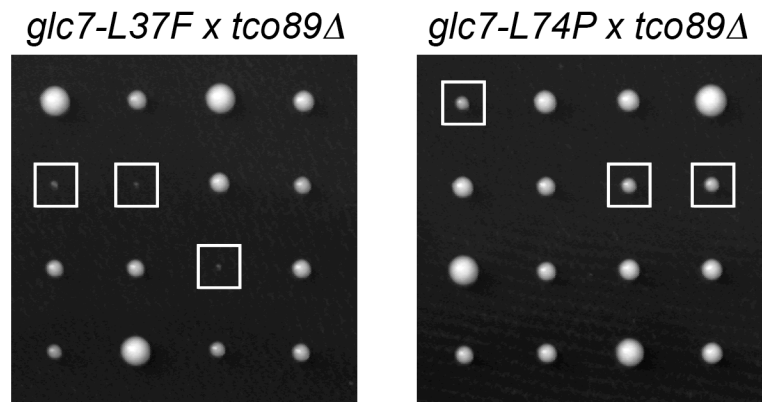
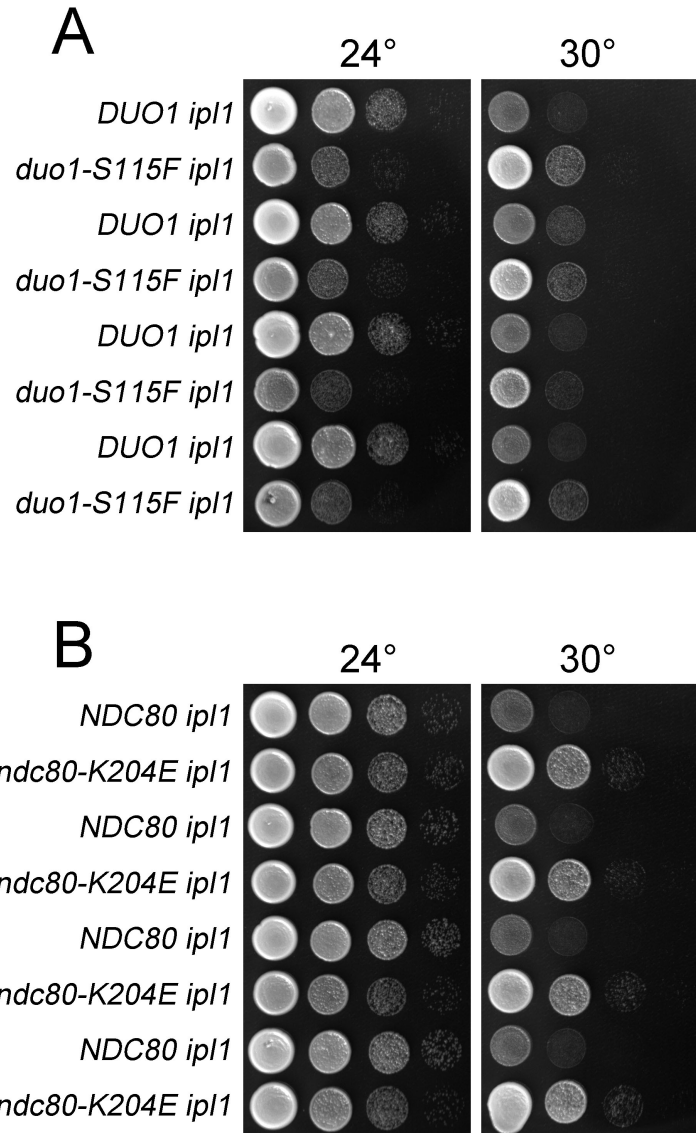


**Figure S1** Dominance of *ipi1* suppressors. Cultures of diploid strains homozygous for *ipi1-2* and heterozygous for the designated *ipi1-2* suppressor mutation were serially diluted onto YPD medium and imaged after 40 hr at the designated temperatures. The WT strain is heterozygous for *ipi1-2* (KT1112 X KT1829). Suppressor mutations in *GLC7*, *SDS22*, *YPI1* and *SHP1* are presented in panels (A), (B), (C), and (D), respectively. Note that only the *GLC7* mutant alleles display semi-dominance.

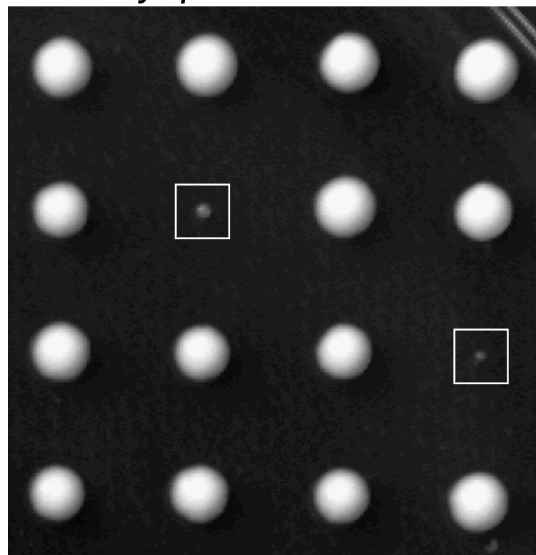


**Figure S2** Genetic interactions between *GLC7* mutant alleles and *TCO89*. Images of four tetrads from a cross between *glc7-L37F* (left panel) and *glc7-L74P* (right panel) and a *tco89Δ::kanMX* strain. The boxes identify the *glc7 tco89Δ* double mutants. Each column represents the four spore clones of a tetrad. Note that the rapamycin-sensitive *GLC7* mutant (*glc7-L37F*) exhibits the strongest genetic interaction with *tco89Δ*.



**Figure S3** Confirmation of *duo1-S115F* and *ndc80-K204E* alleles. Cultures of ascospore clones were serially diluted onto YPD medium and imaged after 28-30 hr at the designated temperatures. (A) Haploid spore clones from a diploid strain KT3349 (*ipl1-2/IPL1 duo1Δ::kan/DUO1* pRS303:*duo1-S115F*). All the clones are *ipl1-2* and contain either wild-type *DUO1* or pRS303:*duo1-S115F*. As expected the pRS303-*duo1-S115F* clones are also cold sensitive, arresting as large budded cells at 14°. (B) Haploid spore clones from a diploid strain KT3350 (*ipl1-2/IPL1 ndc80Δ::kan/NDC80* pRS316:*ndc80-K204E*). All the clones are *ipl1-2* and contain either wild-type *NDC80* or pRS316:*ndc80-K204E*.

*ybp2Δ x mad1Δ*



**Figure S4** Genetic interactions between *ybp2Δ* and *mad1Δ* mutant alleles. Images of four tetrads from a cross between *ybp2Δ::kanMX* and a *mad1Δ::HIS3* strain. The boxes identify the *ybp2Δ mad1Δ* double mutants. Each column represents the four spore clones of a tetrad.

**Table S1 Strain list**

strain	genotype	Source or reference
KT1112	<i>MATa leu2 ura3 his3</i>	(Stuart et al., 1994) <sup>a</sup>
KT1113	<i>MATα leu2 ura3 his3</i>	(Frederick and Tatchell, 1996) <sup>b</sup>
KT1687	<i>MATα mad1::HIS3</i>	this study
KT1688	<i>MATa mad1::HIS3</i>	this study
KT1829	<i>MATα ip11-2 (H352Y)</i>	(Hsu et al., 2000) <sup>c</sup>
KT1963	<i>MATa ip11-2</i>	(Bharucha et al., 2008) <sup>d</sup>
KT2856	<i>MATa SDS22-mCitrine::SpHis5 POM34-mCherry::SpHis5</i>	this study
KT2865	<i>MATa ip11-R150K H352Y</i>	this study
KT2867	<i>MATa ip11-S167L H352Y</i>	this study
KT2869	<i>MATa ip11-G347E H352Y</i>	this study
KT2878	<i>MATa ip11-2 ndc80-K204E ura3::GFP-TUB1::URA3</i>	this study
KT2881	<i>MATa YPI1-13Myc:kanMX6</i>	this study
KT2934	<i>MATα ip11-2 sds22-D2N D119N</i>	this study
KT2936	<i>MATα ip11-2 sds22-77(Cterm frameshift)</i>	this study
KT2938	<i>MATα ip11-2 glc7-L71S</i>	this study
KT2939	<i>MATα ip11-2 glc7-S99L</i>	this study
KT2940	<i>MATα ip11-2 glc7-L37F</i>	this study
KT2963	<i>MATα sds22-D2N D119N</i>	this study
KT2964	<i>MATα sds22-77</i>	this study
KT2961	<i>MATα duo1-S115F (rev76)</i>	this study
KT2967	<i>MATα duo1-S115F (rev81)</i>	this study
KT2969	<i>MATα glc7-L71S</i>	this study
KT2970	<i>MATα glc7-S99L</i>	this study
KT2973	<i>MATα glc7-L37F</i>	this study
KT3052	<i>MATα ip11-2 sds22-E163I L329P</i>	this study

KT3059	<i>MATα ipl1-2 glc7-F118S</i>	this study
KT3062	<i>MATα ipl1-2 glc7-L15S</i>	this study
KT3064	<i>MATα ipl1-2 glc7-K112E</i>	this study
KT3066	<i>MATα ipl1-2 glc7-Q293D</i>	this study
KT3242	<i>MATα GLC7-mCitrine:SpHis5 POM34-mCherry:SpHis5</i>	(Tatchell et al., 2011) <sup>e</sup>
KT3255	<i>MATα ndc80-K204E</i>	this study
KT3257	<i>MATα ipl1-2 ndc80-K204E</i>	this study
KT3292	<i>MATα sds22-E163I L329P</i>	this study
KT3302	<i>MATα glc7-F118S</i>	this study
KT3304	<i>MATα glc7-L15S</i>	this study
KT3308	<i>MATα glc7-K112E</i>	this study
KT3310	<i>MATα glc7-Q293D</i>	this study
KT3317	<i>MATα PDS1-13Myc:kanMX6 SPC42-3XGFP:TRP1 bar1-1 ndc80-K204E</i>	this study
KT3319	<i>MATα PDS1-13Myc:kanMX6 SPC42-3XGFP:TRP1 bar1-1</i>	this study
KT3320	<i>MATα PDS1-13Myc:kanMX6 SPC42-3XGFP:TRP1 bar1-1 ndc80-K204E ipl1-2</i>	this study
KT3351	<i>MATα sds22-F177S</i>	this study
KT3353	<i>MATα ipl1-2 sds22-F177S</i>	this study
KT3355	<i>MATα glc7-Y136N</i>	this study
KT3358	<i>MATα ipl1-2 glc7-Y136N</i>	this study
KT3359	<i>MATα glc7-L74P</i>	this study
KT3361	<i>MATα ipl1-2 glc7-L74P</i>	this study
KT3363	<i>MATα glc7-Y92N R141K</i>	this study
KT3365	<i>MATα ipl1-2 glc7-Y92N R141K</i>	this study
KT3368	<i>MATα ipl1-2 ypi1-F74S</i>	this study
KT3370	<i>MATα ipl1-2 ypi1-F74L</i>	this study
KT3381	<i>MATα ipl1-2 sds22-W187R</i>	this study
KT3383	<i>MATα IPL1-13Myc:kanMX6</i>	this study

KT3385	<i>MAT<math>\alpha</math> duo1-S115F</i>	this study
KT3386	<i>MAT<math>\alpha</math> ipl1-2 duo1-S115F</i>	this study
KT3389	<i>MAT<math>\alpha</math> IPL1-13Myc:kanMX6 tco89-71</i>	this study
KT3391	<i>MAT<math>\alpha</math> IPL1-13Myc:kanMX6 glc7-L74P</i>	this study
KT3392	<i>MAT<math>\alpha</math> IPL1-13Myc:kanMX6 glc7-Y92N R141K</i>	this study
KT3395	<i>MAT<math>\alpha</math> IPL1-13Myc:kanMX6 shp1-105</i>	this study
KT3396	<i>MAT<math>\alpha</math> IPL1-13Myc:kanMX6 shp1-99</i>	this study
KT3400	<i>MAT<math>\alpha</math> ipl1-2 ybp2<math>\Delta</math>::kanMX6</i>	this study
KT3401	<i>MAT<math>\alpha</math> ipl1-2 ybp2<math>\Delta</math>::kanMX6</i>	this study
KT3403	<i>MAT<math>\alpha</math> ybp2<math>\Delta</math>::kanMX6</i>	this study
KT3409	<i>MAT<math>\alpha</math> ipl1-2 duo1-S115F ybp2<math>\Delta</math>::kanMX6</i>	this study
KT3410	<i>MAT<math>\alpha</math> duo1-S115F ybp2<math>\Delta</math>::kanMX6</i>	this study
KT3412	<i>MAT<math>\alpha</math> shp1-99</i>	this study
KT3413	<i>MAT<math>\alpha</math> shp1-99 ipl1-2</i>	this study
KT3415	<i>MAT<math>\alpha</math> shp1-99 ipl1-2</i>	this study
KT3416	<i>MAT<math>\alpha</math> shp1-105</i>	this study
KT3417	<i>MAT<math>\alpha</math> shp1-105 ipl1-2</i>	this study
KT3419	<i>MAT<math>\alpha</math> shp1-105 ipl1-2</i>	this study
KT3424	<i>MAT<math>\alpha</math> GLC7-mCitrine:SpHis5 POM34-mCitrine:SpHis5 ipl1-2 shp1-105</i>	this study
KT3247	<i>MAT<math>\alpha</math> YPI1-13Myc:kanMX6 ipl1-2 shp1-105</i>	this study
KT3428	<i>MAT<math>\alpha</math> SDS22-mCitrine:SpHis5 POM34-mCherry:SpHis5 ipl1-2 shp1-105</i>	this study
KT3449	<i>MAT<math>\alpha</math>/MAT<math>\alpha</math> ipl1-2/IPL1 duo1<math>\Delta</math>::kanMX6 pRS306:duo1-S115F</i>	this study
KT3450	<i>MAT<math>\alpha</math>/MAT<math>\alpha</math> ipl1-2/IPL1 ndc80<math>\Delta</math>::kanMX6 pRS313:ndc80-K204E</i>	this study

<sup>a</sup>Stuart, J.S., Frederick, D.L., Varner, C.M., and Tatchell, K. (1994). The mutant type 1 protein phosphatase encoded by *glc7-1* from *Saccharomyces cerevisiae* fails to interact productively with the *GAC1*-encoded regulatory subunit. *Mol Cell Biol* **14**, 896-905.

<sup>b</sup>Frederick, D.L., and Tatchell, K. (1996). The *REG2* gene of *Saccharomyces cerevisiae* encodes a type1 protein phosphatase-binding protein that functions with Reg1p and the Snf1p protein kinase to regulate growth. *Mol Cell Biol* **16**, 2922-2931.

<sup>c</sup> Hsu, J.-Y., Sun, Z.-W., Li, X., Ruben, M., Tatchell, K., Bishop, D.K., Grushcow, J.M., Brame, C.J., Caldwell, J.A., Hunt, D.F., *et al.* (2000). Mitotic phosphorylation of histone H3 is governed by Ipl1/aurora kinase and Glc7p/PP1 phosphatase in budding yeast and nematodes. *Cell* *102*, 279-291.

<sup>d</sup> Bharucha, J.P., Larson, J.R., Gao, L., Daves, L.K., and Tatchell, K. (2008). Ypi1, a Positive Regulator of Nuclear Protein Phosphatase Type 1 Activity in *Saccharomyces cerevisiae*. *Mol Biol Cell* *19*, 1032-1045.

<sup>e</sup> Tatchell, K., Makrantonis, V., Stark, M.J., and Robinson, L.C. (2011). Temperature-sensitive ipl1-2/Aurora B mutation is suppressed by mutations in TOR complex 1 via the Glc7/PP1 phosphatase. *Proc Natl Acad Sci U S A* *108*, 3994-3999.



**Table S2 Primers Used**

<b>PRIMER</b>	<b>SEQUENCE, 5' - 3'</b>
pBR-247F	GATGCAATTTCTATGCGCAC
pBR-559R	CAAGGAATGGTGCATGCAAG
DUO1-F	GATGAGAGGTTTTGGACCTC
DUO1-R	CCTGTTGAAACTAAGGCATG
DUO1-Fa	CCTGCTCGTTGTACATATC
DUO1-Ra	CAACCTGTAGCCGTATTAAG
GLC7-F	CGTAACACAAGTTAACACC
GLC7-R	GAGTGATGATTGCATCTTCC
IPL1-F	GCAATTGCTCAAGTGAATG
IPL1-R	GCAATATACGATGCCGAAAAG
IPL1-SEQ	CGCTAAATCATCCGAATC
NDC80-F	GAATACATTCACAGGAGAGG
NDC80-R	GGTGAATTGAACAAAATGC
NDC80-1644F	CTGTAGGTGGCTCTAATTG
NDC80-Fa	CTGGATCCCAGATGGTTATAACTG
NDC80-Ra	CTTCTAGACCACCGTCTTCTTATTC
SDS22-F	CTCTCTCCTCCCTTTTTCTC
SDS22-R	GCGAACTTTAACTAGATGG
SHP1-F	GTGGACAGCTTAGTTAAGAC
SHP1-R	CTGCTCCAAAAGCAGTATTG

SHP1-303F	CAAGGTAGCAACGAGTACTTG
YBP2-F	CGTTTCGCTGTGAAACTAAC
YBP2-R	CGTACTTTCTGGGTTCCAATG
YBP2-571F	GTAACAACCAACGTTCAAGG
YPI1-F	CCGTTTCTTAGCTTAAAGGC
YPI1-R	CAATACAGAATGCCAAGGG
IPL1-F2	GATACTAAGAAACAAGCCCTTTTGGGAAAATAAGCGTTACGGATCCCCGGGTTAATTAA
IPL1-R1	ATTAATAGTGCCCTTCAAACGATTCTGTCATACTTTAATTGAATTCGAGCTCGTTTAAAC
NDC80-F2	GTTACGAAATTTGGAGTTTGAAACTGAACATAACGTAACAAATCGGATCCCCGGGTTAATTAA
NDC80-R1	CTTTGCTGTAGATTGCTCGGGTATTATATATCATTATTTTAGAATTCGAGCTCGTTTAAAC
PDS1-F2	AGAAGGCCTCGATCCTGAAGAACTAGAGGACTTAGTTACTCGGATCCCCGGGTTAATTAA
PDS1-R1	ACGTGTATATATGTTGTGTGTATGTGAATGAGCAGTGGATGAATTCGAGCTCGTTTAAAC
SHP1-F2	CGCTGATCTGCTGAACTCCGTTGTCGTGCAAAGATGGGCACGGATCCCCGGGTTAATTAA
SHP1-R1	GTTGAAGTCTTTTCCGTTTCTGTTTTGTATATTTATGCGAATTCGAGCTCGTTTAAAC

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