

# Supplemental Materials

*Molecular Biology of the Cell*

McClure et al.



## Supplemental Experimental Procedures

### Yeast strains and plasmids

Yeast strains and plasmids were constructed using standard molecular genetic procedures and are listed in the tables below. Wild strains are listed, and the two lab strains are BF264-15Du (Richardson et al., 1989) and YEF473 (Bi and Pringle, 1996).

The following have been previously described: *BEM1-GFP* (Howell et al., 2009) and *rsr1::kan<sup>R</sup>*, *HTB2-mCherry* and *GFP-Ste4* (McClure et al., 2015). *STE2-mCherry* (DLB4299) was made by amplifying mCherry using primers that added flanking NotI sites and ligating into DLB3859 as well as using PvuII to change the marker to *nat<sup>R</sup>* (McClure et al., 2015). *MF $\alpha$ 1* was replaced with GFP using the PCR-based method (Baudin et al., 1993) and using pFA6a-GFP(S65T)-KanMX6 as a template. *P<sub>MF $\alpha$ 1</sub>-PSRI-GFP-T<sub>ADH1</sub>:kan<sup>R</sup>* (DLB4273) was made by altering a plasmid from (Kuo et al., 2014) by amplifying *P<sub>MF $\alpha$ 1</sub>* using flanking ApaI and HindIII sites and changing the marker to *kan<sup>R</sup>*. *Psrl* sequences (first 28 residues) target the protein to the plasma membrane. The plasmid was integrated at the *ADH1* locus using SphI. *P<sub>TEF1</sub>-PSRI-GFP-T<sub>ADH1</sub>* was made similarly by amplifying *P<sub>TEF1</sub>* using ApaI and HindIII sites, and integrated into yeast at the *leu2* locus using PpuMI. *P<sub>TDH3</sub>-YFP* was a kind gift from N. Buchler, and the marker was changed to *kan<sup>R</sup>* using PvuII to make DLB4240. It was integrated at the *TDH3* locus using AccI. *STE20-mCherry* was made using the PCR-based method using pFA6a-mCherry-hph<sup>R</sup> as a template.

Supplemental Table S1. Strains list

<i>strain</i>	<i>background</i>	<i>genotype<sup>1</sup></i>	<i>source</i>	<i>original isolation</i>
DLY19552	YJM1418		Strope et al., 2015	oak tree in Japan
DLY19554	YJM1444		Strope et al., 2015	prickly pear in Hawaii, USA
DLY19556	YJM1338		Strope et al., 2015	sour fig in Maryland, USA
DLY19557	YJM451		Strope et al., 2015	clinical sample from UK
DLY19659	YJM1399		Strope et al., 2015	cherry tree from unknown location
DLY19660	YJM1252		Strope et al., 2015	waste water from Spain
DLY19661	YJM1400		Strope et al., 2015	guava from Phillipines
DLY19663	YJM554		Strope et al., 2015	clinical sample from California, USA
DLY19664	YJM1386		Strope et al., 2015	sugar cane juice from Jamaica
DLY19740	YJM1386	<i>MATa<math>\alpha</math></i> , <i>TDH3:P<sub>TDH3</sub>-YFP-T<sub>ADH1</sub>:kan<sup>R</sup></i>	this study	
DLY19741	YJM1386	<i>MATa<math>\alpha</math></i> , <i>HTB2-mCherry:nat<sup>R</sup></i>	this study	
DLY20210	YJM451	<i>MATa<math>\alpha</math></i> , <i>ADH1:P<sub>MF<math>\alpha</math>1</sub>-PSRI-GFP-T<sub>ADH1</sub>:kan<sup>R</sup></i>	this study	
DLY20288	YJM1399	<i>MATa<math>\alpha</math></i> , <i>ADH1:P<sub>MF<math>\alpha</math>1</sub>-PSRI-GFP-T<sub>ADH1</sub>:kan<sup>R</sup></i>	this study	
DLY20651	YJM1418	<i>MATa<math>\alpha</math></i> , <i>ADH1:P<sub>MF<math>\alpha</math>1</sub>-PSRI-GFP-T<sub>ADH1</sub>:kan<sup>R</sup></i>	this study	
DLY20653	YJM1400	<i>MATa<math>\alpha</math></i> , <i>ADH1:P<sub>MF<math>\alpha</math>1</sub>-PSRI-GFP-T<sub>ADH1</sub>:kan<sup>R</sup></i>	this study	
DLY20655	YJM1338	<i>MATa<math>\alpha</math></i> , <i>ADH1:P<sub>MF<math>\alpha</math>1</sub>-PSRI-GFP-T<sub>ADH1</sub>:kan<sup>R</sup></i>	this study	
DLY20657	YJM1444	<i>MATa<math>\alpha</math></i> , <i>ADH1:P<sub>MF<math>\alpha</math>1</sub>-PSRI-GFP-T<sub>ADH1</sub>:kan<sup>R</sup></i>	this study	
DLY20659	YJM1252	<i>MATa<math>\alpha</math></i> , <i>ADH1:P<sub>MF<math>\alpha</math>1</sub>-PSRI-GFP-T<sub>ADH1</sub>:kan<sup>R</sup></i>	this study	

DLY20748	YJM554	<i>MATa/α, ADH1:P<sub>MFa1</sub>-PSR1-GFP-T<sub>ADH1</sub>:kan<sup>R</sup></i>	this study
DLY20845	YJM1386	<i>MATa/α, ADH1:P<sub>MFa1</sub>-PSR1-GFP-T<sub>ADH1</sub>:kan<sup>R</sup></i>	this study
DLY22729	YEF473	<i>MATa/α, ADH1:P<sub>MFa1</sub>-PSR1-GFP-T<sub>ADH1</sub>:kan<sup>R</sup></i>	this study
DLY18930 <sup>2</sup>	YEF473	<i>MATa/α, leu2:P<sub>TEF1</sub>-PSR1-GFP-T<sub>ADH1</sub>:LEU2, rsr1::TRP1</i>	this study
DLY20920	YJM1386	<i>MATa/α, STE2-mCherry:nat<sup>R</sup>, MFa1::GFP:kan<sup>R</sup></i>	this study
DLY20922	YJM451	<i>MATa/α, STE2-mCherry:nat<sup>R</sup>, MFa1::GFP:kan<sup>R</sup></i>	this study
DLY20923	YJM554	<i>MATa/α, STE2-mCherry:nat<sup>R</sup>, MFa1::GFP:kan<sup>R</sup></i>	this study
DLY20931	YJM1418	<i>MATa/α, STE2-mCherry:nat<sup>R</sup>, MFa1::GFP:kan<sup>R</sup></i>	this study
DLY20932	YJM1252	<i>MATa/α, STE2-mCherry:nat<sup>R</sup>, MFa1::GFP:kan<sup>R</sup></i>	this study
DLY21020	YJM1399	<i>MATa/α, STE2-mCherry:nat<sup>R</sup>, MFa1::GFP:kan<sup>R</sup></i>	this study
DLY22160	YJM1444	<i>MATa/α, STE2-mCherry:nat<sup>R</sup>, MFa1::GFP:kan<sup>R</sup></i>	this study
DLY22161	YJM1338	<i>MATa/α, STE2-mCherry:nat<sup>R</sup>, MFa1::GFP:kan<sup>R</sup></i>	this study
DLY19805	YEF473	<i>MATa/α, STE20-mCherry:hph<sup>R</sup>, BEM1-GFP:LEU2</i>	this study
DLY16019	BF264-15Du	<i>MATa/α, GFP-STE4, rsr1::kan<sup>R</sup>, bar1</i>	McClure et al., 2015

<sup>1</sup>genotypes are homozygous except for those noted

<sup>2</sup>heterozygous genotype

### Supplemental references

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