

Supporting material

Supplementary figure legends

Supplementary Figure 1. Functional complementation of Pcp1 by the human mitochondrial rhomboid protease PARL. Wild type (WT) or $\Delta pcp1$ (Δ) spores expressing either the human (*PARL*) or the yeast (*PCP1*) mitochondrial rhomboid protease were used. Growth of indicated strains was tested by drop dilutions on rich media containing indicated carbon sources.

Supporting tables

Supplementary Table 1. Plasmids used in this study.

Name	Description	Vector	Reference
pYES2-OPA1 (URA3)	OPA1 cDNA expression constructs for splice variants 4, 7 or 8	pYES2 (Invitrogen)	this study
pYES2-OPA1 (<i>ura3::TRP1</i>)	OPA1 cDNA expression constructs for splice variants 4, 7 or 8	pYES2 (Invitrogen)	this study
pCAG-OPA1-SP7-IRES-mDsRed	OPA1 splice variant 7 cDNA for overexpression in mammalian cells	pCAG-IRES	this study
pES425-PARL	PARL cDNA expression construct	pES425#1 (Doron Rapaport)	this study
pYES2-Pcp1	Pcp1 expression construct	pYES2 (Invitrogen)	this study
pVT100U-mtGFP	Fluorescence microscopy	pVT100U	(Westermann and Neupert, 2000)
pRS314 ^{ADHI} -Yta10 (1-61)-hAFG3L2 (36-798)-Myc	expression construct for human AFG3L2	pRS314 (Sikorski and Hieter, 1989)	this study
Yeplac111 ^{ADHI} -Yta10 (1-63)-hparaplegin (59-795)-HA,	expression construct for human paraplegin	Yeplac111 (Gietz and Sugino, 1988)	(Atorino <i>et al.</i> , 2003)
Yeplac112 ^{ADHI} -Yta10 (1-61)-hAFG3L2 (36-798)-Myc	expression construct for human AFG3L2	Yeplac112 (Gietz and Sugino, 1988)	(Koppen <i>et al.</i> , 2007)
Yeplac112 ^{ADHI} -Yta10 (1-61)-hAFG3L2 ^{E575Q} (36-798)-Myc	expression construct for proteolytically inactive hAFG3L2	Yeplac112 (Gietz and Sugino, 1988)	(Koppen <i>et al.</i> , 2007)
Yeplac181 ^{YTA10} -Yta10 (1-61)-paraplegin (44-781)	expression construct for murine paraplegin	Yeplac181 (Gietz and Sugino, 1988)	(Nolden <i>et al.</i> , 2005)
Yeplac181 ^{YTA10} -Yta10 (1-61)-paraplegin ^{E575Q} (44-781)	expression construct for proteolytically inactive paraplegin	Yeplac181 (Gietz and Sugino, 1988)	(Koppen <i>et al.</i> , 2007)
Yeplac195 ^{YTA10} -Yta10 (1-61)-Afg31I (25-789)-Myc	expression construct for murine Afg31I	Yeplac195(Gietz and Sugino, 1988)	(Koppen <i>et al.</i> , 2007)
Yeplac195 ^{YTA10} -Yta10 (1-61)-Afg31I ^{E567Q} (25-789)-Myc	expression construct for proteolytically inactive Afg31I	Yeplac195 (Gietz and Sugino, 1988)	(Koppen <i>et al.</i> , 2007)
Yeplac112 ^{YTA10} -Yta10 (1-61)-Afg312 (36-802)-HA	expression construct for murine Afg312	Yeplac112 (Gietz and Sugino, 1988)	(Nolden <i>et al.</i> , 2005)
Yeplac112 ^{YTA10} -Yta10 (1-61)-Afg312 ^{E574Q} (36-802)-HA	expression construct for proteolytically inactive Afg312	Yeplac112(Gietz and Sugino, 1988)	(Koppen <i>et al.</i> , 2007)

Supplementary Table 2. Strains. The following strains were transformed with plasmids encoding OPA1 splice variants as indicated in the text and figures.

Description	Name	Background	Plasmids	Reference
WT		W303a		(Rothstein and Sherman, 1980)
<i>PCP1/Δpcp1</i>		W303a		Euroscarf acc. No. Y24731
<i>Δpcp1 + PCP1</i>	YKO100	spore of <i>PCP1/Δpcp1</i>	pYES2-Pcp1	this study
<i>PCP1</i>	YKO100	spore of <i>PCP1/Δpcp1</i>		this study
<i>Δpcp1 + PARL</i>	YKO100	spore of <i>PCP1/Δpcp1</i>	pES425-PARL	this study
<i>Δpcp1</i>	YKO100	spore of <i>PCP1/Δpcp1</i>		this study
<i>Ayta10Ayta12</i>	YKO100	W303a		(Atorino <i>et al.</i> , 2003)
<i>Ayta10Ayta12+ PARL</i>	YKO100	YKO100	pES425-PARL	this study
<i>Ayta10Ayta12 + hAFG3L2 + hparaplegin(LEU2)</i>	YKO117	W303a	Yeplac111 ^{ADH1} -Yta10 (1-63)-hparaplegin (59-795)-HA, pRS316 ^{ADH1} -Yta10 (1-61)-hAFG3L2 (36-798)-Myc	(Atorino <i>et al.</i> , 2003)
<i>Ayta10Ayta12 + hAFG3L2 + hparaplegin(TRP1)</i>	YKO117	YKO117	Yeplac111 ^{ADH1} -Yta10 (1-63)-hparaplegin (59-795)-HA, pRS314 ^{ADH1} -Yta10 (1-61)-hAFG3L2 (36-798)-Myc	this study
<i>Ayta10Ayta12</i>	YKO200	W303a		(Koppen <i>et al.</i> , 2007)
<i>Ayta10Ayta12 + hAFG3L2</i>	YKO203	YKO200	Yeplac112 ^{ADH1} -Yta10 (1-61)-hAFG3L2 (36-798)-Myc	(Koppen <i>et al.</i> , 2007)
<i>Ayta10Ayta12 + hAFG3L2^{EQ}</i>	YKO204	YKO200	Yeplac112 ^{ADH1} -Yta10 (1-61)-hAFG3L2 ^{E575Q} (36-798)-Myc	(Koppen <i>et al.</i> , 2007)
<i>Ayta10Ayta12 + paraplegin</i>	YKO209	YKO200	Yeplac181 ^{YTA10} -Yta10 (1-61)-paraplegin (44-781)	(Koppen <i>et al.</i> , 2007)
<i>Ayta10Ayta12 + Afg311</i>	YKO211	YKO200	Yeplac195 ^{YTA10} -Yta10 (1-61)-Afg311 (25-789)-Myc	(Koppen <i>et al.</i> , 2007)
<i>Ayta10Ayta12 + Afg311^{EQ}</i>	YKO212	YKO200	Yeplac195 ^{YTA10} -Yta10 (1-61)-Afg311 ^{E567Q} (25-789)-Myc	(Koppen <i>et al.</i> , 2007)
<i>Ayta10Ayta12 + Afg312</i>	YKO213	YKO200	Yeplac112 ^{YTA10} -Yta10 (1-61)-Afg312 (36-802)-HA	(Koppen <i>et al.</i> , 2007)
<i>Ayta10Ayta12 + Afg312^{EQ}</i>	YKO214	YKO200	Yeplac112 ^{YTA10} -Yta10 (1-61)-Afg312 ^{E574Q} (36-802)-HA	(Koppen <i>et al.</i> , 2007)
<i>Ayta10Ayta12 + paraplegin + Afg311^{EQ}</i>	YKO217	YKO200	Yeplac181 ^{YTA10} -Yta10 (1-61)-paraplegin (44-781), Yeplac195 ^{YTA10} -Yta10 (1-61)-Afg311 ^{E567Q} (25-789)-Myc	(Koppen <i>et al.</i> , 2007)
<i>Ayta10Ayta12 + paraplegin^{EQ} + Afg311^{EQ}</i>	YKO218	YKO200	Yeplac181 ^{YTA10} -Yta10 (1-61)-paraplegin ^{E575Q} (44-781), Yeplac195 ^{YTA10} -Yta10 (1-61)-Afg311 ^{E567Q} (25-789)-Myc	(Koppen <i>et al.</i> , 2007)
<i>Ayta10Ayta12 + paraplegin + Afg312^{EQ}</i>	YKO221	YKO200	Yeplac181 ^{YTA10} -Yta10 (1-61)-paraplegin (44-781), Yeplac112 ^{YTA10} -Yta10 (1-61)-Afg312 ^{E574Q} (36-802)-HA	(Koppen <i>et al.</i> , 2007)
<i>Ayta10Ayta12 + paraplegin^{EQ} + Afg312^{EQ}</i>	YKO222	YKO200	Yeplac181 ^{YTA10} -Yta10 (1-61)-paraplegin ^{E575Q} (44-781), Yeplac112 ^{YTA10} -Yta10 (1-61)-Afg312 ^{E574Q} (36-802)-HA	(Koppen <i>et al.</i> , 2007)

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