Supplementary Table S1. *C. neoformans* strains used in this study

Strain	Genotype	Parent	Reference
H99	*MAT*α		(Perfect *et al.*, 1993)
KN99	*MAT***a**		(Nielsen *et al.*, 2003)
YSB64	*MAT*α *hog1*Δ*::NAT-STM#177*	H99	(Bahn *et al.*, 2005)
YSB123	*MAT*α *pbs2*Δ*::NAT-STM#213*	H99	(Bahn *et al.*, 2005)
YSB261	*MAT*α *ssk1*Δ*::NAT-STM#205*	H99	(Bahn *et al.*, 2006)
YSB264	*MAT*α *ssk2*Δ*::NAT-STM#210*	H99	(Bahn *et al.*, 2007)
YSB349	*MAT*α *skn7*Δ*::NAT-STM#201*	H99	(Bahn *et al.*, 2006)
YSB278	*MAT*α *tco1*Δ*::NAT-STM#102*	H99	(Bahn *et al.*, 2006)
YSB281	*MAT*α *tco2*Δ*::NAT-STM#116*	H99	(Bahn *et al.*, 2006)
YSB324	*MAT*α *tco1*Δ*::NAT-STM#102 tco2*Δ*::NEO*	YSB278	(Bahn *et al.*, 2006)
YSB284	*MAT*α *tco3*Δ*::NAT-STM#119*	H99	(Bahn *et al.*, 2006)
YSB417	*MAT*α *tco4*Δ*::NAT-STM#123*	H99	(Bahn *et al.*, 2006)
YSB286	*MAT*α *tco5*Δ*::NAT-STM#125*	H99	(Bahn *et al.*, 2006)
YSB2469	*MAT*α *tco6*Δ*::NAT-STM#58*	H99	(Lee *et al.*, 2016)
YSB348	*MAT*α *tco7*Δ*::NAT-STM#209*	H99	(Bahn *et al.*, 2006)
YSB1719	*MAT*α *sho1*Δ*::NAT-STM#58*	H99	(Kim *et al.*, 2015)
YSB1720	*MAT*α *sho1*Δ*::NAT-STM#58*	H99	(Kim *et al.*, 2015)
YSB1927	*MAT***a** *sho1*Δ*::NEO*	KN99	This study
YSB1928	*MAT***a** *sho1*Δ*::NEO*	KN99	This study
YSB3191	*MAT*α *msb2*Δ*::NAT-STM#150*	H99	This study
YSB3916	*MAT***a** *msb2*Δ*::NEO*	KN99	This study
YSB3605	*MAT*α *msb2*Δ*::NAT-STM#150 sho1*Δ*::NEO*	YSB3191	This study
YSB3606	*MAT*α *msb2*Δ*::NAT-STM#150 sho1*Δ*::NEO*	YSB3191	This study
YSB3945	*MAT***a** *msb2*Δ*::NEO sho1*Δ*::NAT-STM#58*	YSB3916	This study
YSB3592	*MAT*α *SHO1-6×HA*	H99	This study
YSB3593	*MAT*α *SHO1-6×HA*	H99	This study
YSB3631	*MAT*α *MSB2-4×FLAG*	H99	This study
YSB3698	*MAT*α *SHO1-6×HA MSB2-4×FLAG*	YSB3593	This study
YSB2253	*MAT*α *ssk1*Δ*::NAT-STM#205 sho1*Δ*::NEO*	YSB261	This study
YSB2268	*MAT*α *hog1*Δ*::NAT-STM#177 sho1*Δ*::NEO*	YSB64	This study
YSB2753	*MAT*α *sho1*Δ*::SHO1-GFP*	YSB1719	(Malachowski *et al.*, 2016)
YSB3455	*MAT*α *MSB2-mCherry*	H99	This study
YSB4132	*MAT*α *sho1*Δ*::SHO1-GFP MSB2-mCherry*	YSB2753	This study
YSB3814	*MAT*α *mpk1*Δ*::NAT*	H99	(Lee *et al.*, 2016)
YSB3816	*MAT*α *mpk1*Δ*::NAT*	H99	(Lee *et al.*, 2016)
YSB127	*MAT*α *cpk1*Δ*::NAT*	H99	(Lee *et al.*, 2016)
YSB128	*MAT*α *cpk1*Δ*::NAT*	H99	(Lee *et al.*, 2016)
YSB6089	*MAT*α *mpk1*Δ*::NAT cpk1*Δ*::NEO*	YSB3814	This study
YSB6091	*MAT*α *mpk1*Δ*::NAT cpk1*Δ*::NEO*	YSB3814	This study
YSB6674	*MAT*α *msb2*Δ*::NAT-STM#150 ssk1*Δ*::HYG*	YSB3191	This study
YSB6675	*MAT*α *msb2*Δ*::NAT-STM#150 sho1*Δ*::NEO mpk1*Δ*::HYG*	YSB3606	This study
YSB6676	*MAT*α *msb2*Δ*::NAT-STM#150 sho1*Δ*::NEO mpk1*Δ*::HYG*	YSB3606	This study

Each *NAT-STM#* indicates the Nat^r marker with a unique signature tag.

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Nielsen, K., G. M. Cox, P. Wang, D. L. Toffaletti, J. R. Perfect and J. Heitman (2003) Sexual cycle of Cryptococcus neoformans var. grubii and virulence of congenic a and alpha isolates. Infection and immunity **71**: 4831-4841.
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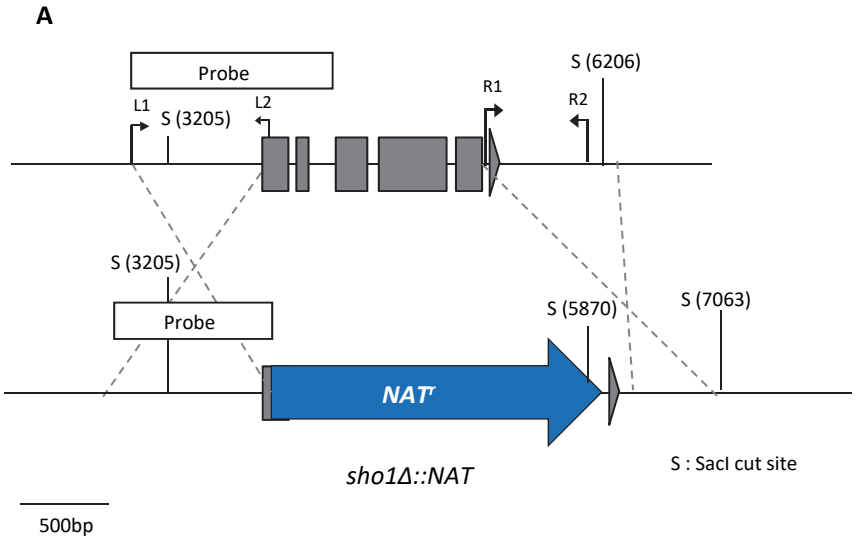
Supplementary Table S2. Primers used in this study

Primer Name	Sequence (5' to 3')	comments
B79	TGTGGATGCTGGCGGAGGATA	Screening primer
B1026	GTAAAACGACGGCCAGTGAGC	M13Forward primer
B1027	CAGGAAACAGCTATGACCATG	M13Reverse primer
B1454	AAGGTGTTCCCCGACGACGAATCG	*NAT* PCR primer SM1
B1455	AACTCCGTCGCGAGCCCCATCAAC	*NAT* PCR primer SM2
B1886	TGGAAGAGATGGATGTGC	*NEO* Split GSR
B1887	ATTGTCTGTTGTGCCCAG	*NEO* Split GSL
B5751	CGAAGAATCTCGTGCTTTC	HSM (Hygromycin Split Marker) 1
B5752	ATTGACCGATTCCTTGCG	HSM2
B4204	ATCTCCCAATCTCCCGAAG	*SHO1* PCR primer L1
B4205	TCACTGGCCGTCGTTTTACAAGAAAGACTGGGTGTCGC	*SHO1* PCR primer L2
B4206	CATGGTCATAGCTGTTTCCTGACACCCGCTGGTATTACAG	*SHO1* PCR primer R1
B4207	AAGTTTTCCTCCACTCGCC	*SHO1* PCR primer R2
B4203	AACGAACGGAAGATTGGC	*SHO1* PCR primer SO
B4208	GCTGCTTACTACATCTGGACG	*SHO1* PCR primer PO
B4504	CGACACCCAGTCTTTCTTG	*SHO1* northern blot probe primer 1
B4505	GTAATACCAGCGGGTGTTC	*SHO1* northern blot probe primer 2
B4794	GTCGACATCTCCCAATCTCCCGAAG	*sho1Δ::SHO1* primer L1
B4795	GCGGCCGCTCACTTATCGTCGTCATCCTTGTAATCAAGCAAGACAAGGTAATTGG	*sho1Δ::SHO1* primer R1
B4796	GCGGCCGCGACTAAAATGATGACTTGAAC	*sho1Δ::SHO1* primer L2
B4797	CCGCGGAAGTTTTCCTCCACTCGCC	*sho1Δ::SHO1* primer R2
B4998	GCTGCTTACTACATCTGGACG	Sho1::GFP primer seq1
B5614	GCGGCCGCATCTCCCAATCTCCCGAAG	Sho1::GFP primer L1
B5610	GCGGCCGCAAGCAAGACAAGGTAATTGGAG	Sho1::GFP primer L2
B6144	TCACCTACACAGCAGACCAC	*MSB2* PCR primer L1
B6145	CTGGCCGTCGTTTTACATGAAGGCGGTGTGTATCG	*MSB2* PCR primer L2
B6146	GTCATAGCTGTTTCCTGGCGTTGAGTAAGCCTAAGC	*MSB2* PCR primer R1
B6147	GAAGAGATGACGGGAGTTATG	*MSB2* PCR primer R2
B6148	AATCTTCGTCCAGTTGTCAC	*MSB2* PCR primer SO
B6395	AGAACTGTTTGAGAAGGCG	*MSB2* PCR primer PO2
B6724	CGCAACATCATAATCGGTG	*MSB2* FLAG PCR primer L1
B6729	GGCTGCTTTTGACAATGG	*SHO1* HA PCR primer L1
B6732	TAATCGCTGAGCCGTTTG	*SHO1* HA PCR primer SO
B6900	CATTCAAACCGCCTCTTC	*MSB2* mCherry PCR primer L1
B6901	CTCCTCGCCCTTGCTCACAGAGCCACCGCCACCTGCGTG	*MSB2* mCherry PCR primer L2
B6902	TGCCACTCGAATCCTGCATGCGACGTGAGACTCGACGG	*MSB2* mCherry PCR primer R1
B6991	CCGTTCAGAATCATACAACC	*MSB2* FLAG PCR primer PO
JOHE12039	CTGTAGAAGATGTGAGTTTGGG	*CPK1* PCR primer L1
JOHE12040	CTGGCCGTCGTTTTACTGATTGATGAGAGATACGGG	*CPK1* PCR primer L2
JOHE12041	GTCATAGCTGTTTCCTGGGCGGAGAAATAGAGGTTG	*CPK1* PCR primer R1
JOHE12042	CGCACAAGAAGTAAGAGGTG	*CPK1* PCR primer R2
JOHE12043	GGCTATGGACCGTATTCAC	*CPK1* PCR primer SO
JOHE12046	ATGCTGCTCACCGTTAGTC	*CPK1* PCR primer PO
B5944	CCGATTTGGAAGTCGTTG	*SSK1* PCR primer L1
B5945	CTGGCCGTCGTTTTACAATCAAAGAGAGGCTGGG	*SSK1* PCR primer L2
B5946	GTCATAGCTGTTTCCTGTCTCCATTTCGCTCGTCAG	*SSK1* PCR primer R1
B5947	GCAAACTCCAAAAACGCTC	*SSK1* PCR primer R2
B5948	GAAACCTTGTCGCTGAGAC	*SSK1* PCR primer SO
B3668	GCAGAGGTTAGAGGAAGTC	*SSK1* PCR primer PO
B5930	TTTGCTTGCTCCTCTTCTC	*MPK1* PCR primer L1
B5931	TCACTGGCCGTCGTTTTACGAGAAGTAGAGGCAGTGACG	*MPK1* PCR primer L2
B5932	CATGGTCATAGCTGTTTCCTGTTGGAGAAACAGTTGGAGAG	*MPK1* PCR primer R1
B5933	TTCAGCAGGTCAATCAGG	*MPK1* PCR primer R2
B5934	CGACTCACGATGTAACTTCC	*MPK1* PCR primer SO
B5935	ACCTCAACTCTCTCAGACACC	*MPK1* PCR primer PO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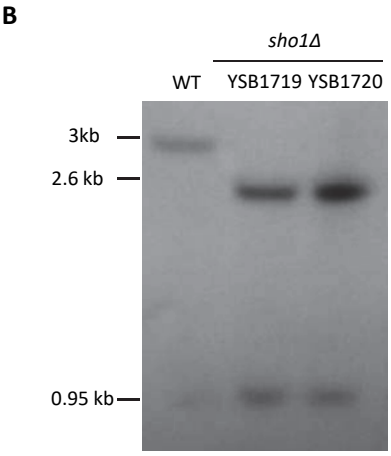
Probe
L1
S (3205)
L2
R1
S (6206)
R2
S (3205)
Probe
S (5870)
S (7063)
NAT^r
sho1Δ::NAT
S : SacI cut site
500bp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YSB1720
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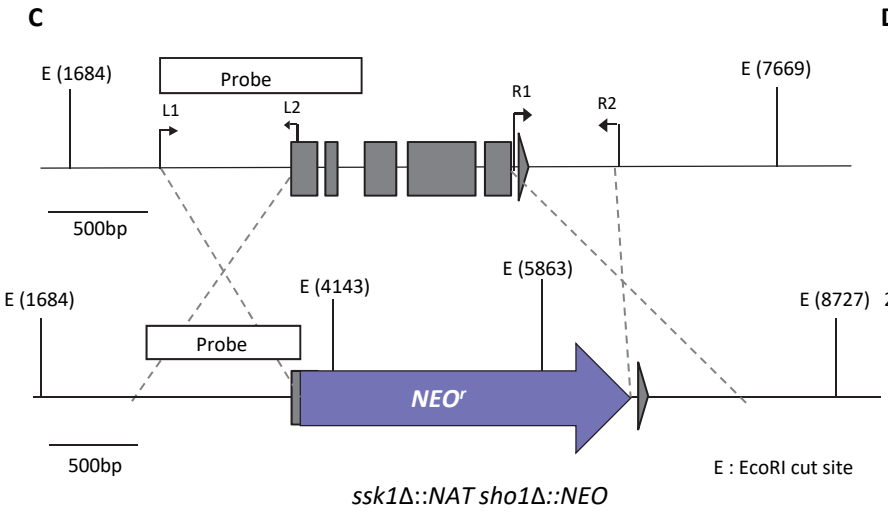
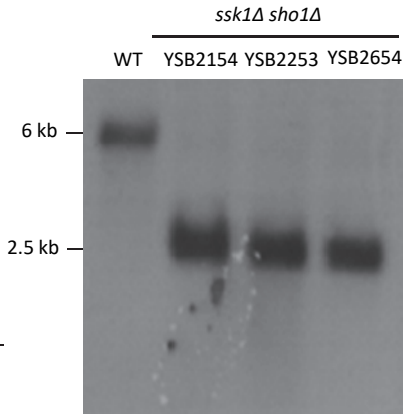
0.95 kb


C
E (1684)
Probe
L1
L2
R1
R2
E (7669)
500bp
E (1684)
E (4143)
E (5863)
E (8727)
Probe
NEO^r
500bp
E : EcoRI cut site
ssk1Δ::NAT sho1Δ::NEO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WT
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YSB2253
YSB2654
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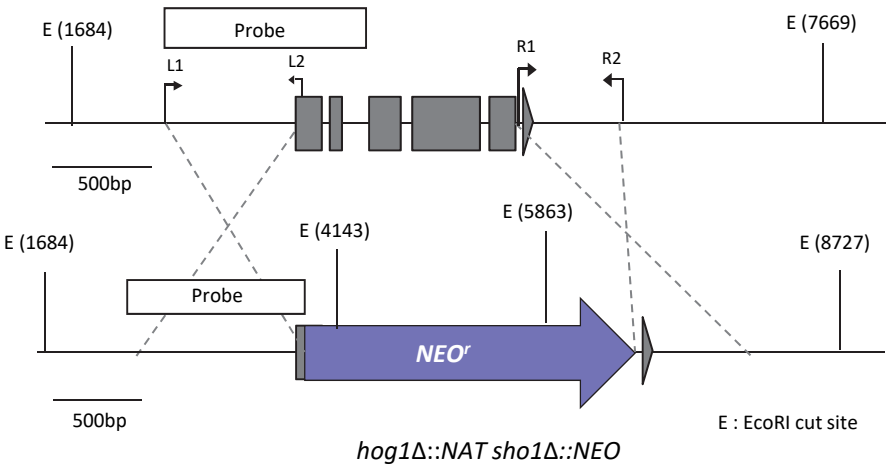
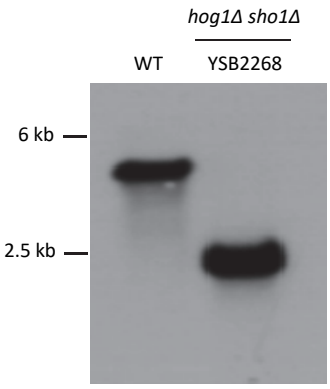
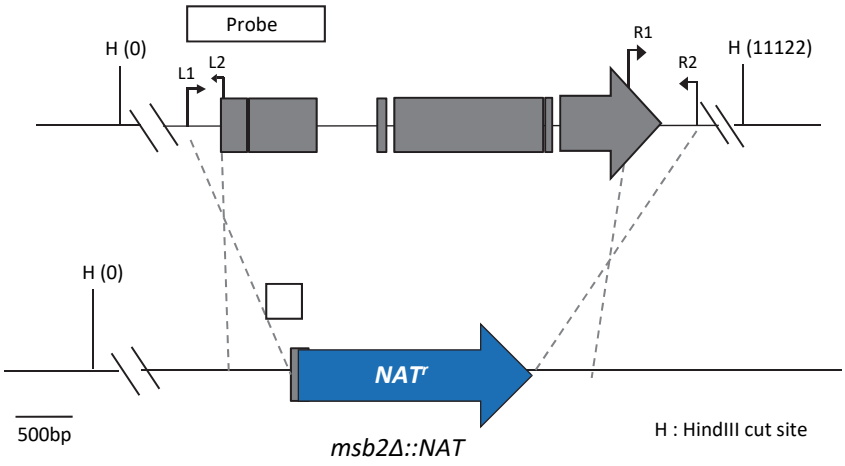
2.5 kb


E
E (1684)
Probe
L1
L2
R1
R2
E (7669)
500bp
E (1684)
E (4143)
E (5863)
E (8727)
Probe
NEO^r
500bp
E : EcoRI cut site
hog1Δ::NAT sho1Δ::NEO


F
hog1Δ sho1Δ
WT
YSB2268
6 kb
2.5 kb

G



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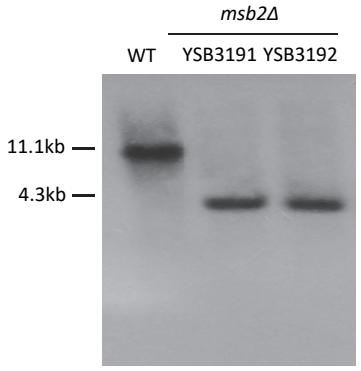

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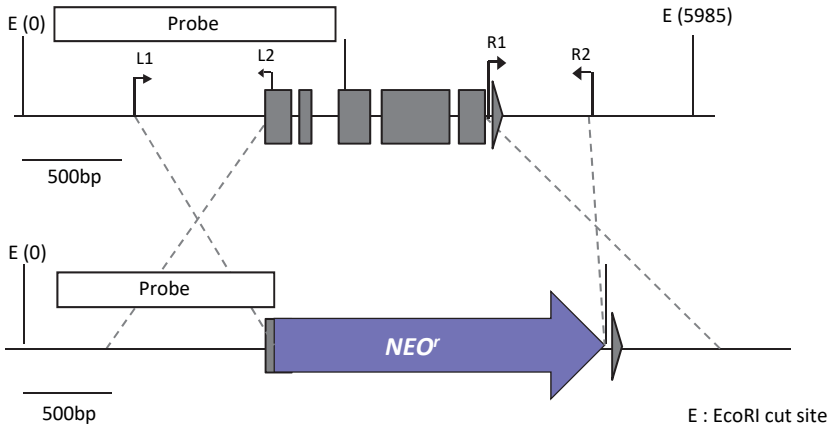
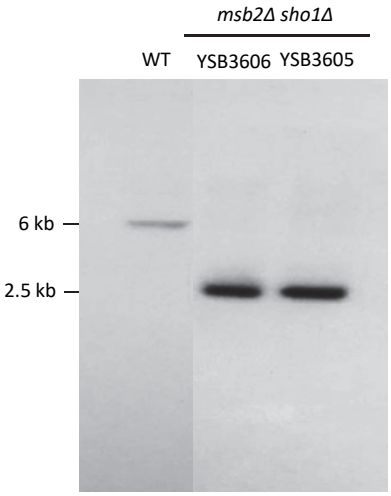


msb2Δ::NAT sho1Δ::NEO

J



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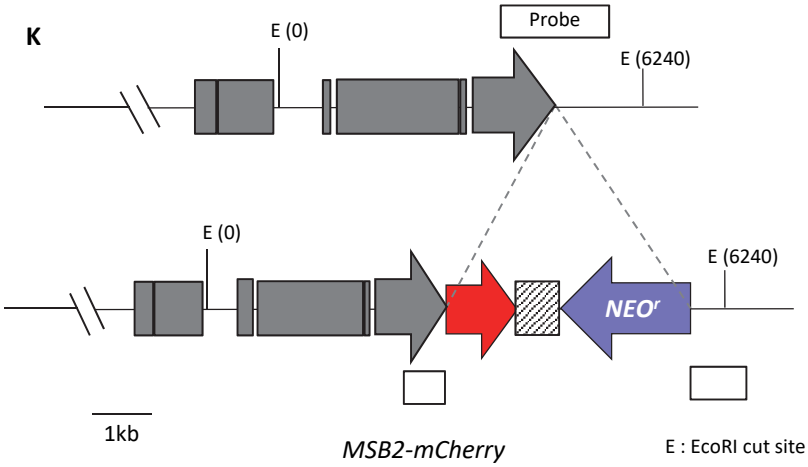

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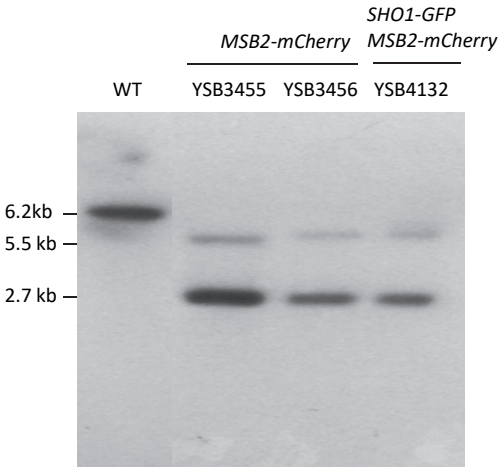

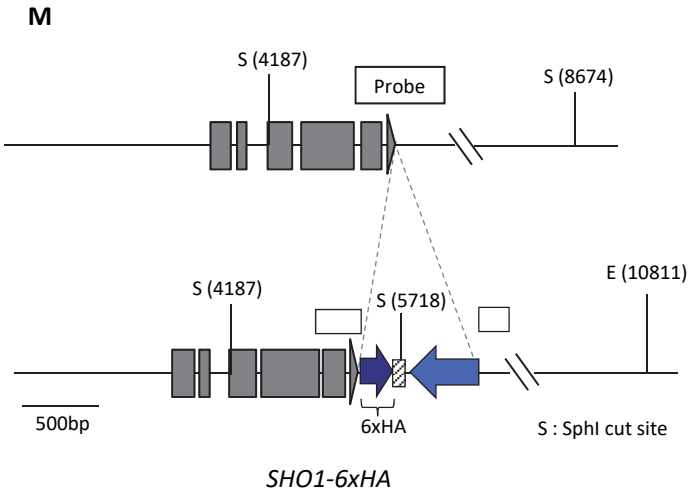
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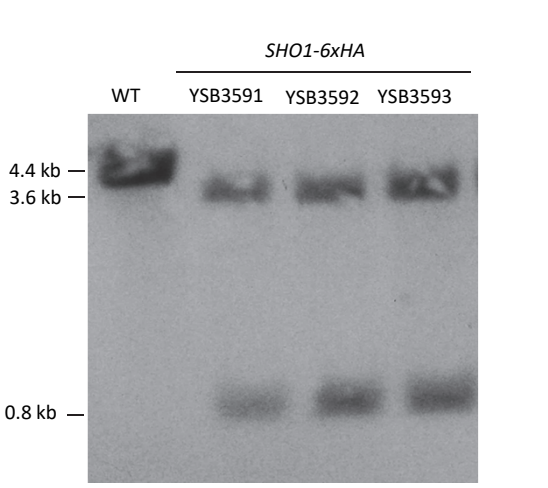
Probe
S (8674)
S (4187)
S (5718)
E (10811)
500bp
6xHA
S : SphI cut site
SHO1-6xHA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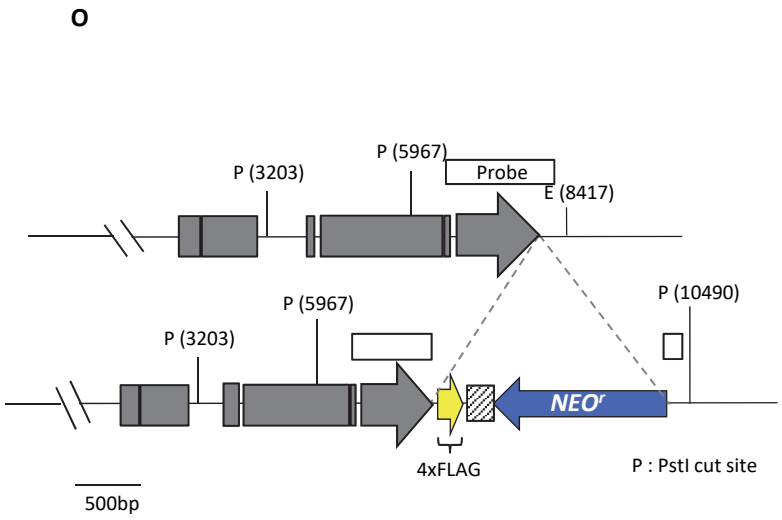
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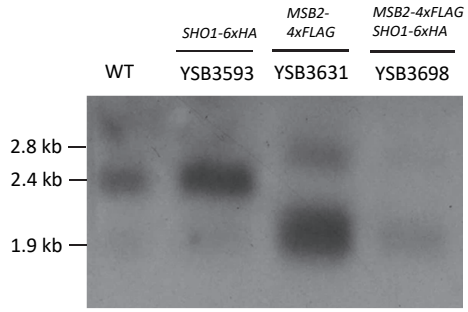
P (5967)
Probe
E (8417)
P (10490)
P (5967)
P (3203)
NEOr
4xFLAG
P : PstI cut site
500bp
MSB2-4xFLAG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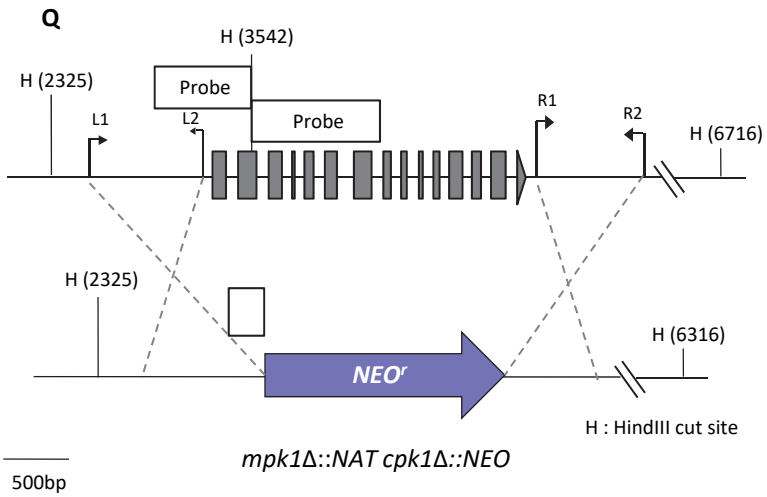
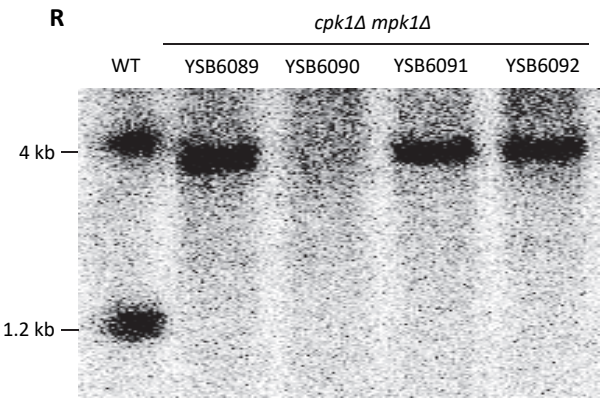

Q
H (3542)
H (2325)
Probe
Probe
L1
L2
R1
R2
H (6716)
H (2325)
H (6316)
NEOr
H : HindIII cut site
500bp
mpk1Δ::NAT cpk1Δ::NEO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WT
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YSB6090
YSB6091
YSB6092
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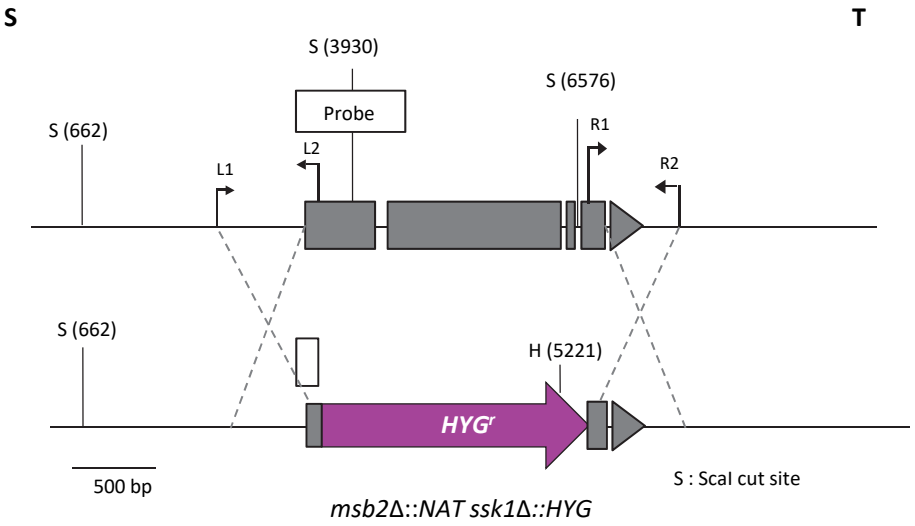
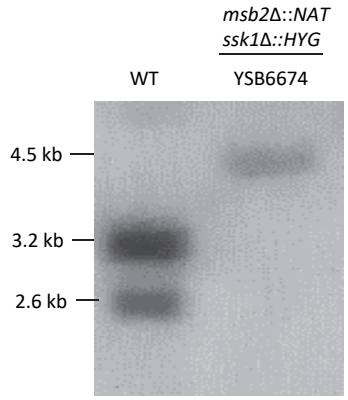
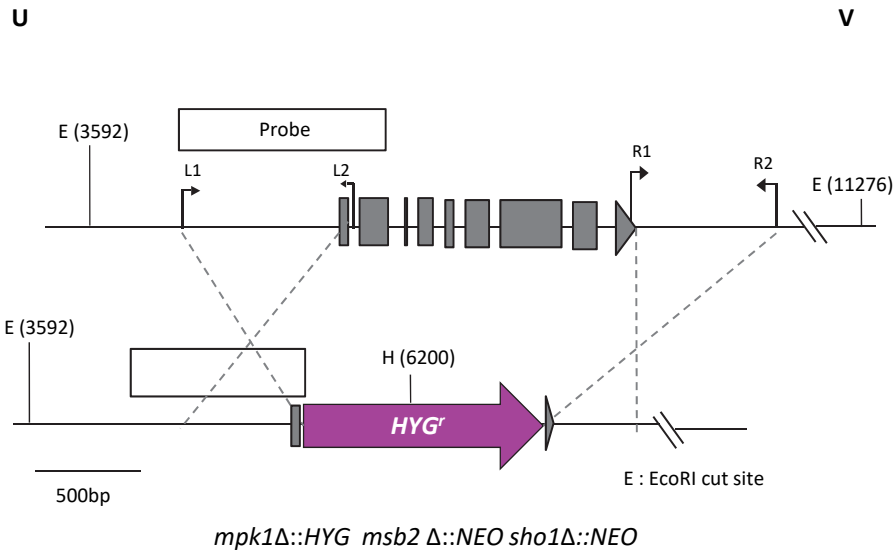
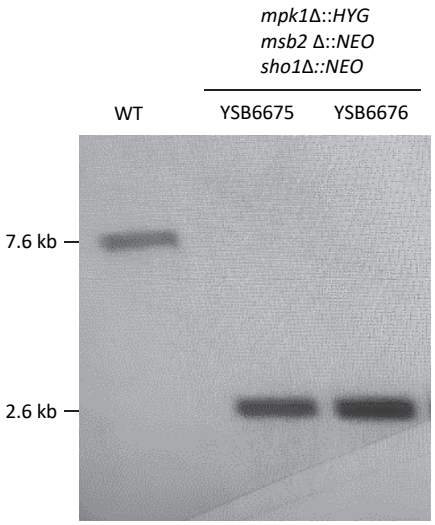
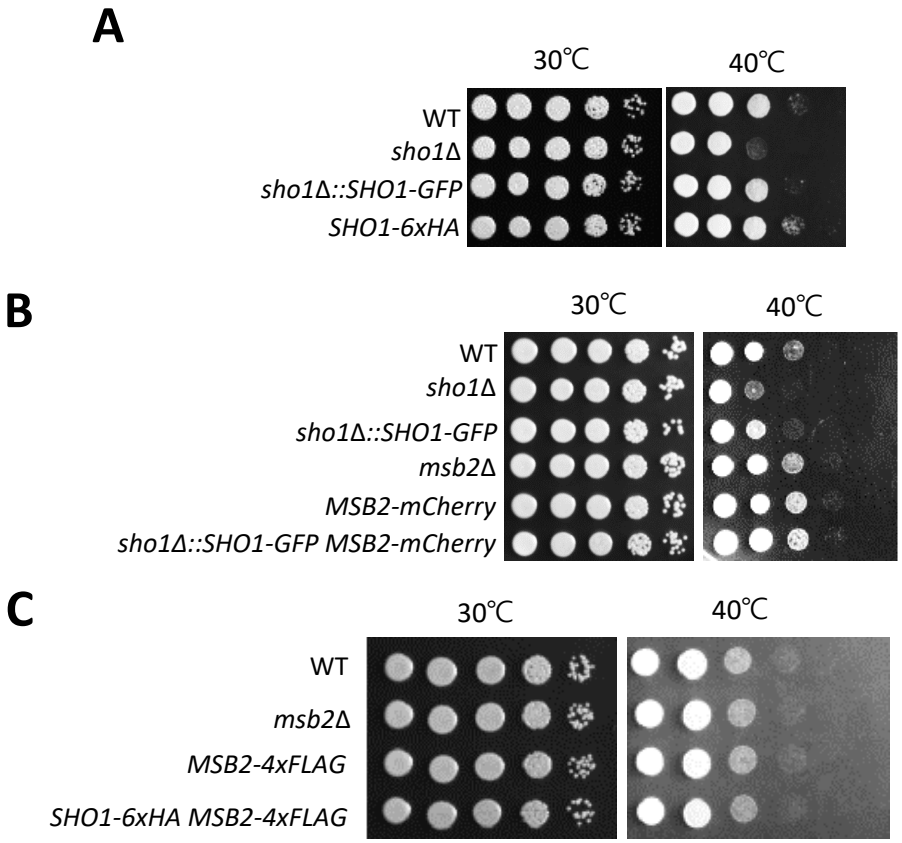




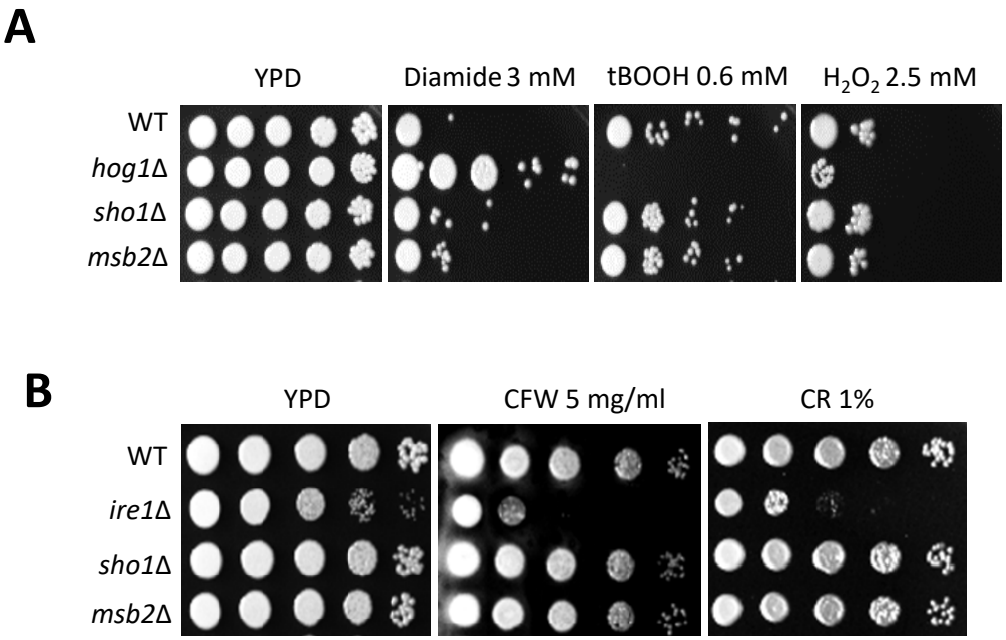




Supplementary Fig. S1. Construction of tagged or deletion strains. (A, C, E, G, I, K, M, O, Q, S and U) Schematic strategy for the construction of tagged or deletion strains. (B, D, F, H, J, L, N, P, R, T and V) The correct genotypes of the strains were confirmed by Southern blot analysis.



Supplementary Figure S2. Phenotypic analyses of tagged strains. *C. neoformans* strains were cultured overnight in liquid YPD medium at 30°C, and then the cultures were spotted on YPD medium. The spotted cells were incubated at 30°C or 40°C. [Strains: WT (H99), *sho1*Δ (YSB1719), *sho1*Δ*::SHO1-GFP* (YSB2753), *SHO1-6xHA* (YSB3593), *msb2*Δ (YSB3191),*MSB2-mCherry* (YSB4128), *sho1*Δ*::SHO1-GFP MSB2-mCherry* (YSB4132), *MSB2-4xFLAG* (YSB3631), *SHO1-6xHA MSB-4xFLAG* (YSB3698)]



Supplementary Fig. S3. Sho1 and Msb2 are dispensable for resistance to oxidative damaging agents and cell wall destabilizers. Strains were grown overnight at 30℃ in liquid YPD medium. The strains were serially diluted and spotted 3µl on YPD medium containing the Diamide, tBOOH, H_2O_2, CFW (calcofluor white)or CR (conco red). The plates were incubated at 30℃ for 2 days and photographed (A-B).