

Supplementary Table 1 | *Saccharomyces cerevisiae* strains used in this study.

Strain	Genetic background	Source	No.
YPH499 (WT)	<i>MATa ura3-52 lys2-801_amber ade2-101_orchre trp1-Δ63 his3-Δ200 leu2-Δ1</i>	Ref. 44	1501
BY4741 (WT)	<i>MATa his3Δ1 leu2Δ0 met15Δ0 ura3Δ0</i>	Euroscarf	1354
334 (WT)	<i>MATa pep4-3 prp1-1122 leu2 ura3-112 reg1-501 gal1</i>	Ref. 27	1079
YWO2 (WT)	<i>MATa trp1-1 his3-Δ200 ura3-52 lys2-801 leu2-3,-112</i>	Ref.45	5360
<i>ubx2Δ</i>	BY4741 <i>ubx2::kanMX4</i>	Euroscarf	2430
<i>vms1Δ</i>	BY4741 <i>vms1::kanMX4</i>	Euroscarf	4348
<i>doa1Δ</i>	BY4741 <i>doa1::kanMX4</i>	Euroscarf	4926
<i>dfm1Δ</i>	BY4741 <i>dfm1::kanMX4</i>	Euroscarf	5372
<i>mss1Δ</i>	BY4741 <i>mss1::kanMX4</i>	Euroscarf	1458
<i>cdc48-2</i>	BY4741 <i>cdc48::CDC48-2-kanMX4</i>	Euroscarf	4779
<i>cdc48-3</i>	BY4741 <i>cdc48::CDC48-3-kanMX4</i>	Euroscarf	4778
<i>ufd1-2</i>	BY4741 <i>ufd1::UFD1-2-kanMX4</i>	Euroscarf	4781
<i>npl4-1</i>	YWO2 <i>npl4-1</i>	Ref. 45	5365
<i>pre9Δ</i>	BY4741 <i>pre9::kanMX4</i>	Euroscarf	4345
<i>shp1Δ</i>	BY4741 <i>shp1::kanMX4</i>	Euroscarf	4927
<i>pam17Δ</i>	BY4741 <i>pam17::hphNT1</i>	this study	5301
<i>pam17Δ</i>	YWO2 <i>pam17::hphNT1</i>	this study	5361
<i>ubx2Δ vms1Δ</i>	BY4741 <i>vms1::kanMX4 ubx2::hphNT1</i>	this study	5303
<i>ubx2Δ mss1Δ</i>	BY4741 <i>mss1::kanMX4 ubx2::hphNT1</i>	this study	5304
<i>ubx2Δ pam17Δ</i>	BY4741 <i>ubx2::kanMX4 pam17::hphNT1</i>	this study	5302
<i>cdc48-2 pam17Δ</i>	BY4741 <i>cdc48::CDC48-2-kanMX4 pam17::hphNT1</i>	this study	5346
<i>pre9Δ pam17Δ</i>	BY4741 <i>pre9::kanMX4 pam17::hphNT1</i>	this study	5347
<i>doa1Δ pam17Δ</i>	BY4741 <i>doa1::kanMX4 pam17::hphNT1</i>	this study	5348
<i>vms1Δ pam17Δ</i>	BY4741 <i>vms1::kanMX4 pam17::hphNT1</i>	this study	5349
<i>ufd1-2 pam17Δ</i>	BY4741 <i>ufd1::UFD1-2-kanMX4 pam17::hphNT1</i>	this study	5350
<i>npl4-1 pam17Δ</i>	YWO2 <i>npl4-1 pam17::hphNT1</i>	this study	5363
<i>ubx2Δ + UBX2</i>	BY4741 <i>ubx2::kanMX4</i> pRS415-UBX2	this study	5305
<i>ubx2Δ pam17Δ + UBX2</i>	BY4741 <i>ubx2::kanMX4 pam17::hphNT1</i> pRS415-UBX2	this study	5351
Tom40 _{HA}	BY4741 <i>tom40::TOM40-HA-HIS3MX6</i>	this study	5035

Tom40 _{HA} <i>ubx2</i> Δ	BY4741 <i>ubx2::kanMX4 tom40::TOM40-HA-HIS3MX6</i>	this study	5037
Tom40 _{HA} <i>ubx2</i> Δ + <i>UBX2</i> Δ <i>UBA</i>	BY4741 <i>ubx2::kanMX4 tom40::TOM40-HA-HIS3MX6</i> pRS415- <i>UBX2</i> Δ <i>UBA</i>	this study	5369
Tom40 _{HA} <i>ubx2</i> Δ + <i>UBX2</i> Δ <i>UBX</i>	BY4741 <i>ubx2::kanMX4 tom40::TOM40-HA-HIS3MX6</i> pRS415- <i>UBX2</i> Δ <i>UBX</i>	this study	5370
Tom40 _{HA} <i>ubx2</i> Δ + <i>UBX2</i> Δ <i>iMTS-L</i>	BY4741 <i>ubx2::kanMX4 tom40::TOM40-HA-HIS3MX6</i> pRS415- <i>UBX2</i> Δ <i>iMTS-L</i>	this study	5371
Tom40 _{HA} <i>vms1</i> Δ	BY4741 <i>vms1::kanMX4 tom40::TOM40-HA-HIS3MX6</i>	this study	5307
Tom40 _{HA} <i>doa1</i> Δ	BY4741 <i>doa1::kanMX4 tom40::TOM40-HA-HIS3MX6</i>	this study	5358
Tom40 _{HA} <i>ufd1-2</i>	BY4741 <i>ufd1::UFD1-2-kanMX4 tom40::TOM40-HA-HIS3MX6</i>	this study	5359
Ubx2 _{HA}	BY4741 <i>ubx2::UBX2-HA-HIS3MX6</i>	this study	5036
Ubx2 _{His}	BY4741 <i>ubx2::UBX2-HIS-HIS3MX6</i>	this study	5308
Tom22 _{His}	YPH499 <i>tom22::TOM22-HIS-HIS3MX6</i>	Ref. 37	1456
Hrd1 _{HA}	BY4741 <i>hrd1::HRD1-HA-HIS3MX6</i>	this study	5038
Doa10 _{ProtA}	BY4741 <i>doa10::DOA10-ProtA-hphNT1</i>	this study	5309
Por1 _{HA}	BY4741 <i>por1::POR1-HA-HIS3MX6</i>	Ref. 46	3716
_{HA} Tom5	YPH499 <i>tom5::HA-TOM5</i>	Ref. 32	4052
_{HA} Tom6	YPH499 <i>tom6::HA-TOM6</i>	Ref. 32	4053
_{HA} Tom7	YPH499 <i>tom7::HA-TOM7</i>	Ref. 32	4054
<i>tom20</i> Δ	YPH499 <i>tom20::URA3 pYep13-TOM22</i>	Ref. 15	1273
<i>tom70</i> Δ	YPH499 <i>tom70::HIS3</i>	Ref. 15	1059
<i>mim1</i> Δ	YPH499 <i>mim1::ADE2</i>	Ref. 15	2195
<i>sam37</i> Δ	YPH499 <i>sam37::LEU2</i>	Ref. 31	1271
<i>ubx2</i> Δ (334)	334 <i>ubx2::hphNT1</i>	this study	5310
334-pCYB2-ΔDHFR-HB	334 pDP-CYB2(1-84)Δ(47-65)-DHFR-HB(81-181)	Ref. 27	1080
334-pCYB2-DHFR-HB	334 pDP-CYB2(1-84)-DHFR-HB(81-181)	Ref. 27	1081
<i>ubx2</i> Δ pCYB2-ΔDHFR-HB	334 <i>ubx2::hphNT1</i> pDP-CYB2(1-84)Δ(47-65)-DHFR-HB(81-181)	this study	5312
<i>ubx2</i> Δ pCYB2-DHFR-HB	334 <i>ubx2::hphNT1</i> pDP-CYB2(1-84)-DHFR-HB(81-181)	this study	5311
334 <i>pYEp352</i>	334 <i>pYEp352</i>	this study	5313
334 <i>ubx2</i> Δ <i>pYEp352</i>	334 <i>ubx2</i> Δ <i>pYEp352</i>	this study	5314
<i>ubx2</i> Δ + <i>UBX2-GFP</i>	BY4741 <i>ubx2::kanMX4</i> pUG35-Ubx2-GFP	this study	5345

Supplementary Data Table 2 | Plasmids used in this study.

Plasmid	Source	No.
pRS415	Ref. 15	X24
pRS415-UBX2	this study	P3114
pRS415-UBX2ΔUBA	this study	P3116
pRS415-UBX2ΔUBX	this study	P3117
pRS415-UBX2ΔiMTS-L	this study	P3118
pUG35-Ubx2-GFP	this study	P3115
pFA6a-3xHA-HIS3MX6	Ref. 47	1450; pYM2
pFA6a-His10-HIS3MX6	Ref. 37	X63
pFA6a-ProtA-hphNT1	Ref. 34	P1450
pFA6a-hphNT1	Ref. 35	P2722
pYEp352	Addgene No. 37673	X51
pDP-CYB2 (1-84)Δ(46-66)-DHFR-HB(81-181)	Ref. 27	B55
pDP-CYB2 (1-84)-DHFR-HB(81-181)	Ref. 27	B54

Supplementary Data Table 3 | Primers used in this study.

Name	Sequence (5'→3')	Source	No.
Tom40-HA_fw (HIS3MX6)	CAACAAGGTTTAGACGCAGATGGTAACCCATT GCAAGCTCTTCCTCAATTGCGTACGCTGCAGG TCGAC	this study	PR2220
Tom40-HA_rv (HIS3MX6)	GATAAAAACAATGATTATTTATTCAACCATAA AAAAGCCAAGGGAAGATTTATCGATGAATTCG AGCTCG	this study	PR2221
Ubx2- His/HA_fw (HIS3MX6)	GGAAGCACTGGATGAAGAAGATGAAGAAGAT GAGGAAAATGAAGAACAACGTACGCTGCAGG TCGAC	this study	PR1193
Ubx2- His/HA_rv (HIS3MX6)	CCAGAACTCTTTGTACGCGTTTGTCGTTTTTA ACGATATGCTATTTTAATCGATGAATTCGAGC TCG	this study	PR1194
Hrd1-HA_fw (HIS3MX6)	CAAGAAAATTGTCATACCAGATAAATTTATCC AGCATATCCGTACGCTGCAGGTCGAC	this study	PR2222
Hrd1-HA_rv (HIS3MX6)	TTTCTTTAAAAAAAATCTATGTATAATATAAAA CATGCAATATCGATGAATTCGAGCTCG	this study	PR2223
Porin-HA_fw (HIS3MX6)	GAAGTTGTCTGAACCTGTTCAAGCTAGGTT GGTCTTTGTCCTTCGACGCTCGTACGCTGCAGG TCGAC	this study	PR728
Porin-HA_rv (HIS3MX6)	CGAGCACATATATGGTATATAGTGAACATATA TATATTAGATATATACGTATCGATGAATTCGA GCTCG	this study	PR729
Doa10- ProtA_fw (hphNT1)	CACTAAGGGTAGAGCTTTAGAAAATTTACCAG ATGAAAGTCGTACGCTGCAGGTCGAC	this study	PR2224

Doa10-ProtA_rv (hphNT1)	TATGCTAGCATTTCATTTTAAATGTAAGGAAGA AAACGCCTATCGATGAATTCGAGCTCG	this study	PR2225
Ubx2Δ_fw (hphNT1)	GCAGCAGGTATTACGATAGAAGTATGTAATAG CTTTCATAGTGTAATCGAAGATGCGTACGCTG CAGGTCGAC	this study	PR2226
Ubx2Δ_rv (hphNT1)	CTCCAGAAACTCTTTGTACGCGTTTGTCGTTTT TAACGATATGCTATTTTATCAATCGATGAATTC GAGCTCG	this study	PR2227
Pam17Δ_fw (hphNT1)	GTTCAAAGAAGTGTTAAAAACATTCAGAAAAC ATTGTCCGCCTCTTCAAAATGCGTACGCTGCA GGTCGAC	this study	PR2228
Pam17Δ_rv (hphNT1)	CATATATGTATATATACAGAGTCTGAGAAGAA GGAAAAGATCACACGTTCAATCAATCGATGAA TTCGAGCTCG	this study	PR2229
XhoI-Ubx2-up	CCGCTCGAGCGGTATATTGTTGGGCTCTAGAA CAG	this study	PR2230
XmaI-Ubx2- down	CCCCCGGGGGGCTGTAGATGCTGCAATGTCG	this study	PR2231
Sp6-Ubx2_fw1	TCGATTTAGGTGACACTATAGAATACGCCGCC GCCATGCCAGTAGTCAATCATG	this study	PR2232
Sp6-Ubx2_fw2	TCGATTTAGGTGACACTATAGAATACGCCGCC GCCATGTGGAAAGGCGAACCAGAC	this study	PR2233
Sp6-Ubx2_fw3	TCGATTTAGGTGACACTATAGAATACGCCGCC GCCATGCCCATGGCTGAGACCTTG	this study	PR2234
Sp6-Ubx2_fw4	TCGATTTAGGTGACACTATAGAATACGCCGCC GCCATGACTGTGTGGTCCATGTTTG	this study	PR2235
Ubx2_rv1	GATCTCATTGTTCTTCATTTTCCTC	this study	PR2236
Ubx2_rv2	GATCTTACGTTGTCTCGAACGGC	this study	PR2237
Ubx2_rv3+3M	GATCTTACATCATCATCTCAGCCATGGGTGTA GAC	this study	PR2238
XbaI-Ubx2_fw	GATCTCTAGAATGCCAGTAGTCAATCATGAG	this study	PR2254
SalI-Ubx2_rv	GATCGTCGACTTGTTCTTCATTTTCCTCATC	this study	PR2255
Ubx2ΔUBA_fw	CACCTTTCGCACACAGAATGGAAAGGCGAACC AGACC	this study	PR2256
Ubx2ΔUBA_rv	GGTCTGGTTCGCCTTTCATTCTGTGTGCGAAA GGTG	this study	PR2257
Ubx2ΔUBX_fw	CAGCCGTTTCGAGACAACGGCACTGGATGAAG AAGATG	this study	PR2258
Ubx2ΔUBX_rv	CATCTTCTTCATCCAGTGCCGTTGTCTCGAACG GCTG	this study	PR2259
Ubx2ΔMTS_fw	CGTCTACACCCATGGCTGAGAGACCATTGCCA GCGAACTTCC	this study	PR2260

Ubx2ΔMTS_rv	GGAAGTTCGCTGGCAATGGTCTCTCAGCCATG GGTGTAGACG	this study	PR2261
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Supplementary Data Table 4 | Antisera/antibodies used in this study.

Antigen	Source	No.
Ubx2	this study	GR1484-6
Cdc48	this study	GR5015-4
Ufd1	this study	GR5059-6 (affinity purified)
Hrd1	this study	GR5067-4
Doa10	this study	GR5065-4
Msp1	this study	GR1468-7
Tom20	Ref. 32	GR3225-7
Tom22	Ref. 32	GR3227-3
Tom40	Ref. 32	GR168-5
Tom5	Ref. 32	GR3419-7
Tom70	Ref. 32	GR657-3
Sam50	Ref. 32	GR312-17 (affinity purified)
Mim1	Ref. 32	GR544-1
Om14	Ref. 32	GR3041-4
Om45	Ref. 32	GR1311-4
Porin	this study	GR3622-4
Pam17	this study	GR3885-5
Tim23	Ref. 38	133-14
Tim44	Ref. 38	GR1835-4
Tim50	Ref. 38	GR 3881-4
Rip1	Ref. 38	GR542-3
Cox4	Ref. 38	GR578-5
Mdj1	Ref. 48	121-7
Sod2 precursor form	Ref. 49	GR3409-4
Sec61	Ref. 50	GR759-5
Sss1	Ref. 50	GR787-6
Ssa1	Ref. 34	GR1011-4
Pgk1	Ref. 34	GR753-4
Cox1	Ref. 38	GR1539-4

Cyt1	Ref. 38	GR540-3
Cox2	Ref. 38	GR1948-4
HA	Roche	Cat. No.11583816001
Ubiquitin	Santa Cruz	sc-8017
DHFR	Santa Cruz	sc-377091

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