

Supplementary Table 1

Genotype	p > F		
	Genotype	Environment	Genotype X Environment
<i>ste7Δ</i>	0.0784	<0.0020*	0.0005*
<i>mat2Δ</i>	0.0723	0.0027*	0.1439
<i>spo11Δ</i>	0.5405	<0.0001*	0.8091

**Supplementary Table 1. Effect of genotypes and environment on competition.** 2-factor ANOVA was used to analyze the effect of genotype and the environment on the outcomes of the competitions. (\*) represents significant deviations.

**Supplementary Table 2**

Competing genotype	p-value		
	YPD	MS	V8
<i>ste7Δ1</i>	<0.0001*	0.0466*	<0.0001*
<i>ste7Δ2</i>	<0.0001*	0.0677	<0.0001*
<i>mat2Δ1</i>	<0.0001*	0.0118*	<0.0001*
<i>mat2Δ2</i>	0.4123	0.1776	<0.0001*
<i>spo11Δ1</i>	<0.0001*	0.1461	0.0022*
<i>spo11Δ2</i>	<0.0001*	0.3458	0.0106*

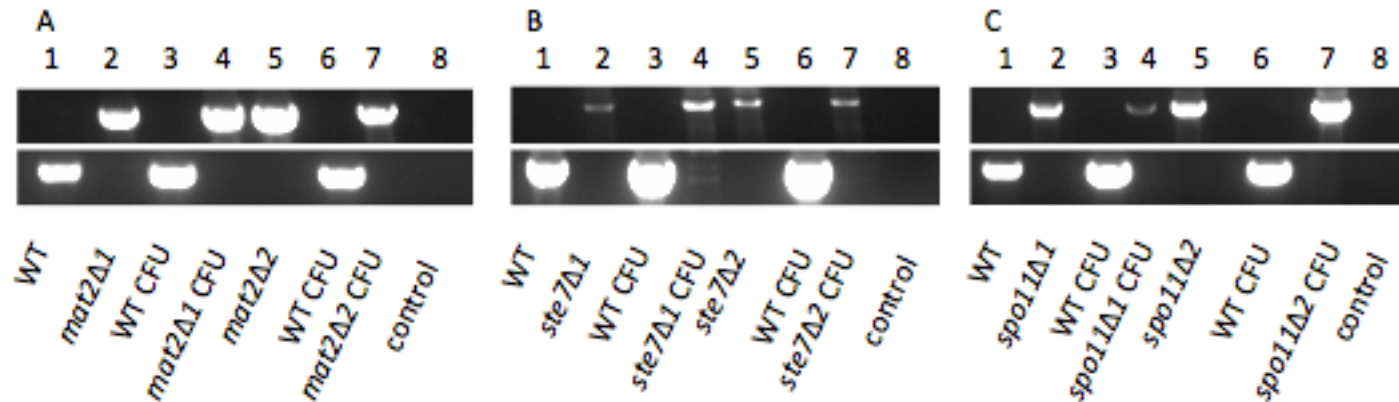
**Supplementary Table 2. Significance testing for the deviation from equality (50% WT).** The binomial two sample proportion test was used to assess the deviation of the observed proportion of wildtype from the equality (50%) expected under the null hypothesis. p-values are provided for the observed proportions at the center of the competition spot. (\*) represents significant deviations.

**Supplementary Table 3**

<b>PCR verification</b>	<b>Primer</b>
<i>ste7</i> $\Delta$	JOHE14073 JOHE15272
<i>mat2</i> $\Delta$	JOHE14087 JOHE15187
<i>spo11</i> $\Delta$	JOHE15249 JOHE15250
NAT <sup>R</sup> cassette	M13F M13R JOHE21081
<b>RT-PCR</b>	<b>Primer</b>
<i>GPD1</i>	XW129 XW130
MF $\alpha$ pheromone	JOHE24434 JOHE24435

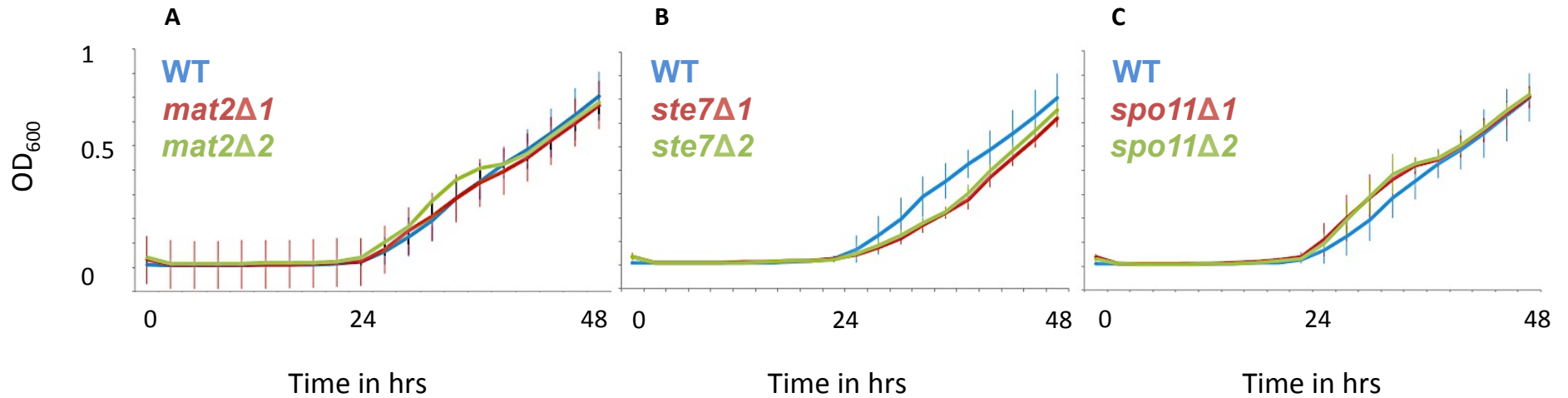
**Supplementary Table 3. Primers used for PCR verification and RT-PCR.**

### Supplementary Figure 1



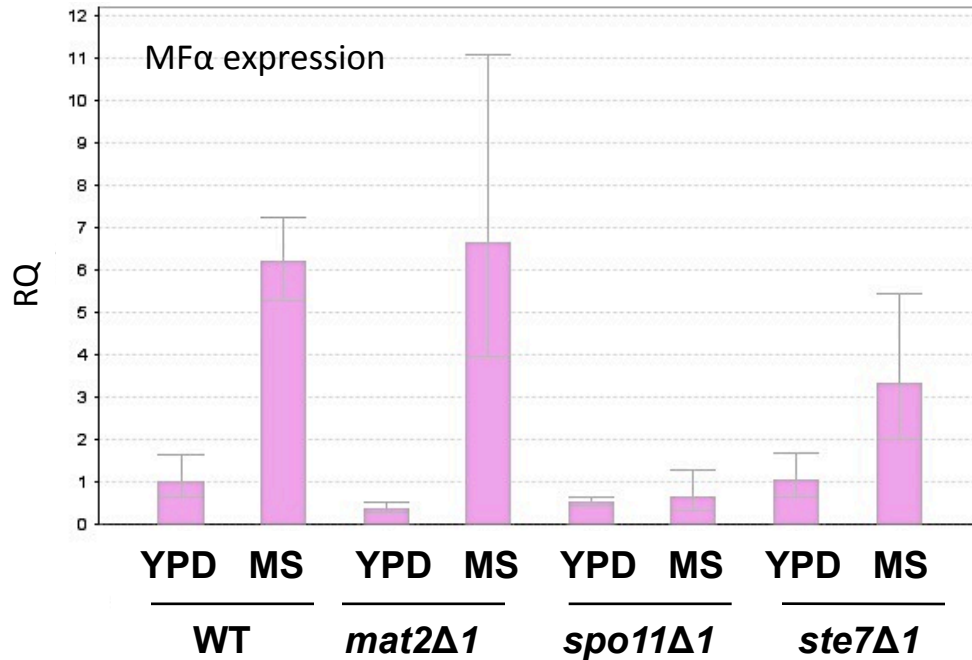
**Supplementary Figure 1. PCR verification of NAT<sup>R</sup> marker and gene deletions.** (Top panel) NAT<sup>R</sup> and (Bottom panels) *MAT2* (A), *STE7* (B) and *SPO11* (C). Each panel shows strains in the following order: XL280 WT, Mutant 1, WT CFU, NAT-R CFU from competitions with mutant 1, Mutant 2, WT CFU, NAT-R CFU from competitions with mutant 2, negative control. As expected, only XL280 WT and WT CFU randomly selected from outcome of competitions have the genes intact and only mutants have the NAT cassette used to replace the genes.

Supplementary Figure 2



**Supplementary Figure 2. Growth curve assays.** (A) *mat2*Δ; (B) *ste7*Δ; (C) *spo11*Δ. OD<sub>600</sub> was measured for 48 hrs at room temperature using the Tecan-Sunrise microplate reader in YPD liquid medium.

Supplementary Figure 3



Genotype	p-value	
	YPD	MS
<i>mat2Δ</i>	0.07	0.58
<i>spo11Δ</i>	0.11	0.0017*
<i>ste7Δ</i>	0.90	0.08

**Supplementary Figure 3. Analysis of pheromone expression.** Expression of the MFα pheromone was compared between YPD and MS agar media for WT and the genotypes *mat2Δ1*, *ste7Δ1*, and *spo11Δ1*. Error bars represent standard deviations based on three replicates. The adjoining table provides p-values based on pairwise t-tests used to compare expression levels between the wildtype and each mutant in the respective media.