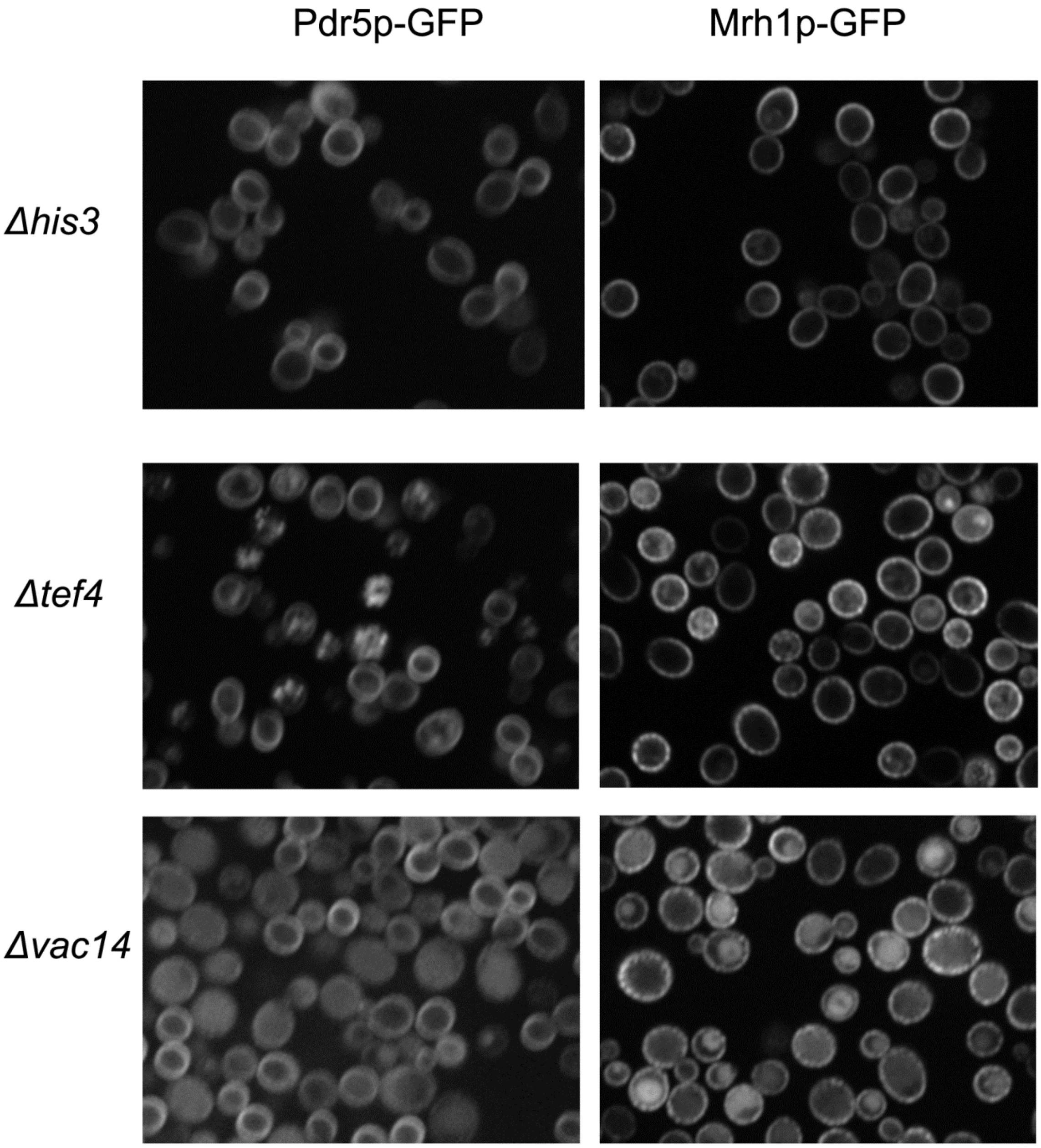
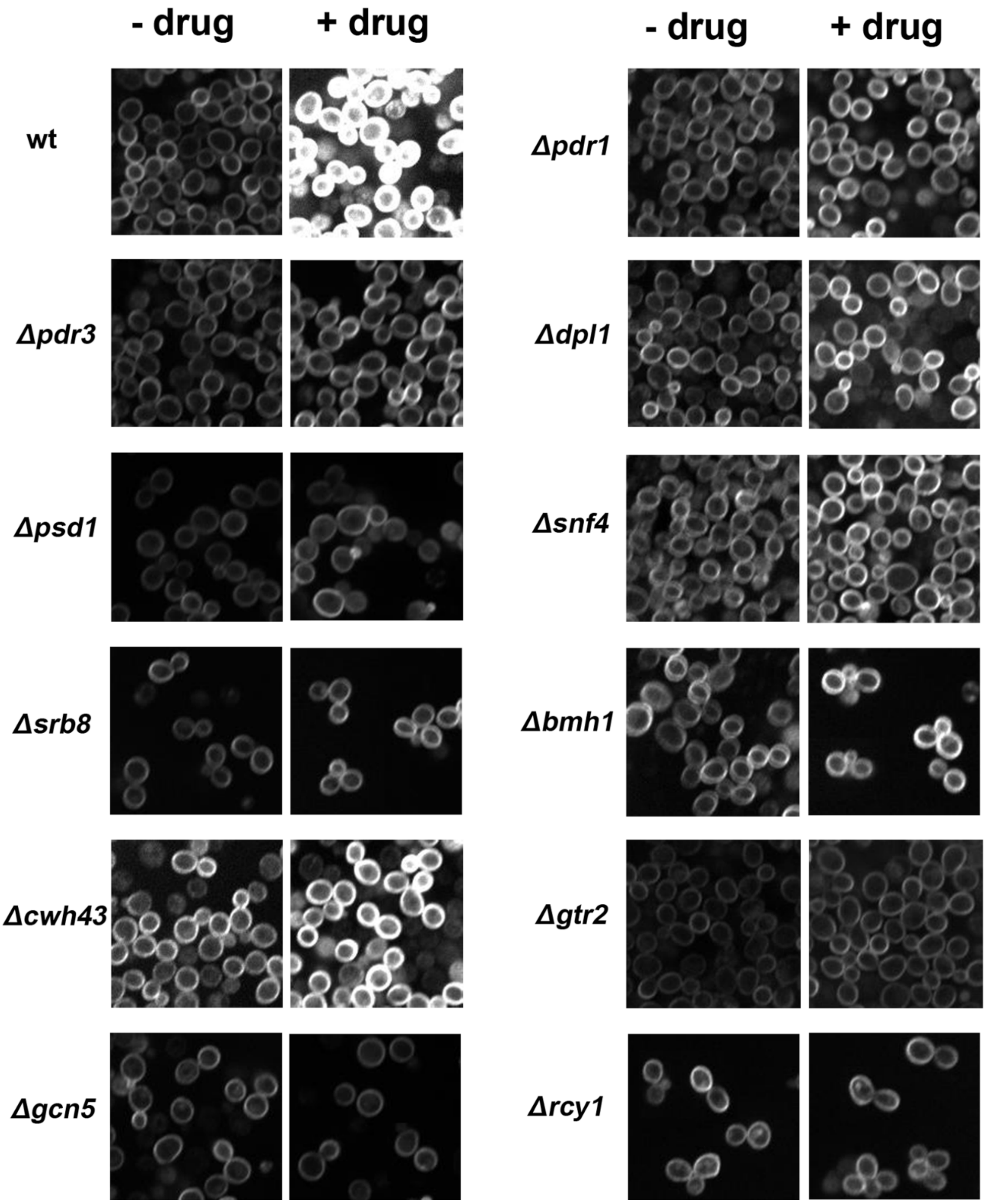
Supplementary Figures

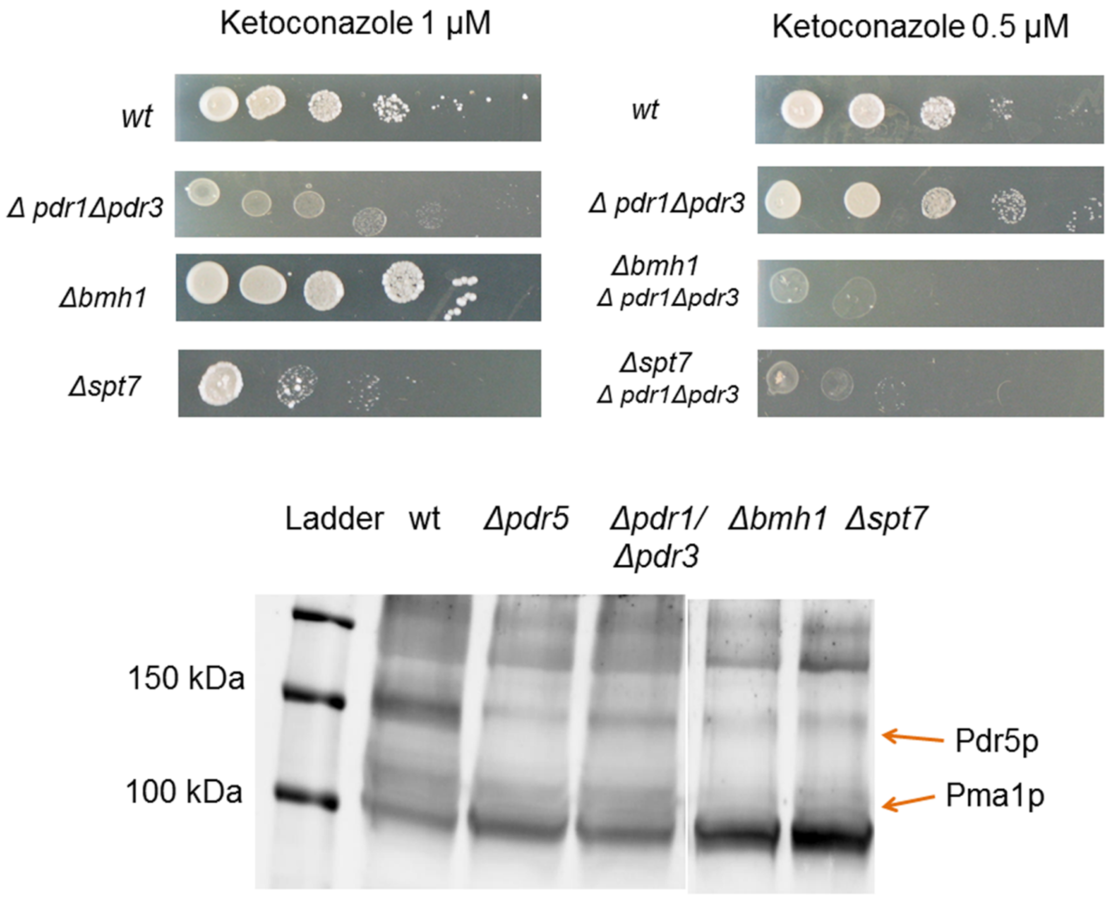


Supplementary Figure1: Examples of gene deletions which result in misfolding or mis-localisationn of the plasma membrane proteins Pdr5p and Mrh1p. Δhis3 is the “wild-type” positice control.

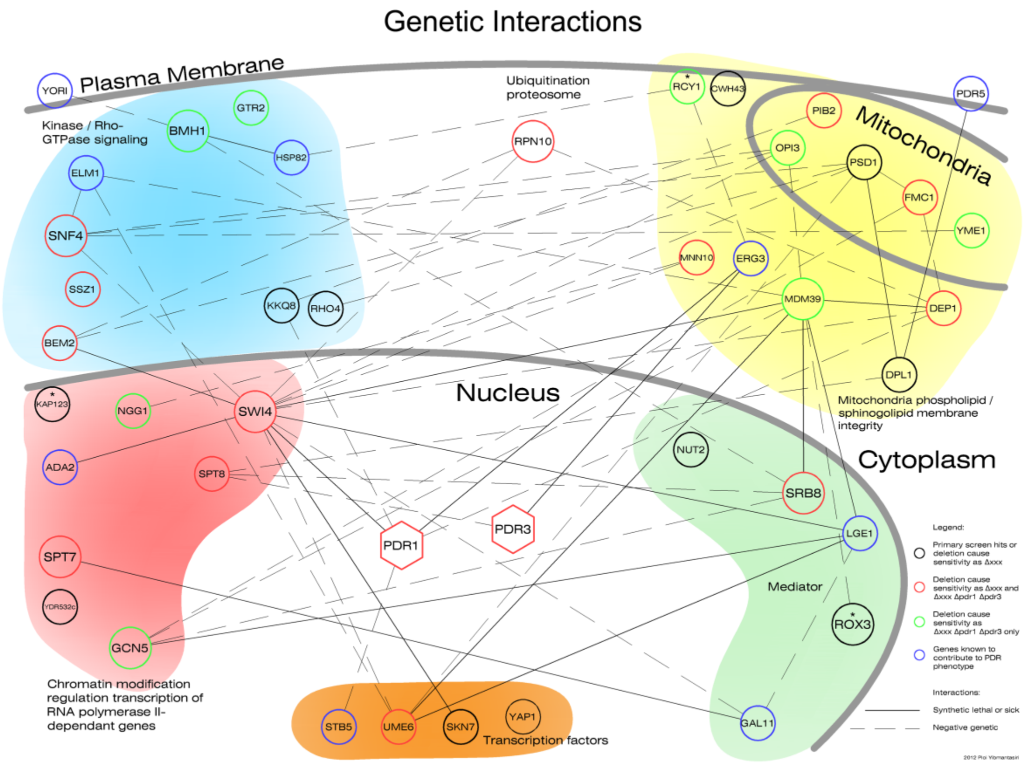


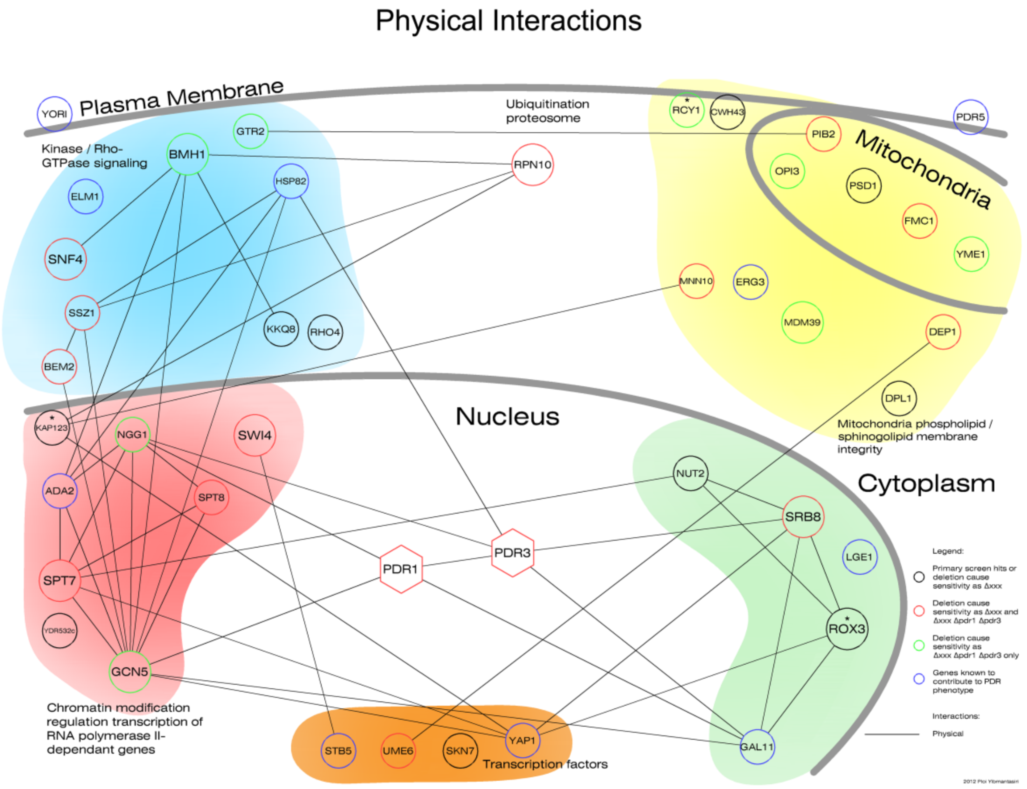
Supplementary Figure 2: Examples of gene deletions that prevented normal upregulation of Pdr5p-GFP and Yor1p-GFP reporters.

The images are of the Pdr5p-GFP reporter in the named gene deletion backgrounds. The images were adjusted so that fluorescence levels of DMSO control and drug treatment of each of the deletions (in 35μM atorvastatin) are directly comparable as to fluorescence intensity in each image filed.



Supplementary Figure 3: Examples of Spot Dilutions Assays and Western Blots. Top panel: Spot Dilution of different deletion strains in 1 and 0.5 µM ketoconazole , which allow *Δpdr1Δpdr3* to grow. Deletion of *SPT7* caused hypersensitivity to ketoconazole in both single deletion and in *Δpdr1Δpdr3* background. Deletion of *BMH1*caused hypersensitivity to ketoconazole in *Δpdr1Δpdr3* background alone. **Bottom panel:** Example of Western Blots of Pdr5p in different deletion backgrounds. Pma1p, another plasma membrane protein, was used as an internal control. It has two isoforms one of about the same size as Pdr5p and shows up as a background band after the Western Blot was exposed to both Pdr5p and Pma1p antibodies.





Supplementary Figure 4: The published genetic and physical interaction http://thebiogrid.org) networks among our screen results and some of the known PDR-contributing genes .

Supplementary Tables

Supplementary Table 1: Gene Deletions that overlap between PDR screens and Mrh1p-GFP control screen

Systematic Name	**Standard Name**	**Discription**
YJR033C	*RAV1*	Subunit of the RAVE complex ,which required for transport between the early and late endosome/PVC and for localization of TGN membrane proteins
YKL081W	*TEF4*	Gamma subunit of translational elongation factor eEF1B
YLR262C	*YPT6*	Rab family GTPase, Ras-like GTP binding protein involved in the secretory pathway; required for fusion of endosome-derived vesicles with the late Golgi
YLR386W	*VAC14*	Protein involved in regulated synthesis of PtdIns(3,5)P(2), in control of trafficking of some proteins to the vacuole lumen via the MVB
YPL154C	*PEP4*	Vacuolar aspartyl protease (proteinase A)

Supplementary Table 2: List of gene deletions which cause insignificant change in PDR reporters upon atorvastatin treatment

Systematic Name	**Standard Name**	**Description**
YDR374C		Putative protein of unknown function
YFR018C		Putative protein of unknown function
YIL152W		Putative protein of unknown function
YNL315C	ATP11	Molecular chaperone, required for the assembly of alpha and beta subunits into the F1 sector of mitochondrial F1F0 ATP synthase
YER155C	BEM2	Rho GTPase activating protein (RhoGAP) involved in the control of cytoskeleton organization and cellular morphogenesis; required for bud emergence
YER177W	BMH1	14-3-3 protein, major isoform; controls proteome at post-transcriptional level, binds proteins and DNA, involved in regulation of many processes including exocytosis, vesicle transport, Ras/MAPK signaling, and rapamycin-sensitive signalling
YOR061W	CKA2	Alpha' catalytic subunit of casein kinase 2 (CK2), a Ser/Thr protein kinase with roles in cell growth and proliferation; CK2, comprised of CKA1, CKA2, CKB1 and CKB2, has many substrates including transcription factors and all RNA polymerase
YDL155W	CLB3	B-type cyclin involved in cell cycle progression; activates Cdc28p to promote the G2/M transition; may be involved in DNA replication and spindle assembly; accumulates during S phase and G2, then targeted for ubiquitin-mediated degradation
YCR017C	CWH43	Putative sensor/transporter protein involved in cell wall biogenesis; contains 14-16 transmembrane segments and several putative glycosylation and phosphorylation sites; null mutation is synthetically lethal with pkc1 deletion
YGL078C	DBP3	Putative ATP-dependent RNA helicase of the DEAD-box family involved in ribosomal biogenesis
YDR294C	DPL1	Dihydrosphingosine phosphate lyase, regulates intracellular levels of sphingolipid long-chain base phosphates (LCBPs), degrades phosphorylated long chain bases, prefers C16 dihydrosphingosine-l-phosphate as a substrate

YGR071C	ENV11	Protein proposed to be involved in vacuolar functions; mutant shows defect in CPY processing and fragmented vacuoles; deletion mutant has increased glycogen accumulation and displays elongated buds; green fluorescent protein (GFP)-fusion protein localizes to the nucleus
YGL054C	ERV14	Protein localized to COPII-coated vesicles, involved in vesicle formation and incorporation of specific secretory cargo; required for the delivery of bud-site selection protein Axl2p to cell surface; related to Drosophila cornichon
YIL065C	FIS1	Protein involved in mitochondrial membrane fission and peroxisome abundance; required for localization of Dnm1p and Mdv1p during mitochondrial division; mediates ethanol-induced apoptosis and ethanol-induced mitochondrial fragmentation
YIL098C	FMC1	Mitochondrial matrix protein, required for assembly or stability at high temperature of the F1 sector of mitochondrial F1F0 ATP synthase; null mutant temperature sensitive growth on glycerol is suppressed by multicopy expression of Odc1p
YHR176W	FMO1	Flavin-containing monooxygenase, localized to the cytoplasmic face of the ER membrane; catalyzes oxidation of biological thiols to maintain the ER redox buffer ratio for correct folding of disulfide-bonded proteins
YGR252W	GCN5	Acetyltransferase, modifies N-terminal lysines on histones H2B and H3; acetylates Rsc4p, a subunit of the RSC chromatin-remodeling complex, altering replication stress tolerance; catalytic subunit of the ADA and SAGA histone acetyltransferase complexes; founding member of the Gcn5p-related N-acetyltransferase superfamily; mutant displays reduced transcription elongation in the G-less-based run-on (GLRO) assay
YGL020C	GET1 (MDM39)	Subunit of the GET complex; involved in insertion of proteins into the ER membrane; required for the retrieval of HDEL proteins from the Golgi to the ER in an ERD2 dependent fashion and for normal mitochondrial morphology and inheritance
YGR163W	GTR2	Putative GTP binding protein that negatively regulates Ran/Tc4 GTPase cycle; activates transcription; subunit of EGO and GSE complexes; required for sorting of Gap1p; localizes to cytoplasm and to chromatin; homolog of human RagC and RagD
YJR147W	HMS2	Protein with similarity to heat shock transcription factors; overexpression suppresses the pseudohyphal filamentation defect of a diploid mep1 mep2 homozygous null mutant
YFR038W	IRC5	Putative ATPase containing the DEAD/H helicase-related sequence motif; null mutant displays increased levels of spontaneous Rad52p foci
YER110C	KAP123	Karyopherin beta, mediates nuclear import of ribosomal proteins prior to assembly into ribosomes and import of histones H3 and H4; localizes to the nuclear pore, nucleus, and cytoplasm; exhibits genetic interactions with RAI1
YKL168C	KKQ8	Putative serine/threonine protein kinase with unknown cellular role
YDR532C	KRE28	Subunit of a kinetochore-microtubule binding complex with Spc105p that bridges centromeric heterochromatin and kinetochore MAPs and motors, and is also required for sister chromatid bi-orientation and kinetochore binding of SAC components
YCL061C	MRC1	S-phase checkpoint protein required for DNA replication; interacts with and stabilizes Pol2p at stalled replication forks during stress, where it forms a pausing complex with Tof1p and is phosphorylated by Mec1p; protects uncapped telomeres
YDR192C	NUP42	Subunit of the nuclear pore complex (NPC) that localizes exclusively to the cytoplasmic side; involved in RNA export, most likely at a terminal step; interacts with Gle1p
YJR073C	OPI3	Phospholipid methyltransferase (methylene-fatty-acyl-phospholipid synthase), catalyzes the last two steps in phosphatidylcholine biosynthesis
YGL013C	PDR1	Zinc cluster protein that is a master regulator involved in recruiting other zinc cluster proteins to pleiotropic drug response elements (PDREs) to fine tune the regulation of multidrug resistance genes

YBL005W	PDR3	Transcriptional activator of the pleiotropic drug resistance network, regulates expression of ATP-binding cassette (ABC) transporters through binding to cis-acting sites known as PDREs (PDR responsive elements); post-translationally upregulated in cells lacking a functional mitochondrial genome
YGL023C	PIB2	Protein binding phosphatidylinositol 3-phosphate, involved in telomere-proximal repression of gene expression; similar to Fab1 and Vps27
YBL051C	PIN4	Protein involved in G2/M phase progression and response to DNA damage, interacts with Rad53p; contains an RNA recognition motif, a nuclear localization signal, and several SQ/TQ cluster domains; hyperphosphorylated in response to DNA damage
YHR075C	PPE1	Protein with carboxyl methyl esterase activity that may have a role in demethylation of the phosphoprotein phosphatase catalytic subunit; also identified as a small subunit mitochondrial ribosomal protein
YNL169C	PSD1	Phosphatidylserine decarboxylase of the mitochondrial inner membrane, converts phosphatidylserine to phosphatidylethanolamine
YAL017W	PSK1	One of two (see also PSK2) PAS domain containing S/T protein kinases; coordinately regulates protein synthesis and carbohydrate metabolism and storage in response to a unknown metabolite that reflects nutritional status
YJL204C	RCY1	F-box protein involved in recycling plasma membrane proteins internalized by endocytosis; localized to sites of polarized growth
YKR055W	RHO4	Non-essential small GTPase of the Rho/Rac subfamily of Ras-like proteins, likely to be involved in the establishment of cell polarity
YDR279W	RNH202	Ribonuclease H2 subunit, required for RNase H2 activity; related to human AGS2 that causes Aicardi-Goutieres syndrome
YBL093C	ROX3	Subunit of the RNA polymerase II mediator complex; associates with core polymerase subunits to form the RNA polymerase II holoenzyme
YHR200W	RPN10	Non-ATPase base subunit of the 19S regulatory particle (RP) of the 26S proteasome; N-terminus plays a role in maintaining the structural integrity of the RP; binds selectively to polyubiquitin chains; homolog of the mammalian S5a protein
YJR127C	RSF2	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 other functions
YOL067C	RTG1	Transcription factor (bHLH) involved in interorganelle communication between mitochondria, peroxisomes, and nucleus
YGR143W	SKN1	Protein involved in sphingolipid biosynthesis; type II membrane protein with similarity to Kre6p
YHR206W	SKN7	Nuclear response regulator and transcription factor; physically interacts with the Tup1-Cyc8 complex and recruits Tup1p to its targets; part of a branched two-component signaling system; required for optimal induction of heat-shock genes in response to oxidative stress; involved in osmoregulation
YGL115W	SNF4	Activating gamma subunit of the AMP-activated Snf1p kinase complex (contains Snf1p and a Sip1p/Sip2p/Gal83p family member); activates glucose-repressed genes, represses glucose-induced genes; role in sporulation, and peroxisome biogenesis
YER161C	SPT2	Protein involved in negative regulation of transcription; required for RNA polyadenylation; exhibits regulated interactions with both histones and SWI-SNF components, has similarity to mammalian HMG1 proteins
YBR081C	SPT7	Subunit of the SAGA transcriptional regulatory complex, involved in proper assembly of the complex; also present as a C-terminally truncated form in the SLIK/SALSA transcriptional regulatory complex
YCR081W	SRB8	Subunit of the RNA polymerase II mediator complex; associates with core polymerase subunits to form the RNA polymerase II holoenzyme; essential for transcriptional regulation; involved in glucose repression
YDR443C	SSN2	Subunit of the RNA polymerase II mediator complex; associates with core polymerase subunits to form the RNA polymerase II holoenzyme; required for stable association of Srb10p-Srb11p kinase; essential for transcriptional regulation

YHR064C	SSZ1	Hsp70 protein that interacts with Zuo1p (a DnaJ homolog) to form a ribosome-associated complex that binds the ribosome via the Zuo1p subunit; also involved in pleiotropic drug resistance via sequential activation of PDR1 and PDR5; binds ATP
YOR027W	STI1	Hsp90 cochaperone, interacts with the Ssa group of the cytosolic Hsp70 chaperones and activates Ssa1p ATPase activity; interacts with Hsp90 chaperones and inhibits their ATPase activity; homolog of mammalian Hop
YER111C	SWI4	DNA binding component of the SBF complex (Swi4p-Swi6p), a transcriptional activator that in concert with MBF (Mbp1-Swi6p) regulates late G1-specific transcription of targets including cyclins and genes required for DNA synthesis and repair
YBR165W	UBS1	Ubiquitin-conjugating enzyme suppressor that functions as a general positive regulator of Cdc34p activity; nuclear protein that may represent a link between nucleocytoplasmic transport and ubiquitin ligase activity
YDR207C	UME6	Key transcriptional regulator of early meiotic genes, binds URS1 upstream regulatory sequence, couples metabolic responses to nutritional cues with initiation and progression of meiosis, forms complex with Ime1p, and also with Sin3p-Rpd3p
YPR024W	YME1	Catalytic subunit of the mitochondrial inner membrane i-AAA protease complex, which is responsible for degradation of unfolded or misfolded mitochondrial gene products; mutation causes an elevated rate of mitochondrial turnover

Supplementary Table 3: Eleven Genes relating to RNA transcription were GO enriched as revealed in the PDR primary screens.

GO-term	P-value	Gene(s) annotated to the term
sequence-specific DNA binding transcription factor activity	0.00588	PDR3/YBL005W: UME6/YDR207C: SWI4/YER111C: PDR1/YGL013C: SKN7/YHR206W: RSF2/YJR127C: RTG1/YOL067C
nucleic acid binding transcription factor activity	0.00613	PDR3/YBL005W UME6/YDR207C: SWI4/YER111C: PDR1/YGL013C: SKN7/YHR206W: RSF2/YJR127C: RTG1/YOL067C
transcription factor binding transcription factor activity	0.04211	ROX3/YBL093C: SRB8/YCR081W: UME6/YDR207C: SSN2/YDR443C: GCN5/YGR252W
transcription coactivator activity	0.04829	SRB8/YCR081W: SSN2/YDR443C: GCN5/YGR252W

Supplementary Table 4: LOPAC compounds that activate PDR reporters and inhibit signalling-related pathways or molecules

Compounds	Mode-of-action
Farnesylthiosalicylic acid	Non-toxic Ras inhibitor
3-alpha,21-dihydroxy-5-alpha- pregnan-20-one	Positive allosteric modulator of GABA-A receptors

Dequalinium analog, C-14 linker	Protein kinase C-alpha (PKC-alpha) inhibitor
N-phenylanthranilic acid	Cl- channel blocker
7-cyclopentyl-5-(4-phenoxy) phenyl-7H-pyrolo	Potent and selective Ick (src family tyrosine kinase) inhibitor.
DL-stearoylcarnitine chloride	Protein kinase C (PKC) inhibitor
SU 6656	Selective Src family kinase inhibitor
U0126	Specific inhibitor of MEK1 and MEK2 (MAP kinase kinase; MAPKK)