

Supplementary Table S5 - Yeast Strains used in this study

Strain code	Name	Genotype	Origin
yABA19	WT	MATa <i>his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 trp1Δ::NatMX</i>	This study
yABA21	<i>cka2Δ ckb2Δ</i>	MATa <i>his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 trp1Δ::NatMX cka2Δ::HphMX ckb2Δ::KanMX</i>	This study ^{d,g}
LV1434	WT	MATα Ty1- <i>his3AI-Δ1-3114 his3-Δ1 leu2-Δ0 lys2-Δ0 ura3Δ0</i>	(1)
yABA15	Ty1- <i>his3AI cka1Δ</i>	MATα Ty1- <i>his3AI-Δ1-3114 his3-Δ1 leu2-Δ0 lys2-Δ0 ura3Δ0 cka1Δ::KanMX</i>	This study ^a
yABA16	Ty1- <i>his3AI cka2Δ</i>	MATα Ty1- <i>his3AI-Δ1-3114 his3-Δ1 leu2-Δ0 lys2-Δ0 ura3Δ0 cka2Δ::KanMX</i>	This study ^b
yABA17	Ty1- <i>his3AI ckb1Δ</i>	MATα Ty1- <i>his3AI-Δ1-3114 his3-Δ1 leu2-Δ0 lys2-Δ0 ura3Δ0 ckb1Δ::KanMX</i>	This study ^c
yABA18	Ty1- <i>his3AI ckb2Δ</i>	MATα Ty1- <i>his3AI-Δ1-3114 his3-Δ1 leu2-Δ0 lys2-Δ0 ura3Δ0 ckb2Δ::KanMX</i>	This study ^d
yABA24	Ty1- <i>his3AI cka1Δ ckb1Δ</i>	MATα Ty1- <i>his3AI-Δ1-3114 his3-Δ1 leu2-Δ0 lys2-Δ0 ura3Δ0 cka1Δ::KanMX ckb1Δ::HphMX</i>	This study ^{a,e}
yABA25	Ty1- <i>his3AI cka1Δ ckb2Δ</i>	MATα Ty1- <i>his3AI-Δ1-3114 his3-Δ1 leu2-Δ0 lys2-Δ0 ura3Δ0 cka2Δ::KanMX ckb2Δ::HphMX</i>	This study ^{b,f}
yABA26	Ty1- <i>his3AI cka2Δ ckb1Δ</i>	MATα Ty1- <i>his3AI-Δ1-3114 his3-Δ1 leu2-Δ0 lys2-Δ0 ura3Δ0 cka2Δ::HphMX ckb1Δ::KanMX</i>	This study ^{c,g}
yABA27	Ty1- <i>his3AI ckb2Δ ckb1Δ</i>	MATα Ty1- <i>his3AI-Δ1-3114 his3-Δ1 leu2-Δ0 lys2-Δ0 ura3Δ0 ckb2Δ::KanMX ckb1Δ::HphMX</i>	This study ^{d,e}
yABA28	Ty1- <i>his3AI cka2Δ ckb2Δ</i>	MATα Ty1- <i>his3AI-Δ1-3114 his3-Δ1 leu2-Δ0 lys2-Δ0 ura3Δ0 cka2Δ::HphMX ckb2Δ::KanMX</i>	This study ^{d,g}
BY4741	WT	MATa <i>his3Δ1 leu2Δ0 met15Δ0 ura3Δ0</i>	Euroscarf
yABA11	<i>cka2Δ ckb2Δ</i>	MATa <i>his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 cka2Δ::HphMX ckb2Δ::KanMX</i>	This study ^{d,g}
yABA35	<i>ste12Δ</i>	MATa <i>his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 ste12Δ::LEU2</i>	This study
yABA37	<i>ste12Δ cka2Δ ckb2Δ</i>	MATa <i>his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 cka2Δ::HphMX ckb2Δ::KanMX ste12Δ::LEU2</i>	This study ^{d,g,h}
LV1757	Ty1- <i>his3AI spt3Δ</i>	MATα Ty1- <i>his3AI-Δ1-3114 his3-Δ1 leu2-Δ0 lys2-Δ0 ura3Δ0 spt3Δ::HphMX</i>	(2)
LV47	<i>spt3-101</i>	MATα <i>ura3Δ851 trp1Δ63 his3Δ200 spt3-101</i>	
ByΔste12	<i>ste12Δ</i>	MATalpha <i>his3Δ1 leu2Δ0 trp1Δ met15Δ0 ste12Δ::URA3</i>	This study
dCKΔste12	<i>ste12Δ cka2Δ ckb2Δ</i>	MATalpha <i>his3Δ1 leu2Δ0 trp1Δ met15Δ0 ste12Δ::URA3 cka2Δ::HphMX ckb2Δ::KanMX</i>	This study ^{d,g}
Y190		MATa, <i>ura3-52, his3-Δ200, lys2-801, ade2-101, trp1-901, leu2-3,112, gal4Δ, gal80Δ, URA3::GAL1-lacZ, LYS2::GAL4(UAS)-HIS3, cyh^R</i>	(3)
TAP-tagged strains	SC1131 (TAP-Cka1) SC6163 (TAP-Cka2) SC1987 (TAP-Ckb1) SC1485 (TAP-Ckb2) SC1870 (TAP-Tup1) SC5005 (TAP-YKR011C)		Yeast TAP-tagged library, Cellzome

a. *CKA1* complete CDS was deleted by a KanMX cassette amplified from the gDNA of strain BY4741

cka1Δ::KanMX from Euroscarf strain collection clone Id:Y01428

b. *CKA2* complete CDS was deleted by a KanMX cassette amplified from the gDNA of BY4741

cka2Δ::KanMX from Euroscarf strain collection clone Id:Y01837

c. *CKB1* complete CDS was deleted by a KanMX cassette amplified from the gDNA of BY4741

ckb1Δ::KanMX from Euroscarf strain collection clone Id: Y04387

d. *CKB2* complete CDS was deleted by a KanMX cassette amplified from the gDNA of BY4741

ckb1Δ::KanMX from Euroscarf strain collection clone Id: Y01815

e. *CKB1* complete CDS was deleted by a HphMX cassette amplified from pAG32.

f. *CKB2* complete CDS was deleted by a HphMX cassette amplified from pAG32.

g. *CKA2* complete CDS was deleted by a HphMX cassette amplified from pAG32.

Supplementary Table S6 - Plasmids used in this study

Name	Description	Origin
pAG32	for deletion	(4)
pRS305	for deletion	
pFA6a-HphMX6	for deletion	(5)
pGAL-Ty1	2 μ <i>AmpR URA3 pGAL1-Ty1-his3AI</i>	(6)
pABA19	CEN <i>AmpR LEU2 pPSP2-Ty1-his3AI</i>	J. Curcio
pABA20	CEN <i>AmpR URA3 pPSP2-Ty1-his3AI</i>	This study
pPSP2-Ty1 ^{M1}	CEN <i>AmpR URA3 pPSP2-Ty1^{M1}</i> (IN mutations S _{469A} , S _{471A} , Y _{472F} , S _{473A} , T _{477A} , T _{480A} , S _{499A})	This study
pPSP2-Ty1 ^{M3}	CEN <i>AmpR URA3 pPSP2-Ty1^{M3}.his3AI</i> (IN mutations S _{347A} , S _{354A} , S _{360A} , S _{411A} , S _{469A} , S _{471A} , Y _{472F} , S _{473A} , T _{477A} , T _{480A} , S _{499A})	This study
pPSP2-Ty1 ^{M6}	CEN <i>AmpR URA3 pPSP2-Ty1^{M6}.his3AI</i> (IN mutations S _{43A} , S _{347A} , S _{354A} , S _{360A} , S _{411A} , S _{469A} , S _{471A} , Y _{472F} , S _{473A} , T _{477A} , T _{480A} , S _{499A})	This study
pCM185-IN-HBH	CEN <i>AmpR TRP1 TetO₇-CYC1p</i> -IN-HBH	(7)
pCM185-IN	CEN <i>AmpR TRP1 TetO₇-CYC1p</i> -IN	This study
pCM185-IN ^{M6}	CEN <i>AmpR TRP1 TetO₇-CYC1p</i> -IN ^{M6} (IN mutations S _{43A} , S _{347A} , S _{354A} , S _{360A} , S _{411A} , S _{469A} , S _{471A} , Y _{472F} , S _{473A} , T _{477A} , T _{480A} , S _{499A})	This study
pCM185-Ty1	CEN <i>AmpR TRP1 TetO₇-CYC1p</i> -Ty1	This study
pCM185-Ty1 ^{M6}	CEN <i>AmpR TRP1 TetO₇-CYC1p</i> -Ty1 ^{M6} (IN mutations S _{43A} , S _{347A} , S _{354A} , S _{360A} , S _{411A} , S _{469A} , S _{471A} , Y _{472F} , S _{473A} , T _{477A} , T _{480A} , S _{499A})	This study
pAS2-IN-C-tag	2 μ <i>AmpR TRP1</i> GBD-IN-C-tag	This study
pAS2-Cka1	2 μ <i>AmpR TRP1</i> GBD-Cka1	This study
pAS2-Cka2	2 μ <i>AmpR TRP1</i> GBD-Cka2	This study
pAS2-Ckb1	2 μ <i>AmpR TRP1</i> GBD-Ckb1	This study
pAS2-Ckb2	2 μ <i>AmpR TRP1</i> GBD-Ckb2	This study
pACTII-IN-C-tag	2 μ <i>AmpR LEU2</i> GAD-IN-C-tag	This study
pPL121/pACTII-IN	2 μ <i>AmpR LEU2</i> GAD-IN	(8)
pAT26/ pACTII-IN ⁵⁷⁸	2 μ <i>AmpR LEU2</i> GAD-IN ⁵⁷⁸	(8)
pABA10/ pACTII-IN ³⁸⁶⁻⁵¹¹	2 μ <i>AmpR LEU2</i> GAD-IN ³⁸⁶⁻⁵¹¹	This study
pACTII-Cka1	2 μ <i>AmpR LEU2</i> GAD-Cka1	This study
pACTII-Cka2	2 μ <i>AmpR LEU2</i> GAD-Cka2	This study
pACTII-Ckb1	2 μ <i>AmpR LEU2</i> GAD-Ckb1	This study
pACTII-Ckb2	2 μ <i>AmpR LEU2</i> GAD-Ckb2	This study
pET17b-6H-Fh8-IN-C-tag	pBR322 <i>AmpR T7p</i> -6his-Fh8-IN optimized codon-C-tag	(2)
pET17b-6H-Fh8-IN _c -C-tag	pBR322 <i>AmpR T7p</i> -6his-Fh8-IN ⁵⁷⁸⁻⁶³⁵ optimized codon-C-tag	This study
pET17b-6H-Fh8-IN ^{M1} -C-tag	pBR322 <i>AmpR T7p</i> -6his-Fh8IN ^{M1} optimized codon-C-tag (IN mutations S _{469A} , S _{471A} , Y _{472F} , S _{473A} , T _{477A} , T _{480A} , S _{499A})	This study
pET17b-6H-Fh8-IN ^{M2} -C-tag	pBR322 <i>AmpR T7p</i> -6his-Fh8IN ^{M2} optimized codon-C-tag (IN mutations S _{43A} , S _{347A} , S _{354A} , S _{360A})	This study
pET17b-6H-Fh8-IN ^{M3} -C-tag	pBR322 <i>AmpR T7p</i> -6his-Fh8IN ^{M3} optimized codon-C-tag (IN mutations S _{347A} , S _{354A} , S _{360A} , S _{411A} , S _{469A} , S _{471A} , Y _{472F} , S _{473A} , T _{477A} , T _{480A} , S _{499A})	This study
pET17b-6H-Fh8-IN ^{M4} -C-tag	pBR322 <i>AmpR T7p</i> -6his-Fh8IN ^{M4} optimized codon-C-tag (IN mutations S _{43A} , S _{347A} , S _{354A} , S _{360A} , S _{469A} , S _{471A} , S _{473A})	This study

pET17b-6H-Fh8-IN ^{M5} -C-tag	pBR322 <i>AmpR</i> <i>T7p</i> -6his-Fh8IN ^{M5} optimized codon-C-tag (IN mutations S ₄₃ A, S ₃₄₇ A, S ₃₅₄ A, S ₃₆₀ A, S ₄₆₉ A, S ₄₇₁ A, S ₄₇₃ A, S ₄₉₉ A)	This study
pET17b-6H-Fh8-IN ^{M6} -C-tag	pBR322 <i>AmpR</i> <i>T7p</i> -6his-Fh8IN ^{M6} optimized codon-C-tag (IN mutations S ₄₃ A, S ₃₄₇ A, S ₃₅₄ A, S ₃₆₀ A, S ₄₁₁ A, S ₄₆₉ A, S ₄₇₁ A, Y ₄₇₂ F, S ₄₇₃ A, T ₄₇₇ A, T ₄₈₀ A, S ₄₉₉ A)	This study
pET17b-6H-Sso7dIN-C-tag	pBR322 <i>AmpR</i> <i>T7p</i> -6his-Sso7d-IN optimized codon-C-tag optimized codon-C-tag	(2)
pET17b-6H-Sso7dIN ^{M5} -C-tag	pBR322 <i>AmpR</i> <i>T7p</i> -6his-Sso7d IN ^{M5D} optimized codon-C-tag (IN mutations S ₄₃ D, S ₃₄₇ D, S ₃₅₄ D, S ₃₆₀ D, S ₄₆₉ D, S ₄₇₁ D, S ₄₇₃ D, S ₄₉₉ D)	This study

pAB19 is a derivative of pBJC1280 given by Joan Curcio (CEN *AmpR* *LEU2* p*PSP2*-Ty1-*his3AI*) in which *LEU2* has been replaced by *URA3*.

Supplementary Table S7 - Primers used in this study

Name	Sequence
O-AB18_Act1F	TCGTGCTGTCTTCCCATC
O-AB19_Act1R	AAACGGCTTGGATGGAACCG
O-AB46_TYBOUT	GTGATGACAAAACCTCTTCCG
O-AB91_SNR33OUT	TTTTAGAGTGACACCATCGTAC
O-ABA27_HXT (HXT17, HXT16, HXT15, HXT13)	GACATGGGCCCTGTTGCT TATATTGT
O-AMA14_Ty1-5'2F	TGGAACGCCTCTGAGCACTC
O-AMA15_Ty1-5'2R	CATTAGGTGAGGTTAACATTG
O-AMA34-HIS3-5R	GGCGCAAATCCTGATCCAAA
O-AMA35- HIS3-5F	ACGACCATCACACCACTGAA
O-AMA10_Act1F	ACGTTACCCAATTGAACACG
O-AMA11_Act1R	AGAACAGGGTGTCTTCTGG
O-AMA158_Adapteur F	GTAATACGACTCACTATAGGGCACGCGTGGTTCGACGGCCC GGGCTGGT
O-AMA159_Adapteur R	ACCAGCCC
O-AMA344_PSP2-5F	AGCTTGACTCACCAGCTCTT
O-AMA345_PSP2-3R	TATCGTCGCCACCTTTCTCA

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