SI Appendix 3

Results of synthetic lethality experiments for highly connected proteins:

Hub	Paralog	genetic interaction	control	genetic interaction	paralog 2	genetic interaction	paralog 3	genetic interaction
YJL138C	YKR059W	SL	YDL188C	N. SSL				
YIL035C	YOR061W	SL	YDR293C	N. SSL				
YBL039C	YJR103W	SL	YIL114C	N. SSL				
YDL226C	YER122C	SL	YKR028W	N. SSL	YNL204C	N. SSL		
YNL055C	YIL114C	SL	YDR277C	N. SSL				
YMR105C	YKL127W	SL on Galactose	YIL114C	N. SSL				
YER081W	YIL074C	SL w/o serin	YOR061W	N. SSL	YOR388C	N. SSL		
YDL188C	YDL134C	SS	YER122C	N. SSL				
YJL098W	YKR028W	N. SSL	YKL127W	N. SSL	YFR040W	N. SSL	YGL229C	N. SSL
YOR047C	YDR277C	N. SSL	YKRO59W	N. SSL				
YOR136W	YNL037C	N. SSL	YJR103W	N. SSL	YILO94C	N. SSL		
YJR091C	YPR042C	N. SSL	YDL134C	N. SSL				
YDL047W	YDL188C	ND						

YDL160C	YDR293C	ND						

Results of synthetic lethality experiments for lowly connected proteins:

Paralog 1	Paralog 2	Genetic interaction
YBR015C	YJL186W	N. SSL
YHR021C	YKL156W	N. SSL
YJL134W	YKR053C	N. SSL
YGL089C	YPL187W	N. SSL
YBR005W	YDR003W	N. SSL
YEL029C	YNR027W	N. SSL
YFL053W	YML070W	N. SSL
YOR237W	YPL145C	N. SSL
YLR287C-A	YOR182C	SL
YDR185C	YLR168C	N. SSL
YER037W	YGL224C	N. SSL
YIL089W	YLR036C	N. SSL
YIR031C	YNL117W	N. SSL
YKL133C	YMR115W	N. SSL
YJR127C	YML081W	N. SSL
YHR162W	YGR243W	N. SSL
YLR130C	YGL255W	SS

Paralog 1	Paralog 2	Genetic interaction
YLR351C	YJL126W	N. SSL

SL, synthetic lethal; SS, synthetic sick; N.SSL, no interaction; ND, not determined.

Literature examination of highly connected proteins and their duplicate Partners.

HUB	Paralog	Function (HUB)	Function (paralog)	Literature indication for redundancy
YJL138C (TIF2)	YKR059W (TIF1)	Translation initiation factor eIF4A, identical to Tif1p; DEA(D/H)-box RNA helicase that couples ATPase activity to RNA binding and unwinding; forms a dumbbell structure of two compact domains connected by a linker; interacts with eIF4G	Translation initiation factor eIF4A, identical to Tif2p; DEA(D/H)-box RNA helicase that couples ATPase activity to RNA binding and unwinding; forms a dumbbell structure of two compact domains connected by a linker; interacts with eIF4G	Redundancy established by PMID: 2648398 *tif1tif2* double mutant is inviable.
YIL035C (CKA1)	YOR061W (CKA2)	Alpha catalytic subunit of casein kinase 2, a Ser/Thr protein kinase with roles in cell growth and proliferation; the holoenzyme also contains CKA2, CKB1 and CKB2, the many substrates include	Alpha' catalytic subunit of casein kinase 2, a Ser/Thr protein kinase with roles in cell growth and proliferation; the holoenzyme also contains CKA1, CKB1 and CKB2, the many substrates include transcription factors and all RNA polymerases	Redundancy established by PMID: 2196445 *cka1cka2* double mutant is inviable. lethality rescue by *Drosophila* casein kinase 2

HUB	Paralog	Function (HUB)	Function (paralog)	Literature indication for redundancy
		transcription factors and all RNA polymerases		
YBL039C (URA7)	YJR103W (URA8)	Major CTP synthase isozyme (see also *URA8*), catalyzes the ATP-dependent transfer of the amide nitrogen from glutamine to UTP, forming CTP, the final step in de novo biosynthesis of pyrimidines; involved in phospholipid biosynthesis	Minor CTP synthase isozyme (see also *URA7*), catalyzes the ATP-dependent transfer of the amide nitrogen from glutamine to UTP, forming CTP, the final step in de novo biosynthesis of pyrimidines; involved in phospholipid biosynthesis	Redundancy established by PMID: 8121398 The two CTP synthetases, Ura7p and Ura8p, share 78% amino acid identity and are functionally overlapping. Null mutations in either gene decrease intracellular levels of CTP, leading to a reduced growth rate, while deletion of both gene products results in lethality. Ura7p is also responsible for the majority of CTP synthesis (78%) and the difference in activity between the two isoforms is due to differential regulation in addition to their differential expression.
YDL226C (GCS1)	YER122C (GLO3)	ADP-ribosylation factor GTPase activating protein (*ARF GAP*), involved in ER-Golgi transport; shares functional similarity with Glo3p	ADP-ribosylation factor GTPase activating protein (*ARF GAP*), involved in ER-Golgi transport; shares functional similarity with Gcs1p	Redundancy established by PMID: 9927415 *gcs1 glo3* double deletion is inviable. Genetic interactions with *BET1*, *BOS1*, and *SEC22*, which encode v-SNARES, implicate Gcs1p and Glo3p in transport between the ER and the Golgi.
YNL055C (POR1)	YIL114C (POR2)	Mitochondrial porin (voltage-dependent anion channel), outer membrane protein required for	Putative mitochondrial porin (voltage-dependent anion channel), related to Por1p but not required for	Redundancy established by PMID: 9435273, 9315631 *por1 por2* double deletion exhibits synthetic growth defect. *POR1* is

HUB	Paralog	Function (HUB)	Function (paralog)	Literature indication for redundancy
		the maintenance of mitochondrial osmotic stability and mitochondrial membrane permeability	mitochondrial membrane permeability or mitochondrial osmotic stability	redundant with *POR2* but redundancy is partial in the sense that *POR1* is significantly more efficient
YMR105C (PGM2)	YKL127W (PGM1)	Translation initiation factor eIF4A, identical to Tif1p; DEA(D/H)-box RNA helicase that couples ATPase activity to RNA binding and unwinding; forms a dumbbell structure of two compact domains connected by a linker; interacts with eIF4G	Phosphoglucomutase, minor isoform; catalyzes the conversion from glucose-1-phosphate to glucose-6-phosphate, which is a key step in hexose metabolism	Redundancy established by PMID: 8119301 *Saccharomyces cerevisiae* contains a major phosphoglucomutase isoform, Pgm2p, and a minor phosphoglucomutase isoform, Pgm1p. Pgm2p and Pgm1p functions are involved in glycolysis, the pentose phosphate shunt, and the metabolism of glycogen, trehalose, and galactose. Phosphoglucomutase is also required for the synthesis of N-linked glycoproteins, extracellular glycans, and UDP-glucose. Pgm2p accounts for approximately 80%-90% of all phosphoglucomutase activity in *S. cerevisiae*. *pgm1 pgm2* double null mutants are viable, but cannot use galactose as a sole carbon source, and accumulate lower levels of glycogen and trehalose than wild type.
YER081W (SER3)	YIL074C (SER3)	Alpha catalytic subunit of casein kinase 2, a Ser/Thr protein	3-phosphoglycerate dehydrogenase, catalyzes the first step in serine and	Redundancy established by PMID:12525494 *ser3 ser33* double deletion is lethal in the

HUB	Paralog	Function (HUB)	Function (paralog)	Literature indication for redundancy
		kinase with roles in cell growth and proliferation; the holoenzyme also contains CKA2, CKB1 and CKB2, the many substrates include transcription factors and all RNA polymerases	glycine biosynthesis; isozyme of Ser3p	absence of serine. *ser3 ser33* double mutant showed no phosphoglycerate dehydrogenase activity, indicating that only these two genes encode such an activity. The specific activity determined in the wild-type strain was equivalent to the sum of the activities found in the single mutants. The reduction of phosphoglycerate dehydrogenase activity was more pronounced in the *ser33* mutant, indicating that Ser33p is the major isoenzyme
YDL188C (PPH22)	YDL134C (PPH21)	Catalytic subunit of protein phosphatase 2A, functionally redundant with Pph21p; methylated at C terminus; forms alternate complexes with several regulatory subunits; involved in signal transduction and regulation of mitosis	Catalytic subunit of protein phosphatase 2A, functionally redundant with Pph22p; methylated at C terminus; forms alternate complexes with several regulatory subunits; involved in signal transduction and regulation of mitosis	Redundancy established by PMID: 1656215, 2176150 *PPH21* and *PPH22* encode highly similar proteins and show less than 10% amino acid sequence divergence from each other. While disruption of either *PPH* gene alone is without any major effect, the *pph21 pph22* double disruption is lethal. Measurement of type 2A protein phosphatase activity in yeast strains lacking one or other of the genes indicates that they account for most, if not all, protein phosphatase 2A activity in the cell.

HUB	Paralog	Function (HUB)	Function (paralog)	Literature indication for redundancy
YJL098W (SAP185)	YKR028W (SAP190)	Protein that forms a complex with the Sit4p protein phosphatase and is required for its function; member of a family of similar proteins including Sap4p, Sap155p, and Sap190p	Protein that forms a complex with the Sit4p protein phosphatase and is required for its function; member of a family of similar proteins including Sap4p, Sap155p, and Sap185p	Redundancy established by PMID: 8649382 Cells with deletions of both *SAP185* and *SAP190* had a moderate slow-growth-rate phenotype on YPD medium. This finding suggests that *SAP185* and *SAP190* have full or nearly full functional overlap and that their combined function is important when the cells are grown on YPD medium
YOR047C (STD1)	YDR277C (MTH1)	Protein involved in control of glucose-regulated gene expression; interacts with protein kinase Snf1p, glucose sensors Snf3p and Rgt2p, and TATA-binding protein Spt15p; acts as a regulator of the transcription factor Rgt1p	Negative regulator of the glucose-sensing signal transduction pathway, required for repression of transcription by Rgt1p; interacts with Rgt1p and the Snf3p and Rgt2p glucose sensors; phosphorylated by Yck1p, triggering Mth1p degradation	Redundancy established by PMID: 8114728 *MTH1* encode a protein 61% identical to *STD1*. Both are also homologous to chicken fimbrin, human plastin, and yeast *SAC6* over a 43-residue region. Deletion of *STD1* and *MTH1* together impaired derepression of invertase in response to glucose limitation and sporulation. These findings confirm that the two genes are functionally related and that mutations in both genes together confer phenotypes similar to those conferred by *snfl* and *snf4.*
YOR136W (IDH2)	YNL037C (IDH1)	Subunit of mitochondrial NAD(+)-dependent isocitrate dehydrogenase, which catalyzes	Subunit of mitochondrial NAD(+)-dependent isocitrate dehydrogenase, which catalyzes the oxidation of	

HUB	Paralog	Function (HUB)	Function (paralog)	Literature indication for redundancy
		the oxidation of isocitrate to alpha-ketoglutarate in the TCA cycle	isocitrate to alpha-ketoglutarate in the TCA cycle	
YJR091C (JSN1)	YPR042C (PUF2)	Member of the Puf family of RNA-binding proteins, interacts with mRNAs encoding membrane-associated proteins; overexpression suppresses a tub2-150 mutation and causes increased sensitivity to benomyl in wild-type cells	Member of the PUF protein family, which is defined by the presence of Pumilio homology domains that confer RNA binding activity; preferentially binds mRNAs encoding membrane-associated proteins	
YDL047W (SIT4)	YDL188C (PPH22)	Type 2A-related serine-threonine phosphatase that functions in the G1/S transition of the mitotic cycle; cytoplasmic and nuclear protein that modulates functions mediated by Pkc1p including cell wall and actin cytoskeleton organization	Catalytic subunit of protein phosphatase 2A, functionally redundant with Pph21p; methylated at C terminus; forms alternate complexes with several regulatory subunits; involved in signal transduction and regulation of mitosis	PMID: 1848673 Elevated gene dosage of *PPH22* partially suppressed the *sit4-102* mutation. PMID: 10856229 The paper suggests a common function for Cdc55p and Pph21p/Pph22p/Pph3p, but not Sit4p**.**
YDL160C (DHH1)	YDR293C (SSD1)	Cytoplasmic DExD/H-box helicase, stimulates mRNA decapping, coordinates distinct steps in mRNA function and	Protein with a role in maintenance of cellular integrity, interacts with components of the TOR pathway; ssd1 mutant of a clinical S. cerevisiae strain	Redundancy established by PMID:10234786 Specific *ssd1* mutants are synthetic lethal in combination with the *dhh1* disruption. Furthermore, *DHH1* and *SSD1* could

HUB	Paralog	Function (HUB)	Function (paralog)	Literature indication for redundancy
		decay, interacts with both the decapping and deadenylase complexes, may have a role in mRNA export and translation Dhh1p has been suggested to play a role in partitioning mRNAs between translatable and nontranslatable pools, which has been implicated in the recovery from G1/S cell cycle arrest following DNA damage.	displays elevated virulence	functionally complement each other in the *ade2* red colour pigment formation, hypersensitivity to SDS, growth on synthetic media and at high temperature.

- Gene functions were taken from the gene "Description" category in SGD (http://www.yeastgenome.org/) and is based on community annotation and references provided thereof.

Literature examination of sparsely connected proteins and their duplicate Partners.

Paralog 1	Paralog 2	Function (paralog1)	Function (paralog 2)	Literature examination and indication for redundancy
YBR015C (MNN2)	YJL186W (MNN5)	Alpha-1,2-mannosyltransferase, responsible for addition of the first alpha-1,2-linked mannose to form the branches on the mannan backbone of oligosaccharides, localizes to an	Alpha-1,2-mannosyltransferase, responsible for addition of the second alpha-1,2-linked mannose of the branches on the mannan backbone of oligosaccharides, localizes to an early Golgi	

Paralog 1	Paralog 2	Function (paralog1)	Function (paralog 2)	Literature examination and indication for redundancy
		early Golgi compartment.	compartment	
YHR021C (RPS27B)	YKL156W (RPS27A)	Protein component of the small (40S) ribosomal subunit; nearly identical to Rps27Ap and has similarity to rat S27 ribosomal protein	Protein component of the small (40S) ribosomal subunit; nearly identical to Rps27Bp and has similarity to rat S27 ribosomal protein	
YJL134W (LCB3)	YKR053C (YSR3)	Long-chain base-1-phosphate phosphatase, regulates ceramide and long-chain base phosphates levels, involved in incorporation of exogenous long chain bases in sphingolipids	Dihydrosphingosine 1-phosphate phosphatase, membrane protein involved in sphingolipid metabolism; has similarity to Lcb3p	PMID:10477278 D*ysr2*D*ysr3* accumulated more DHS-1-P than either of the single-deletion
YGL089C (MF(ALPHA)2)	YPL187W (MF(ALPHA)1)	Mating pheromone alpha-factor, made by alpha cells; interacts with mating type a cells to induce cell cycle arrest and other responses leading to mating; also encoded by *MF(ALPHA)1*, which is more highly expressed than *MF(ALPHA)2*	Mating pheromone alpha-factor, made by alpha cells; interacts with mating type a cells to induce cell cycle arrest and other responses leading to mating; also encoded by *MF(ALPHA)2*, although *MF(ALPHA)1* produces most alpha-factor	
YBR005W (RCR1)	YDR003W (RCR2)	Protein of the ER membrane involved in cell wall chitin	Vacuolar protein that presumably functions within the endosomal-	

Paralog 1	Paralog 2	Function (paralog1)	Function (paralog 2)	Literature examination and indication for redundancy
		deposition; may function in the endosomal-vacuolar trafficking pathway, helping determine whether plasma membrane proteins are degraded or routed to the plasma membrane	vacuolar trafficking pathway, affecting events that determine whether plasma membrane proteins are degraded or routed to the plasma membrane; similar to Rcr1p	
YEL029C (BUD16)	YNR027W (BUD17)	Protein involved in bud-site selection and telomere maintenance; diploid mutants display a random budding pattern instead of the wild-type bipolar pattern; has similarity to pyridoxal kinases	Protein involved in bud-site selection; diploid mutants display a random budding pattern instead of the wild-type bipolar pattern	
YFL053W (*DAK2)*	YML070W (*DAK1)*	Dihydroxyacetone kinase, required for detoxification of dihydroxyacetone (DHA); involved in stress adaptation	Dihydroxyacetone kinase, required for detoxification of dihydroxyacetone (DHA); involved in stress adaptation	Redundancy established by PMID: 12401799 *dak1dak2* double deletion is highly sensitive to DHA (dihydroxyacetone) during saline growth
YOR237W (HES1)	YPL145C (KES1)	Protein implicated in the regulation of ergosterol biosynthesis; one of a seven member gene family with a common essential function and	Member of the oxysterol binding protein family, which includes seven yeast homologs; involved in negative regulation of Sec14p-dependent Golgi complex secretory functions,	

Paralog 1	Paralog 2	Function (paralog1)	Function (paralog 2)	Literature examination and indication for redundancy
		non-essential unique functions; similar to human oxysterol binding protein	peripheral membrane protein that localizes to the Golgi complex	
YLR287C-A (RPS30A)	YOR182C (RPS30B)	Protein component of the small (40S) ribosomal subunit; nearly identical to Rps30Bp and has similarity to rat S30 ribosomal protein	Protein component of the small (40S) ribosomal subunit; nearly identical to Rps30Ap and has similarity to rat S30 ribosomal protein	Redundancy established by PMID: 8662789 Deletion of the *RPS30A* gene is not lethal but confers a slow growth phenotype. Ribosomes in the mutant strains contain an authentic yrpS30 protein, indicating that a functional yrpS30 is expressed from the duplicated gene.
YDR185C	YLR168C (MSF1*)*	Mitochondrial protein of unknown function; has similarity to Ups1p, which is involved in regulation of alternative topogenesis of the dynamin-related GTPase Mgm1p	Putative protein of unknown function that may be involved in intramitochondrial sorting; has similarity to Ups1p and to human PRELI; the green fluorescent protein (GFP)-tagged protein localizes to mitochondria	
YER037W (PHM8)	YGL224C (SDT1)	Protein of unknown function, expression is induced by low phosphate levels and by inactivation of Pho85p	Pyrimidine nucleotidase; overexpression suppresses the 6-AU sensitivity of transcription elongation factor S-II, as well as resistance to other pyrimidine derivatives	
YIL089W	YLR036C	Putative protein of unknown	Putative protein of unknown function	

Paralog 1	Paralog 2	Function (paralog1)	Function (paralog 2)	Literature examination and indication for redundancy
		function	predicted to have transmembrane domains; interacts with HSP90 by yeast two-hybrid analysis; *YLR036C* is not an essential protein	
YIR031C (DAL7)	YNL117W (MLS1)	Malate synthase, role in allantoin degradation unknown; expression sensitive to nitrogen catabolite repression and induced by allophanate, an intermediate in allantoin degradation	Malate synthase, enzyme of the glyoxylate cycle, involved in utilization of non-fermentable carbon sources; expression is subject to carbon catabolite repression; localizes in peroxisomes during growth in oleic acid medium	
YKL133C	YMR115W (FMP24)	Putative protein of unknown function	Putative protein of unknown function; the authentic, non-tagged protein is detected in highly purified mitochondria in high-throughput studies	
YJR127C (RSF2)	YML081W	Zinc-finger protein involved in transcriptional control of both nuclear and mitochondrial genes, many of which specify products required for glycerol-based growth, respiration, and	Putative protein of unknown function; green fluorescent protein (GFP)-fusion protein localizes to the nucleus; *YML081W* is not an essential gene	

Paralog 1	Paralog 2	Function (paralog1)	Function (paralog 2)	Literature examination and indication for redundancy
		other functions		
YHR162W	YGR243W (FMP43)	Putative protein of unknown function; green fluorescent protein (GFP)-fusion protein localizes to the mitochondrion	Putative protein of unknown function; the authentic, non-tagged protein is detected in highly purified mitochondria in high-throughput studies	
YLR130C (ZRT2)	YGL255W (ZRT1)	Low-affinity zinc transporter of the plasma membrane; transcription is induced under low-zinc conditions by the Zap1p transcription factor	High-affinity zinc transporter of the plasma membrane, responsible for the majority of zinc uptake; transcription is induced under low-zinc conditions by the Zap1p transcription factor	PMID: 8798516 Over expression of *ZRT2* restored the ability of the *zrt1* mutant to grow under moderately zinc-limiting conditions, but not on severely zinc-limited media. The regulatory pool of intracellular zinc is at a lower level in the *zrt1zrt2* strain grown under than in the *zrt1* single mutant. Disrupting *zrt2* eliminated the low affinity uptake activity, but had little effect on the high affinity system (*ZRT1*). Therefore, the high and low affinity systems are separate uptake path-ways. *zrt1zrt2* mutant was viable ,indicating the existence of additional zinc uptake pathways.
YLR351C (NIT3)	YJL126W (NIT2)	Nit protein, one of two proteins in *S. cerevisiae* with similarity to the Nit domain of NitFhit from	Nit protein, one of two proteins in *S. cerevisiae* with similarity to the Nit domain of NitFhit from fly and	

Paralog 1	Paralog 2	Function (paralog1)	Function (paralog 2)	Literature examination and indication for redundancy
		fly and worm and to the mouse and human Nit protein which interacts with the Fhit tumor suppressor; nitrilase superfamily member	worm and to the mouse and human Nit protein which interacts with the Fhit tumor suppressor; nitrilase superfamily member	