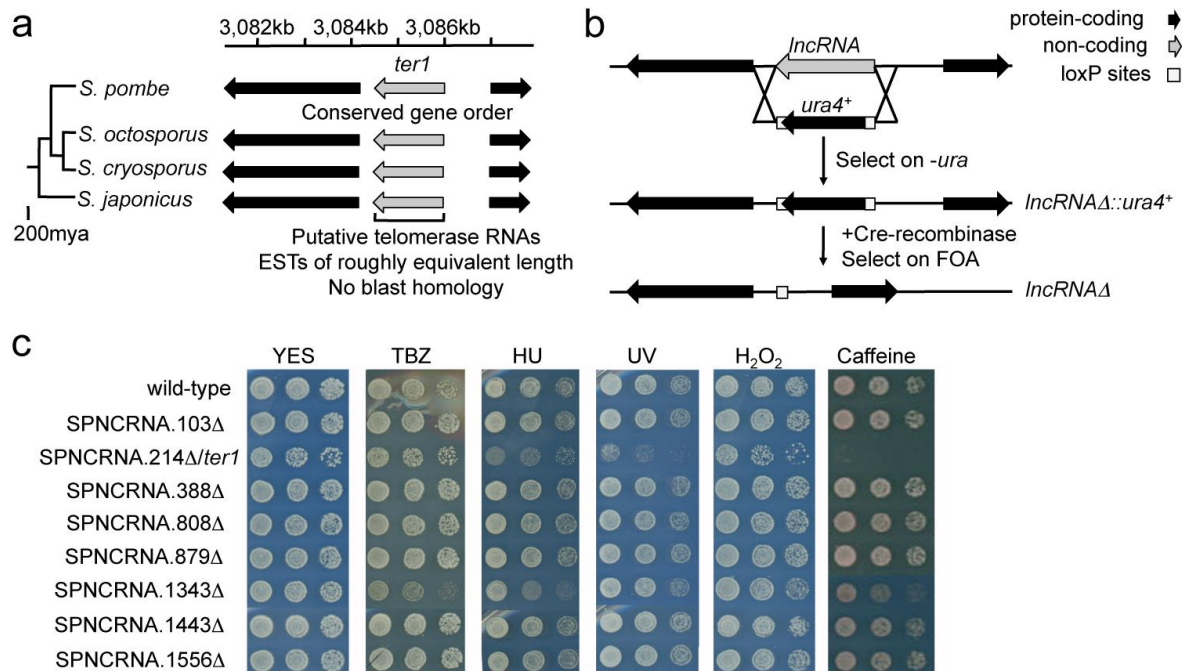
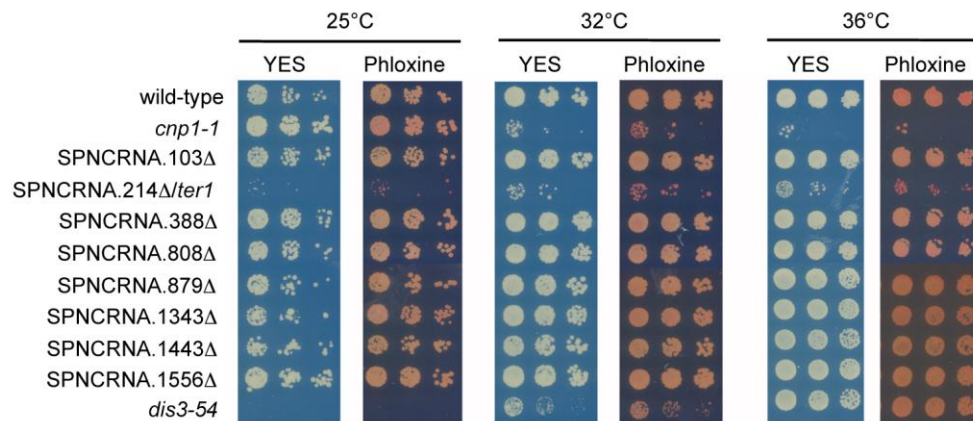


Ard\_Supplementary-Figure-1



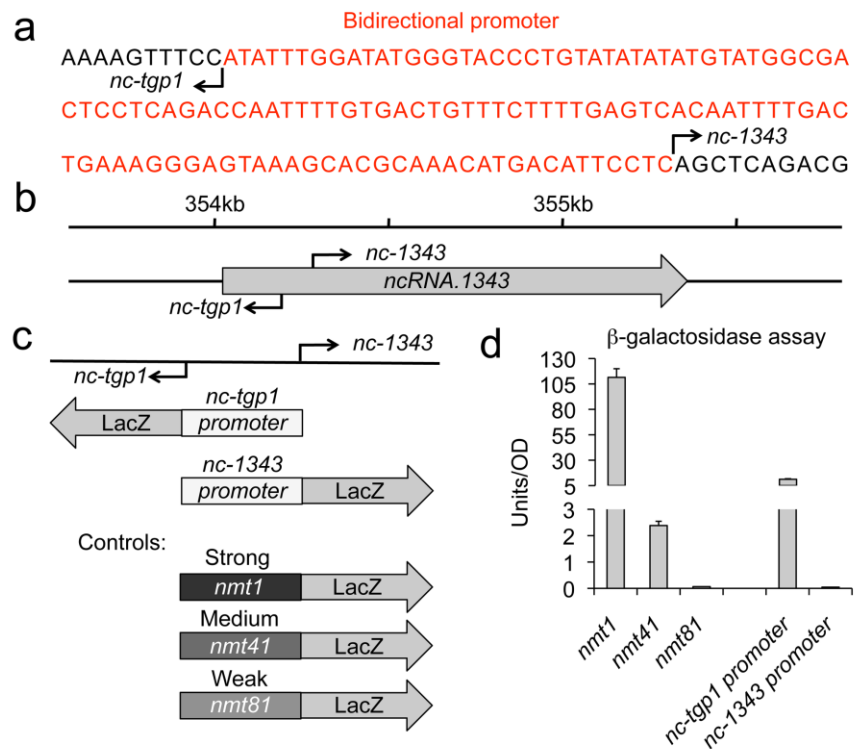
**Supplementary Figure 1. Deletion of the IncRNA locus SPNCRNA.1343 results in sensitivity to multiple drugs.** (a) Schematic representation of *S. pombe* IncRNA (in this case: SPNCRNA.214, telomerase RNA or *ter1*) with conserved synteny in related *Schizosaccharomyces* species. (b) Schematic diagram of the strategy employed to delete IncRNAs in *S. pombe*. (c) Serial dilutions of IncRNA deletions were spotted on non-selective YES medium or in the presence of various stresses, including exposure to the microtubule destabilizing drug thiabendazole (TBZ; 20  $\mu$ g/mL), DNA synthesis inhibitor hydroxyurea (HU; 10 mM), UV-irradiation (80 J/m<sup>2</sup>), oxidative stress (H<sub>2</sub>O<sub>2</sub>; 1 mM), or caffeine (15 mM).

Ard\_Supplementary-Figure-2



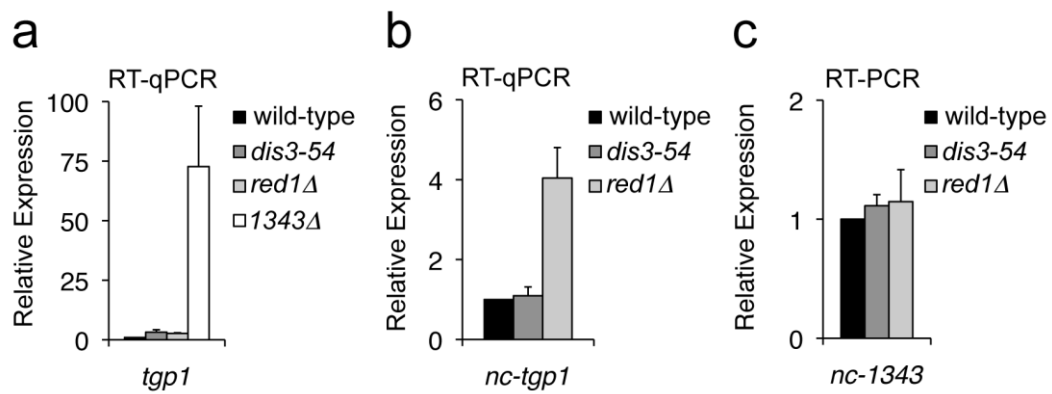
**Supplementary Figure 2. IncRNA deletions, excluding *ter1*<sup>+</sup> control, are viable at 25, 32, and 36°C.** Serial dilutions of IncRNA candidate deletions were spotted on non-selective YES medium or on plates containing phloxine B, which indicates the proportion of dead cells in a colony. *cnp1-1* and *dis3-54* are temperature-sensitive strain controls.

Ard\_Supplementary-Figure-3

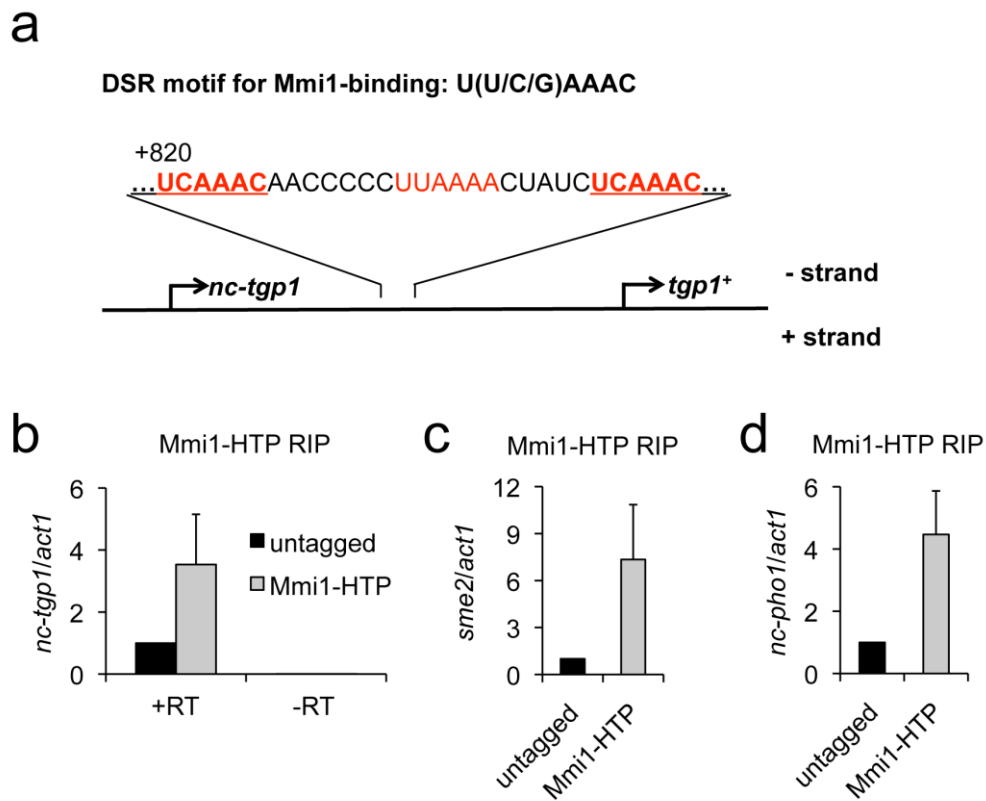


**Supplementary Figure 3. A bidirectional lncRNA promoter upstream of *tgp1*<sup>+</sup> preferentially drives transcription of *nc-tgp1*.** (a) Transcription start sites (TSSs) for *nc-tgp1* and *nc-1343* were mapped by 5'RACE and indicated by arrows. Bidirectional non-coding promoter is underlined and displayed in red. (b) Schematic of *nc-tgp1* and *nc-1343* TSSs depicted within the annotated SPNCRNA.1343 locus. (c) Diagram of LacZ reporter under the control of the bidirectional promoter encoded within the *ncRNA.1343* locus (in both orientations) or control *nmt* promoters of varying strength (as indicated). (d) β-galactosidase assays from wild-type cells transformed with LacZ vectors.

Ard\_Supplementary-Figure-4



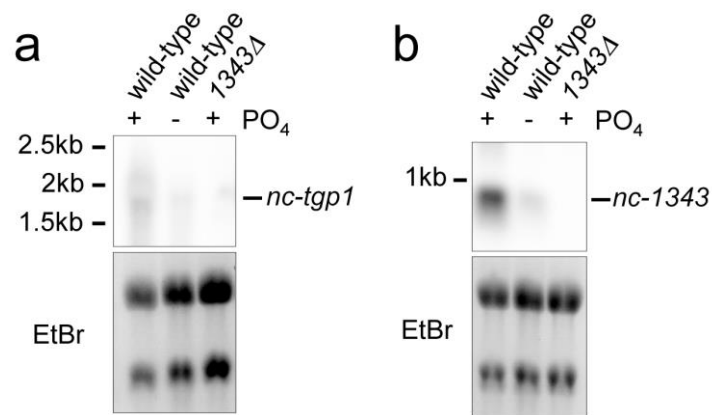
**Supplementary Figure 4. Transcript levels at the *tgp1*<sup>+</sup> locus in Dis3 and Red1 mutants.** RT-qPCR experiments to measure *tgp1*<sup>+</sup> (a), *nc-tgp1* (b), and *nc-1343* (c) transcript levels in wild-type, *dis3-54* and *red1Δ*. RNA levels quantified as relative to wild-type after normalization to *act1*<sup>+</sup>; error bars indicate standard error from three independent experiments.



**Supplementary Figure 5. *nc-tgp1* contains putative DSR sites for Mmi1-binding. (a)**

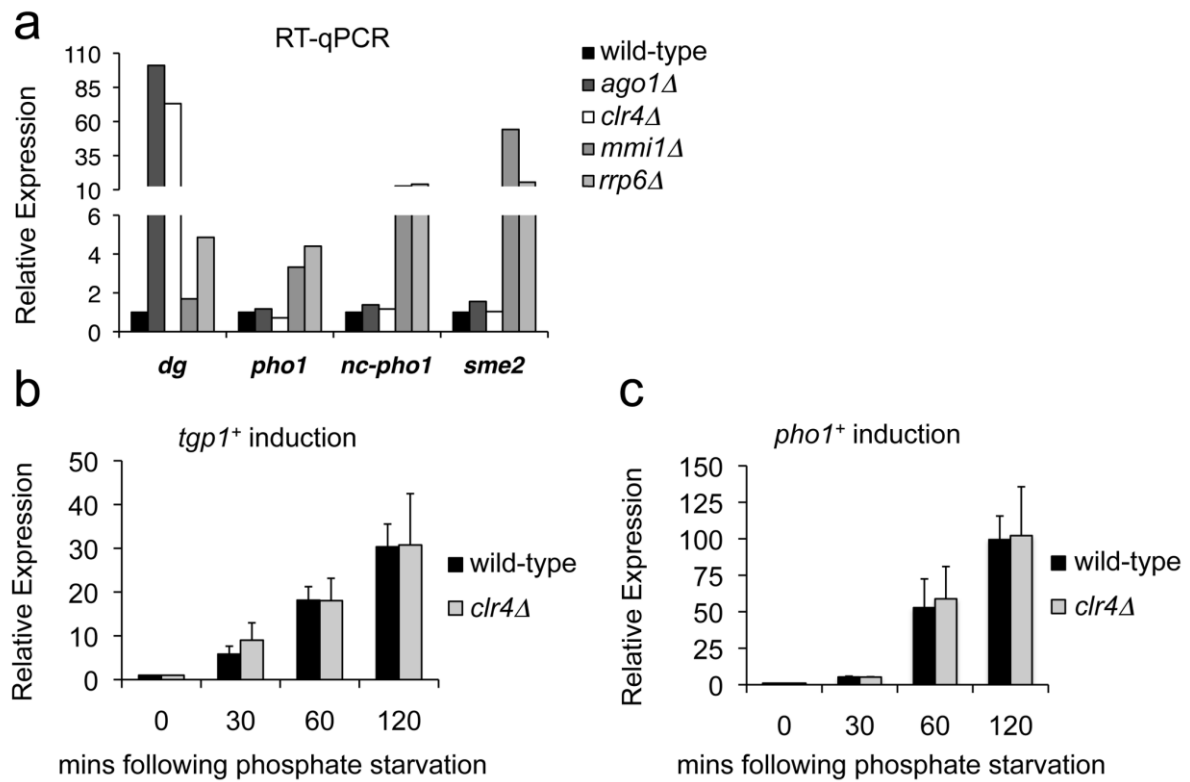
Schematic showing three putative DSR elements (two canonical: bold red text/underlined; one suboptimal: red text) embedded within the *nc-tgp1* transcription unit. Mmi1-HTP RIP and quantification by RT-qPCR for (a) *nc-tgp1*, (b) *sme2*<sup>+</sup>, and (c) *nc-pho1* binding. Error bars indicate standard error from two independent experiments.

Ard\_Supplementary-Figure-6



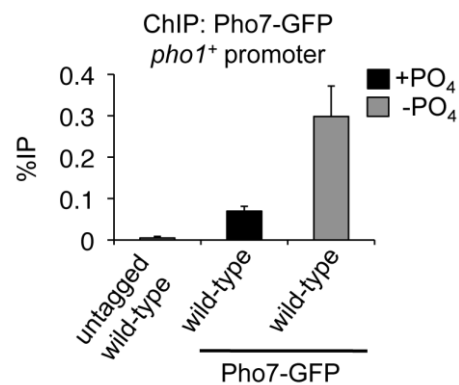
**Supplementary Figure 6. Decreased *nc-tgp1* and *nc-1343* lncRNA levels following phosphate starvation.** Northern analysis of *nc-tgp1* (a) and *nc-1343* (b) in the presence or absence of phosphate, and *1343*Δ grown in the presence of phosphate.

Ard\_Supplementary-Figure-7



**Supplementary Figure 7. Transcript levels in various mutants and conditions.** (a) RT-qPCR experiments to measure *dg* (centromere repeats), *pho1<sup>+</sup>*, *nc-pho1*, and *sme2<sup>+</sup>* transcript levels in wild-type, *ago1Δ*, *clr4Δ*, *mmi1Δ*, and *rrp6Δ*. RNA levels quantified as relative to wild-type after normalization to *act1<sup>+</sup>*. RT-qPCR to measure (b) *tgp1<sup>+</sup>* and (c) *pho1<sup>+</sup>* mRNA induction kinetics following phosphate starvation in wild-type and *clr4Δ* cells. Error bars indicate standard error from two independent experiments.

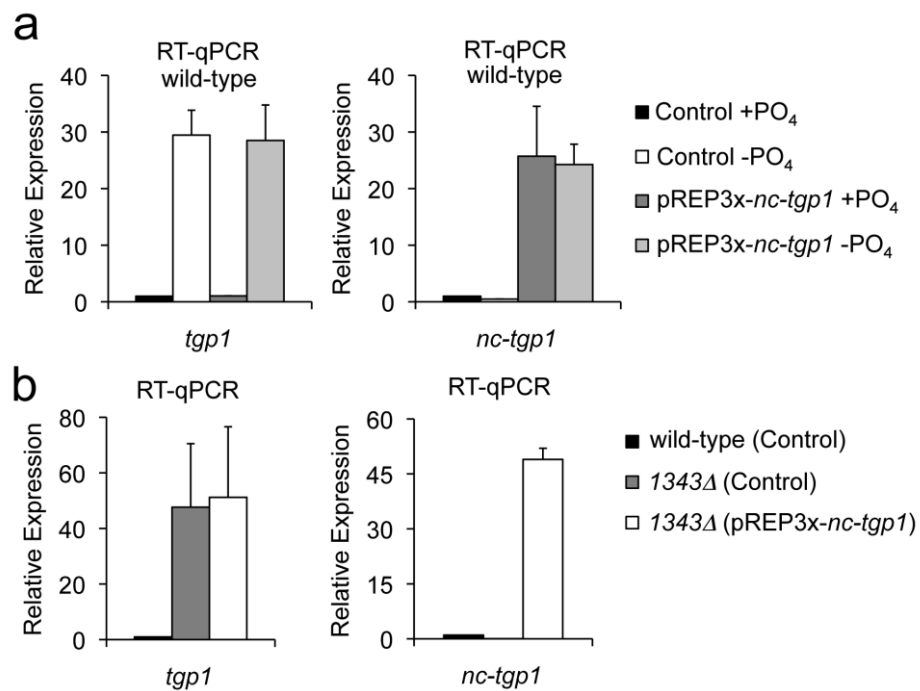
Ard\_Supplementary-Figure-8



**Supplementary Figure 8. Pho7-GFP accumulates at the *pho1*<sup>+</sup> promoter following phosphate starvation.** The endogenous Pho7 transcription factor was C-terminally tagged with GFP in wild-type cells. GFP ChIP experiments were performed in the presence or absence of phosphate. An untagged strain was used as a negative control. Pho7-GFP levels at the *pho1*<sup>+</sup> promoter were measured by qPCR and quantified as %IP (ChIP/Input); error bars indicate standard error from three independent experiments.



Ard\_Supplementary-Figure-9



**Supplementary Figure 9. *nc-tgp1* does not repress *tgp1*<sup>+</sup> *in trans*.** RT-qPCR

experiments to measure *tgp1*<sup>+</sup> mRNA and *nc-tgp1* lncRNA levels in (a) wild-type responding to phosphate availability, and (b) 1343Δ cells, each transformed with an empty pREP3x vector (Control) or pREP3x vector containing *nc-tgp1* under the control of the strong *nmt1* promoter (pREP3x-*nc-tgp1*). Error bars indicate standard error from two independent experiments.

**Supplementary Table 1. List of *S. pombe* strains used in this study.**

Name	ID #	Genotype	Source
wild-type	143	"972 h-"	Lab stock
wild-type	1645	<i>h+ ade6-210 arg3-D4 his3-D1 leu1-32 ura4-D18</i>	Lab stock
wild-type	1646	<i>h- ade6-210 arg3-D4 his3-D1 leu1-32 ura4-D18</i>	Lab stock
<i>rrp6Δ</i>	7865	<i>h+ rrp6Δ::kan ade6-210 ura4-D18 leu1-32</i>	Lab stock
<i>ago1Δ</i>	8061	<i>h+ ago1Δ::ura4 otr1R(SphI):ade6+ ura4-D18 leu1-32 ade6-M210</i>	Lab stock
<i>dcr1Δ</i>	8146	<i>h? dcr1Δ::KAN (G418R) ade6-210</i>	Lab stock
<i>swi6Δ</i>	951	<i>h90 swi6Δ::ura4 ura4-D18</i>	Lab stock
<i>clr4Δ</i>	8435	<i>h- clr4Δ::ura4 his7-366 ade6-210/216 leu1-32 ura4-D18</i>	Lab stock
<i>dis3-54</i>	A1264	<i>h+ dis3-54 ade6-216 leu1-32 arg3-D4</i>	Lab stock
<i>1343Δ::ura4<sup>+</sup></i>	A9016	<i>h+ SPNCRNA.1343Δ::ura4<sup>+</sup>ade6-210 arg3-D4 his3-D1 leu1-32 ura4-D18</i>	This study
<i>1343Δ</i>	A9032	<i>h+ SPNCRNA.1343Δ ade6-210 arg3-D4 his3-D1 leu1-32 ura4-18</i>	This study
<i>tgp1Δ1343Δ</i>	A9352	<i>h+ 1343Δ tgp1Δ::ura4<sup>+</sup> ade6-210 arg3-D4 his3-D1 leu1-32 ura4-D18</i>	This study
<i>red1Δ</i>	A9392	<i>h90 red1Δ::kan leu1-32 ura4-D18 ade6-M210</i>	Sugiyama, T.
<i>mmi1Δ</i>	A9393	<i>h- mmi1Δ::kan leu1-32 mei4-P572</i>	Sugiyama, T.
<i>AΔ</i>	A9520	<i>h+ 1343AΔ ade6-210 arg3-D4 his3-D1 leu1-32 ura4-D18</i>	This study
<i>BΔ</i>	A9522	<i>h+ 1343BΔ ade6-210 arg3-D4 his3-D1 leu1-32 ura4-D18</i>	This study
<i>nc-tgp1::ura4<sup>+</sup></i>	A9523	<i>h+ nc-tgp1::ura4+ ade6-210 arg3-D4 his3-D1 leu1-32 ura4-D18</i>	This study
<i>Pho7-GFP</i>	A9827	<i>h- pho7-GFP:NAT ade6-210 arg3-D4 his3-D1 leu1-32 ura4-D18</i>	This study
<i>Pho7-GFP/1343Δ::ura4<sup>+</sup></i>	A9974	<i>h- pho7-GFP:NAT 1343Δ::ura4<sup>+</sup>ade6-210 arg3-D4 his3-D1 leu1-32 ura4-D18</i>	This study
<i>103Δ</i>	A9027	<i>h+ SPNCRNA.103Δ ade6-210 arg3-D4 his3-D1 leu1-32 ura4-18</i>	This study
<i>214Δ</i>	A9028	<i>h+ SPNCRNA.214Δ ade6-210 arg3-D4 his3-D1 leu1-32 ura4-18</i>	This study
<i>388Δ</i>	A9029	<i>h+ SPNCRNA.388Δ ade6-210 arg3-D4 his3-D1 leu1-32 ura4-18</i>	This study
<i>808Δ</i>	A9030	<i>h+ SPNCRNA.808Δ ade6-210 arg3-D4 his3-D1 leu1-32 ura4-18</i>	This study
<i>879Δ</i>	A9031	<i>h+ SPNCRNA.879Δ ade6-210 arg3-D4 his3-D1 leu1-32 ura4-18</i>	This study
<i>1443Δ</i>	A9033	<i>h+ SPNCRNA.1443Δ ade6-210 arg3-D4 his3-D1 leu1-32 ura4-18</i>	This study
<i>1556Δ</i>	A9034	<i>h+ SPNCRNA.1556Δ ade6-210 arg3-D4 his3-D1 leu1-32 ura4-18</i>	This study
<i>nmt1-nc-tgp1</i>	B0200	<i>h- nc-tgp1-promoter:nmt1-NAT ade6-210 arg3-D4 his3-D1 leu1-32 ura4-D18</i>	This study
<i>cnp1-1</i>	6960	<i>h- leu1-32 ura4-D18 cnp1Δ::ura4 lys1::cnp1-1</i>	Lab stock
<i>Mmi1-HTP</i>	B0398	<i>h+ mmi1-his6-TEV-ProA::KAN MX imr1R(NcoI)::ura4<sup>+</sup> ura4D-18 ade6-M216 leu1-32</i>	Vasilieva, L.

**Supplementary Table 2. List of oligonucleotides used in this study.**

<b>Name</b>	<b>Sequence</b>
qAct1_F	GGTTTCGCTGGAGATGATG
qAct1_R	ATACCACGCTTGCTTTGAG
qSme2_F	AAACAAGGGAGGTAACAGACTTAG
qSme2_R	GCATGCATATTCCGTCTTACAATAG
q1271.10c_F	CGCTTCGTATCTTTCTCTTTCC
q1271.10c_R	CCAGTCCTCTTCTTCGGTTGTA
q1271.09a_F (PP: 1*)	TCGGTTGGAATGTTCTAATCAATAC
q1271.09a_R (PP: 1*)	AGACCGGTGATCAAACAATATTTAG
q1271.09b_F (PP: 2*)	TGAAGTAGTTAGACAGGTTAGCGA
q1271.09b_R (PP: 2*)	CTTGTCGTCCAACCTTCTCTTCATC
qnctgp1c_F (PP: 3; pro*)	GGCAGTAAATCTATCTGTAGCGAGT
qnctgp1c_R (PP: 3; pro*)	TACACGGTAAATGTCAAGTCTGCTA
qnctgp1b_F (PP: 4*)	CTGACAAACCAATTATCCCTACACG
qnctgp1b_R (PP: 4*)	GTATTACGATTTGGCAACCTCATCC
qnctgp1a_F (PP: 5*)	TTAAATGCTGCACTCACATACTGAC
qnctgp1a_R (PP: 5*)	ACTCTCCCTTGGGTTCAATTTGATTA
qncRNA1343_F (PP: 6*)	ATACAGACGTGTGGATTGCAA
qncRNA1343_R (PP: 6*)	CCTCTTCTATACGCAATCAATGTC
q1271.08c_F	TTCAAGGAGCATTTCATTCTAAAC
q1271.08c_R	TATGTATCGTTAGTTATGCCTCGTG
qMug96_F	CATCCTATGTTTATTTGTCTGTTGC
qMug96_R	CTCATGATGGTCCTTAAACCTATTG
qPho1_F	CTTTGGACCCTCTAATACATCCGAT
qPho1_R	AAGAGTGTCAAAGTTCTGGATACCA
qncPho1_F	ATGATGTTTGAGATTTACGGGAAGT
qncPho1_R	TTCTGTAAATGTGTCCCGAACCAAA
qDg_F	AATTGTGGTGGTGTGGTAATAC
qDg_R	GGGTTTCATCGTTTCCATTTCAG
lacZ_1_F	TACTACGTCGACCGACTGACCTCAAACCAAACAGCA
lacZ_1_R	TACTACCTGCAGTCACTAATGTCATACTCGGCTTGAG
lacZ_2_F	TACTACCTGCAGCGACTGACCTCAAACCAAACAGCA
lacZ_2_R	TACTACGTCGACTCACTAATGTCATACTCGGCTTGAG
nc-tgp1_Sall_F	TACTACGTCGACCATATCCAAATATGGAAACT
nc-tgp1_XmaI_R	TACTACCCCGGGCTGCCGACTTACAAGTCTCG

\*(PP: primer pairs; pro: promoter)