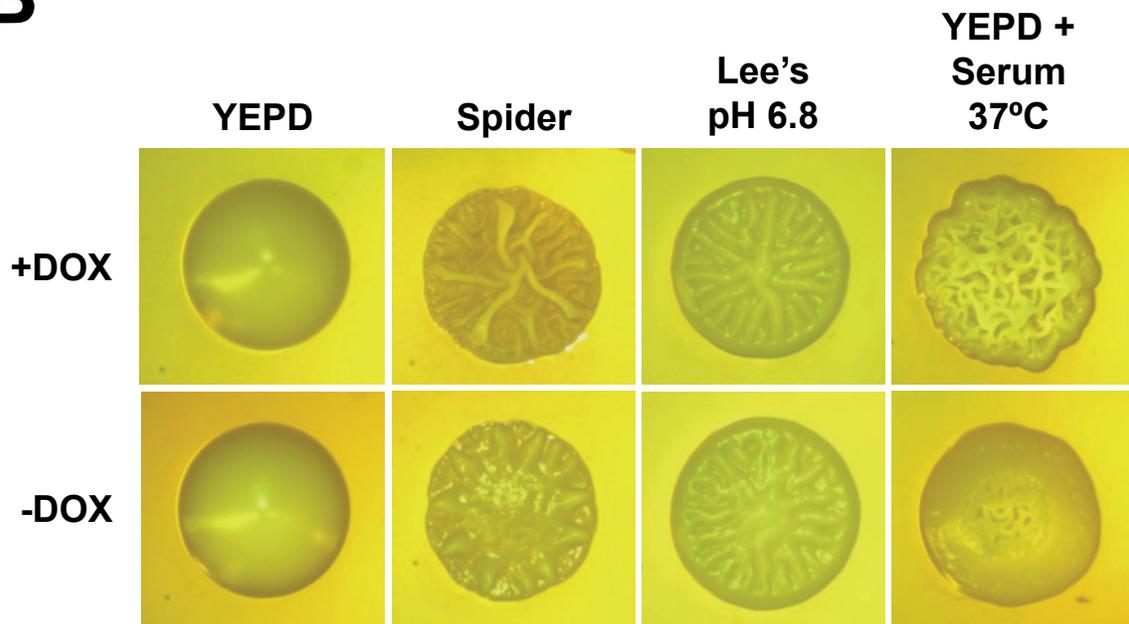
A**B**

FIG S2. High-level constitutive *PPG1* expression affects colony morphology under solid filament-inducing conditions. (A) The indicated strains were grown in YEPD at 30°C overnight in the presence and absence of 20 µg/mL Dox. Cells were harvested for RNA preparation and Northern analysis. 3 µg of total RNA was loaded in each lane and the Northern blot was probed with *PPG1*. *ACT1* is shown as a loading control. (B) Colonies of the *tetO-PPG1* strain were grown on the indicated solid media at 30°C (or 37°C for YEPD + serum) for 2 days in the presence and absence of 20 µg/mL Dox and visualized using light microscopy.

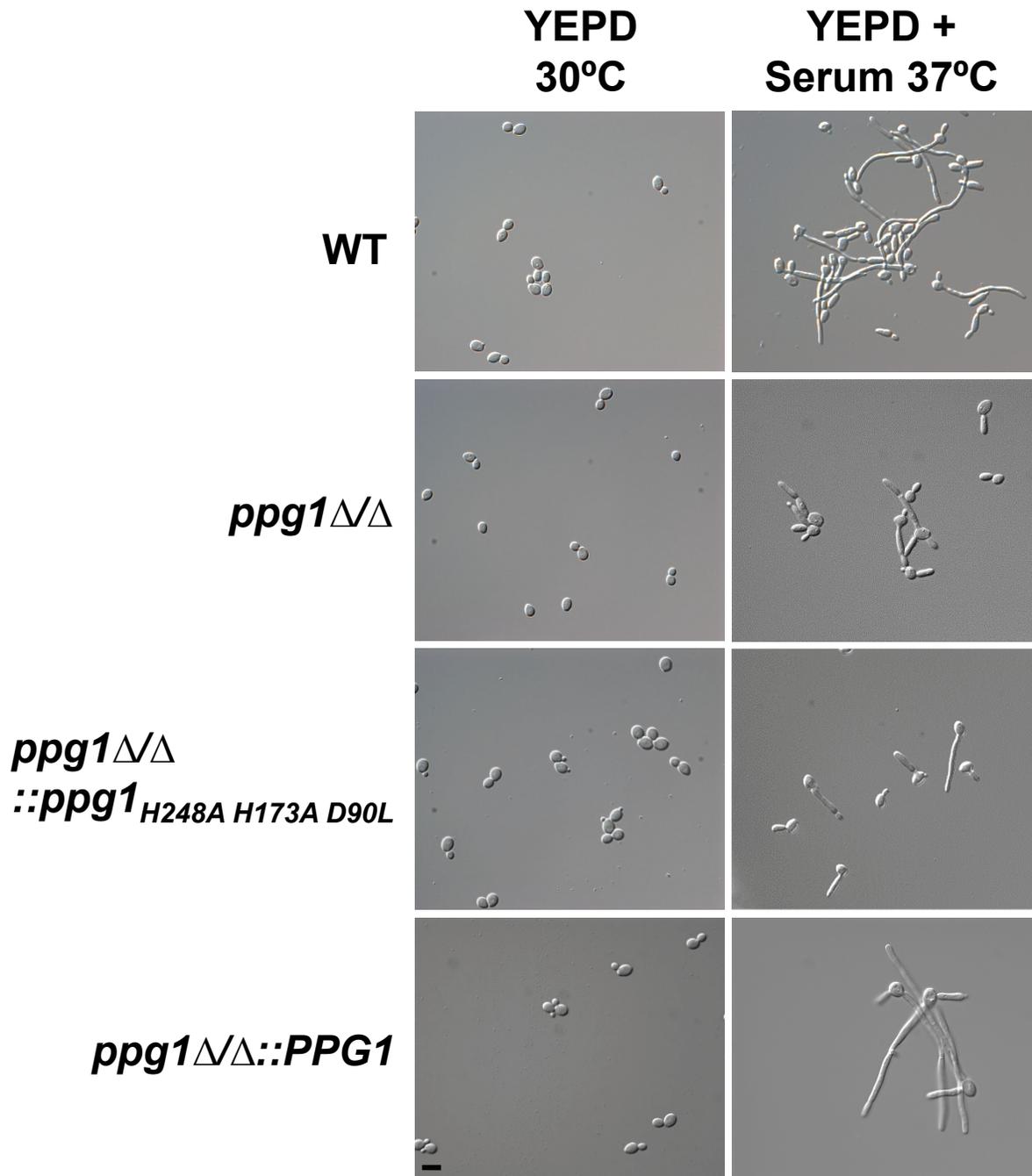


FIG S3. The *ppg1*_{H248A H173A D90L} catalytic mutant shows an equivalent filament extension defect to that of the *ppg1*Δ/Δ strain. The indicated strains were grown in YEPD at 30°C overnight, diluted 1:10 into the indicated pre-warmed media and harvested at the 3 h. time point. Cells were fixed and prepared for DIC microscopy as described in Figure 3. Bar =10 μm.

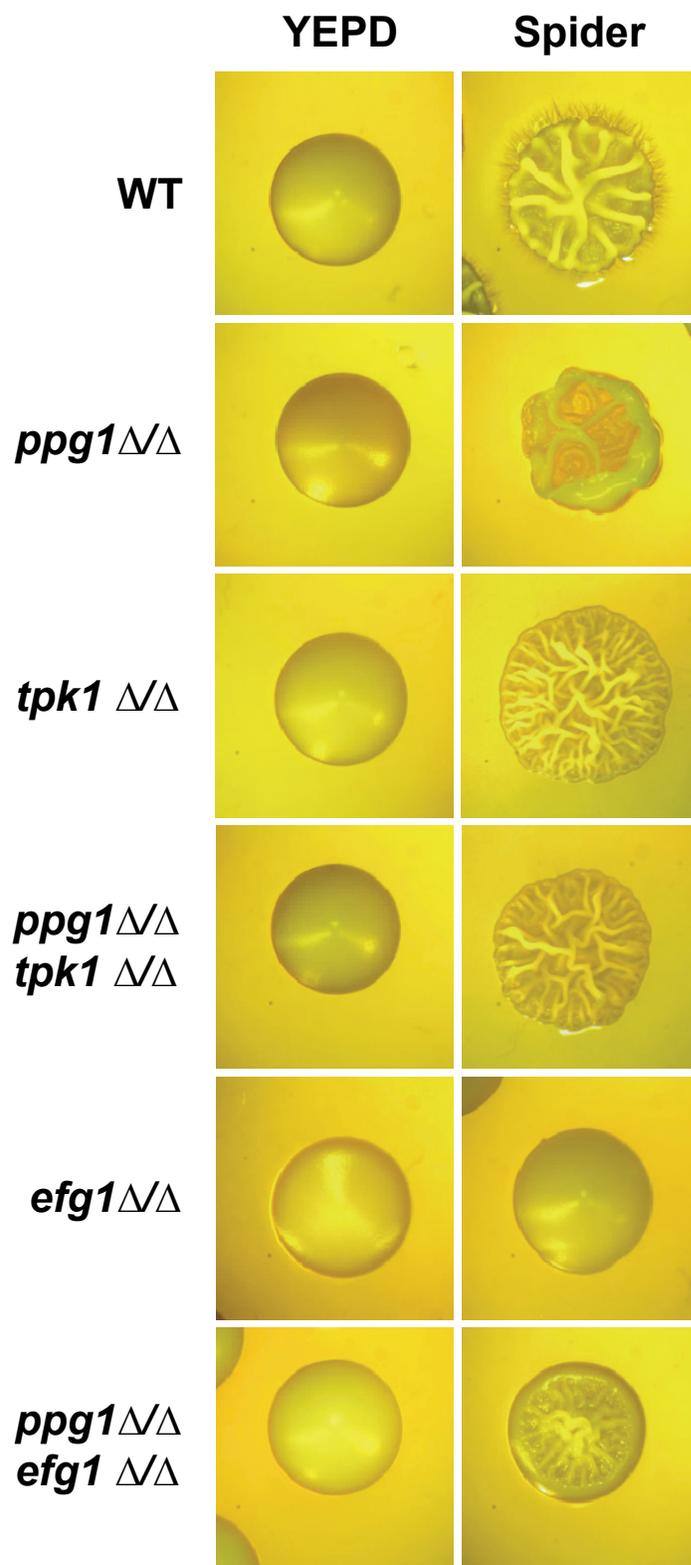


FIG S4. The *tpk1*Δ/Δ mutation is epistatic to the *ppg1*Δ/Δ mutation under solid filament-inducing conditions. The indicated strains were grown for three days on solid medium under non-filament-inducing (YEPD at 30°C) and filament-inducing (Spider at 30°C) conditions. Colonies were visualized by light microscopy.