

SUPPLEMENTAL MATERIALS AND METHODS

Yeast strains

Yeast strains were either purchased from Euroscarf, Open Biosystems, or purposely made following the transformation, and integration on the chromosome by homologous recombination, of suitable PCR cassettes generated with the DNA templates and the oligonucleotides listed below. FF19 and FF20 were obtained through sporulation of strain Y25421, and isolation of haploids following G418 selection.

Strains purchased:

Strain Ref ID	Strain name	Strain genotype	Strain origin
YDL1542	BY4741 (Y00000)	<i>MAT a his3Δ1 leu2Δ0 met15Δ0 ura3Δ0</i>	Euroscarf
YDL1543	BY4742 (Y10000)	<i>MAT α his3Δ1 leu2Δ0 lys2Δ0 ura3Δ0</i>	Euroscarf
YSC1178-7502608 (YDL2366)	TRM112-TAP	<i>Mat a (his3Δ1) leu2Δ0 met15Δ0 ura3Δ0 TRM112::TAP-HIS3MX6</i>	Open Biosystems
Y25421 (YDL2369)	<i>trm112::kanMX4/ TRM112</i>	<i>Mat a/α; his3Δ1/his3Δ1 leu2Δ0/leu2Δ0 lys2Δ0/LYS2 MET15/met15Δ0 ura3Δ0/ura3Δ0 ynr046w::kanMX4/YNR046w</i>	Euroscarf
YDL2616	<i>bud23::kanMX4</i>	<i>Mat a his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 bud23Δ::kanMX4</i>	Charlie Boone Lab

Strains generated in this work:

Strain Ref ID	Strain name	Strain genotype	Strain construction	Host strain
FF19 (YDL2367) and FF20 (YDL2368)	<i>trm112Δ</i>	<i>ura3Δ0 his3Δ1 leu2Δ0 trm112::kanMX4</i>	Y25421 sporulation	n/a
YDL2492	<i>trf5Δ</i>	<i>MAT a his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 trf5Δ::CgHIS3 MX6</i>	pDL0092 LD808 x LD809	YDL1542
YDL2494	<i>trm112Δ trf5Δ</i>	<i>ura3Δ0 his3Δ1 leu2Δ0 trm112::kanMX4 trf5Δ::CgHIS3MX6</i>	pDL0092 LD808 x LD809	FF19
YDL2488	<i>trf4Δ</i>	<i>Mat a his3Δ1 leu2Δ0 met15Δ0 ura3Δ0 trf4Δ::CgHIS3MX6</i>	pDL0092 LD982 x LD983	YDL1542

YDL2490	<i>trm112Δ trf4Δ</i>	<i>ura3Δ0 his3Δ1 leu2Δ0</i> <i>trm112::kanMX4</i> <i>trf4Δ::CgHIS3MX6</i>	pDL0092 LD982 x LD983	FF19
yVH90 (YDL2649)	<i>trm112Δ</i>	<i>MAT a his3Δ1 leu2Δ0</i> <i>met15Δ0 ura3Δ0</i> <i>trm112Δ::LEU2</i>	pRS315 TRM112delup x TRM112dellow	YDL1542
YDL2618	TAP-alone	<i>MAT a his3Δ1 leu2Δ0</i> <i>met15Δ0 +</i> <i>pBAT1::TAP-URA3-KI</i>		Lafontaine Lab
yHE4	<i>bud23Δ</i>	<i>MAT a his3Δ1 leu2Δ0</i> <i>met15Δ0 ura3Δ0</i> <i>bud23Δ::ura3</i>	pRS316 BUD23auxdelup x BUD23auxdellow	YDL1542

Plasmids used in this study

Plasmid name	Marker, origin of replication	Insert	Reference
pYeDP60	<i>URA3</i> , 2μm	n/a	Prof Pompon
pYeDP60 (Mtq2H6) = pYeVH407	<i>URA3</i> , 2μm	Mtq2(His)6 expressed from GAL10-CYC1 promoter	1
pRS316-RPL25eGFP	<i>URA3</i> , CEN/ARS	Rpl25-eGFP	Prof Hurt
pRS316-RPS2-eGFP	<i>URA3</i> , CEN/ARS	Rps2-eGFP	Prof Hurt
pRS423	<i>HIS3</i> , 2μm		
pRS423 (<i>pGAL::TRM112</i>)	<i>HIS3</i> , 2μm	Trm112 expressed from Gal1 promoter	This study
pACDuet (His ₆ -BUD23)	Cm, p15A <i>E. coli</i> origin	(His)6 Bud23 expressed from T7 promoter	This study
pET11a (TRM112) = pVH451	Ap, Kn, ColE1 origin	Trm112 expressed from T7 promoter	2
pDL0092 (pBS1762)	HIS3 Cg	n/a	Prof Séraphin

pACDuet(His₆Bud23), a derivative of pACYC-Duet1 (Novagen) expressing Bud23

with a His₆ tag on its N terminus was constructed by inserting BUD23's ORF between the NdeI and Bgl II sites. BUD23's ORF was PCR-amplified from yeast (strain BY4741) chromosomal DNA with the following oligonucleotides introducing NdeI and BglII sites (up 5'-

CAGAGAAGAACATATGCACCACCACCACCACT

CACGTCCTGAGGAGTTGGCAC-3'; down 5'-CGTGTGAAGATCTGTGTTTC
TTCTAGAAC-3').

Oligonucleotides used for the construction of yeast strains

BUD23 auxdelup	AGTAGTGCTAAAAAGGTGCGGTACGGAAACACAGG AAATATATCAGAGAAGCGTTCGGTGATGAC
BUD23 auxdellow	GTGTGTATTTTATAGTATACAAAAGAATATGCGTGT GAGGATGTGTGTTCCCTGATGCGGTATTTCTCCT
LD808	ATTTTTATTTCAAATAAACAAACGAGGGCGGAGT TTATTGGGTCGTCCACAGGAAACAGCTATGACC
LD809	TGTGGTATTCTGTATAAATAGTAAATAGTCTATAAG AGTCTATATTGTGGTTGTAAAACGACGCCAGT
LD982	AACGTTACGCTTCATAAAGTGTGAATAAGCAAGGG AACTATACTTGAATCACAGGAAACAGCTATGACC
LD983	GGTACACAGTGATGTACAGTTCACTGCATCATTTAA ACAAAAAGGCACATAGTTGTAAAACGACGCCAGT
TRM112de lup	CATCCTATGATCTCTCGGCTCTACACATCATATTAC TAGCCTAGTCAACCCTTCGGTGATGAC
TRM112de llow	TAGCTGTGTATATAGTTGTCTTCGTTGCGTGC ACACACAGAGTCCTGATGCGGTATTTCTCCT

Oligonucleotides used for Northern-blot and primer extension analysis

LD336	CGGTTTAATTGTCCTA
LD359	TTGTTACCTCTGGGCC
LD339	GGCCAGCAATTCAAGTTA
LD871	CATGGCTTAATCTTGAGAC
LD1148	CTCCGCTTATTGATATGC
LD915	GCGTTCTGATCGATGC
LD1290	ATCCCGGCCGCCTCCATCAC
LD2092	CTCTGAGATGGAGTTGCC
LD2484	CGCTGCTCACCAATGG

Oligonucleotides used for RTqPCR analysis

LD714 (actin-Fwd)	CGTTCCAATTACGCTGGTT
LD715 (actin-Rev)	AGCGGTTTGCATTCTTGT
LD2480 (BUD23-Fwd)	CAGCATATCCAGGCGAAGAT
LD2481 (BUD23-Rev)	CGCCCTCCAGCTCTACTA

SUPPLEMENTAL REFERENCES

1. **Heurgué-Hamard, V., S. Champ, L. Mora, T. Merkulova-Rainon, L. L. Kisseelev, and R. H. Buckingham.** 2005. The glutamine residue of the conserved GGQ motif in *Saccharomyces cerevisiae* release factor eRF1 is methylated by the product of the YDR140w gene. *J Biol Chem* **280**:2439-45.
2. **Heurgué-Hamard, V., M. Graille, N. Scrima, N. Ulryck, S. Champ, H. van Tilburgh, and R. H. Buckingham.** 2006. The zinc finger protein Ynr046w is plurifunctional and a component of the eRF1 methyltransferase in yeast. *J Biol Chem* **281**:36140-8.