Supplemental tables

Table S1. Efficiency of site-specific photocrosslinking *in vivo,* related to Figure 4.

Photo-crosslinking was performed with probes at positions at which maximum crosslinking was observed for the various components. The percentage of sCPY*-DHFR-HA crosslinked was estimated after immunoprecipitation with Myc antibodies followed by immunoblotting with HA antibodies. Serial dilution of the extract were loaded on the same gel and served as reference. The percentage of the ERAD components and of Sec61p crosslinked to substrate was estimated from reciprocal immunoprecipitation experiments.

Percentage of sCPY*-DHFR-HA crosslinked to	
Hrd1p	.06-.3
Hrd3p	0.1
Der1p	.01-.02
Sec61p	not detectable

Percentage of component crosslinked to sCPY*-DHFR-HA	
Hrd1p	.05-.1
Hrd3p	0.1
Der1p	0.02
Sec61p	0.0005

Table S2. Yeast strains used in this study

Strain	Genotype	Source
BY4741	*MATa ura3Δ0 his3Δ1 leu2Δ0 met15Δ0*	OpenBiosystems
BY4742	*MATα ura3Δ0 his3Δ1 leu2Δ0 lys2Δ0*	OpenBiosystems
FY250	*MATα ura3-52 leu2Δ1 trp1Δ63 his3Δ200*	Winston et al., 1995
FY251	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200*	Winston et al., 1995
yPC1659	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 hrd1::KANR npl4-1*	This study
yPC1664	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 hrd1::KANR cdc48-3*	This study
yPC1790	*MAT a ura3D0 his3D1 leu2D0 met15D0 usa1::KANR*	Carvalho et al., 2006
yPC2247	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 hrd1::HYGB*	This study
yPC2329	*MATα ura3Δ0 his3Δ1 leu2Δ0 lys2Δ0 HIS-GALp-6his-Hrd1*	This study
yPC2339	*MATα ura3Δ0 his3Δ1 leu2Δ0 lys2Δ0 HIS-GALp-6his-Hrd1 hrd3::KANR*	This study
yPC2357	*MATα ura3Δ0 his3Δ1 leu2Δ0 lys2Δ0 HIS-GALp-6his-Hrd1 hrd3::KANR der1::HYGB*	This study
yPC2382	*MATα ura3Δ0 his3Δ1 leu2Δ0 lys2Δ0 HIS-GALp-6his-Hrd1 usa1::HYGB*	This study
yPC2383	*MATα ura3Δ0 his3Δ1 leu2Δ0 lys2Δ0 HIS-GALp-6his-Hrd1 der1::HYGB*	This study
yPC2438	*MATa ura3Δ0 his3Δ1 leu2Δ0 HIS-GALp-6his-Hrd1 hrd3::KANR usa1::NAT der1::HYGB*	This study
yPC2476	*MATα ura3Δ0 his3Δ1 leu2Δ0 lys2Δ0 HIS-GALp-6his-Hrd1 ubc7::KANR*	This study
yPC2610	*MATa ura3Δ0 his3Δ1 leu2Δ0 met15Δ0 Hrd1-13myc-KANR usa1::HYGB*	This study
yPC3098	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 Hrd1-13myc-KANR*	This study
yPC3129	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 Hrd1-13myc-KANR cdc48-3*	This study
yPC3176	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 mini-hrd1*	This study
yPC3184	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 Hrd1-13myc-HIS ubc7::KANR*	This study
yPC3186	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 mini-Hrd1-13myc-KANR*	This study

yPC3338	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 Hrd1-13myc-KANR der1::HYGB*	This study
yPC3339	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 Hrd1-13myc-KANR usa1::HIS*	This study
yPC3340	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 Hrd1-13myc-KANR yos9::HYGB*	This study
yPC3377	*MATα ura3-52 leu2Δ1 trp1Δ63 his3Δ200 Hrd1-13myc-KANR der1::HIS yos9::HYGB*	This study
yPC3381	*MATα ura3-52 leu2Δ1 trp1Δ63 his3Δ200 Hrd1-13myc-KANR usa1::HIS yos9::HYGB*	This study
yPC3390	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 hrd1::HYGB usa1::NAT*	This study
yPC3430	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 Hrd1-13myc-NAT hrd3::KANR GAPDH-HRD3::LEU2*	This study
yPC3431	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 Hrd1-13myc-NAT hrd3::KANR GAPDH-HRD3(357-833)::LEU2*	This study
yPC3432	*MATa ura3-52 leu2Δ1 trp1Δ63 his3Δ200 Hrd1-13myc-NAT hrd3::KANR GAPDH-HRD3(1-767)::LEU2*	This study
yPC3515	*MATα ura3-52 leu2Δ1 trp1Δ63 his3Δ200 Hrd1-13myc-KANR der1::HIS usa1::NAT*	This study

Table S3. Plasmids used in this study

Plasmid	Description	Source
pDN431	*CPY*-HA, CEN, URA*	Ng et al., 2000
pSM70	*KHN-HA, CEN, URA*	Vashist and Ng, 2004
pESC-Bpa	*TyrRS-tRNACUA* for pBpa incorporation	Chin et al., 2003
pPC234	*USA1, CEN, HIS*	This study
pPC254	*Usa1-HA, CEN, lEU*	Carvalho et al., 2006
pPC289	*FLAG-Usa1, CEN, HIS*	This study
pPC312	*Gal-Hrd1, CEN, LEU*	This study
pPC321	*Gal-Hrd1C399S, CEN, LEU*	This study
pPC364	*Hrd1-13Myc, CEN, HIS*	This study
pPC430	*Hrd1C399S-13MYC, CEN, HIS*	This study
pPC448	*Hrd1C399S-6his-3HA, CEN, LEU*	This study
pPC449	*Hrd1-6his-3HA, CEN, LEU*	This study
pPC490	*sCPY*-DHFR-HA, CEN, URA*	This study
pPC628	*FLAG-Usa1(D319-524), CEN, HIS*	This study
pPC633	*FLAG-Usa1(D319-371), CEN, HIS*	This study
pPC634	*FLAG-Usa1(D319-418), CEN, HIS*	This study
pPC635	*FLAG-Usa1(D491-524), CEN, HIS*	This study
pPC636	*FLAG-Usa1(D437-490), CEN, HIS*	This study
pPC644	*Usa1(D319-371), CEN, HIS*	This study
pPC645	*Usa1(D319-418), CEN, HIS*	This study
pPC646	*Usa1(D437-490), CEN, HIS*	This study