Supporting Information

Live-cell assays reveal selectivity and sensitivity of the multidrug response in budding yeast

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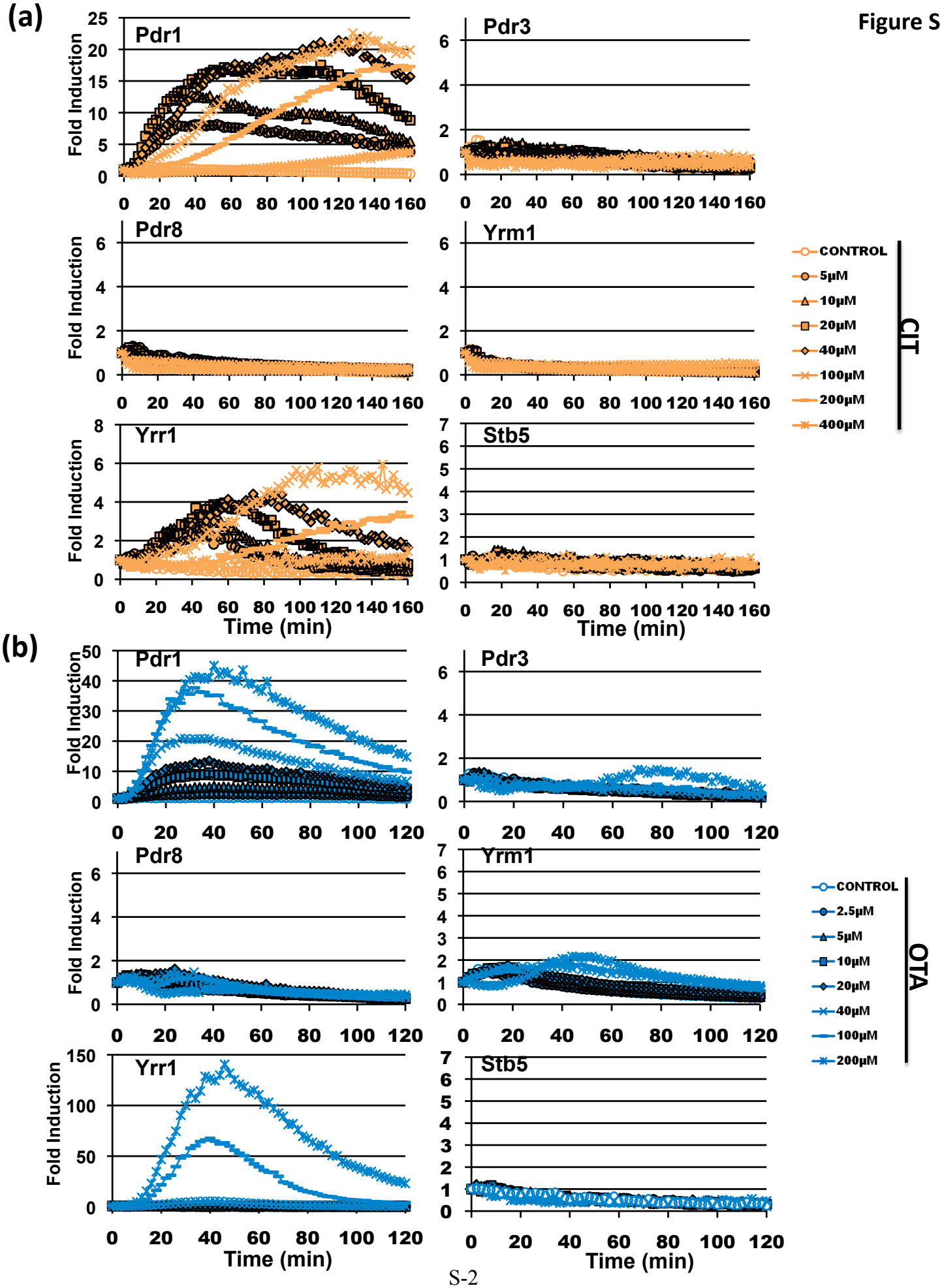
Figure S1


(a)
Pdr1
Pdr3
Pdr8
Yrm1
Yrr1
Stb5
Fold Induction
Time (min)
CONTROL
5μM
10μM
20μM
40μM
100μM
200μM
400μM
CIT
(b)
Pdr1
Pdr3
Pdr8
Yrm1
Yrr1
Stb5
Fold Induction
Time (min)
CONTROL
2.5μM
5μM
10μM
20μM
40μM
100μM
200μM
OTA

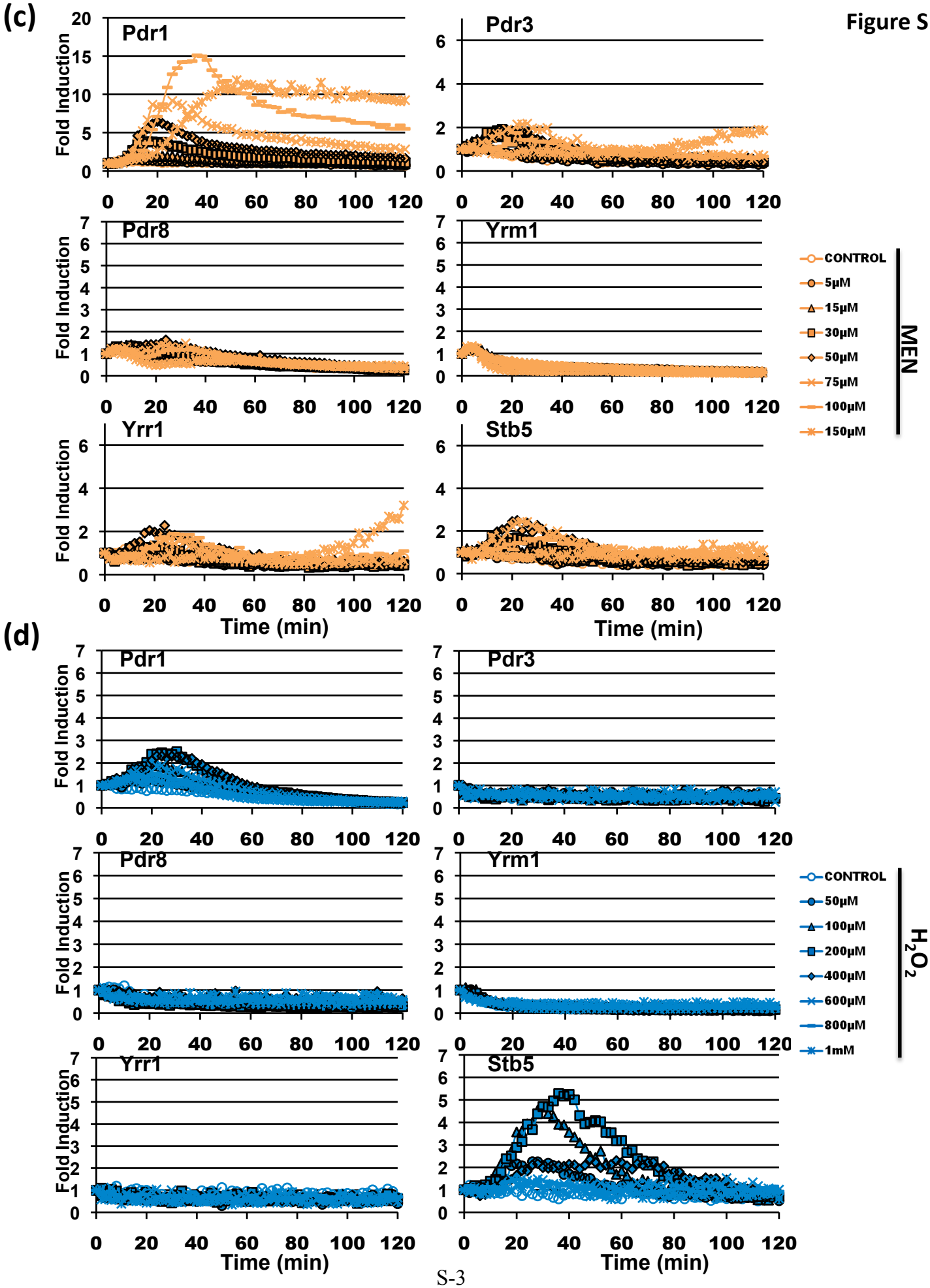

(c)
Pdr1
Pdr3
Pdr8
Yrm1
Yrr1
Stb5
Fold Induction
Time (min)
CONTROL
5µM
15µM
30µM
50µM
75µM
100µM
150µM
MEN
(d)
Pdr1
Pdr3
Pdr8
Yrm1
Yrr1
Stb5
Fold Induction
Time (min)
CONTROL
50µM
100µM
200µM
400µM
600µM
800µM
1mM
H2O2

Figure S1. **Dose response profiles of six Pdr TFs upon different xenobiotic treatment.** The indicated transcription factors were expressed as $Gal4_{DBD}$ hybrid proteins and their transactivation activity measured in real time with a $GAL1_{UAS}$-lucCP^{+} reporter as explained in Fig. 4. The gene expression profiles were determined upon exposure of the indicated concentrations of CIT (a), OTA (b), MEN (c) and H_2O_2 (d). The light emission from three independent culture aliquots was continuously measured. The fold induction was calculated as described in Materials. SD was <15% throughout the experiment, but is not included in the graphs.

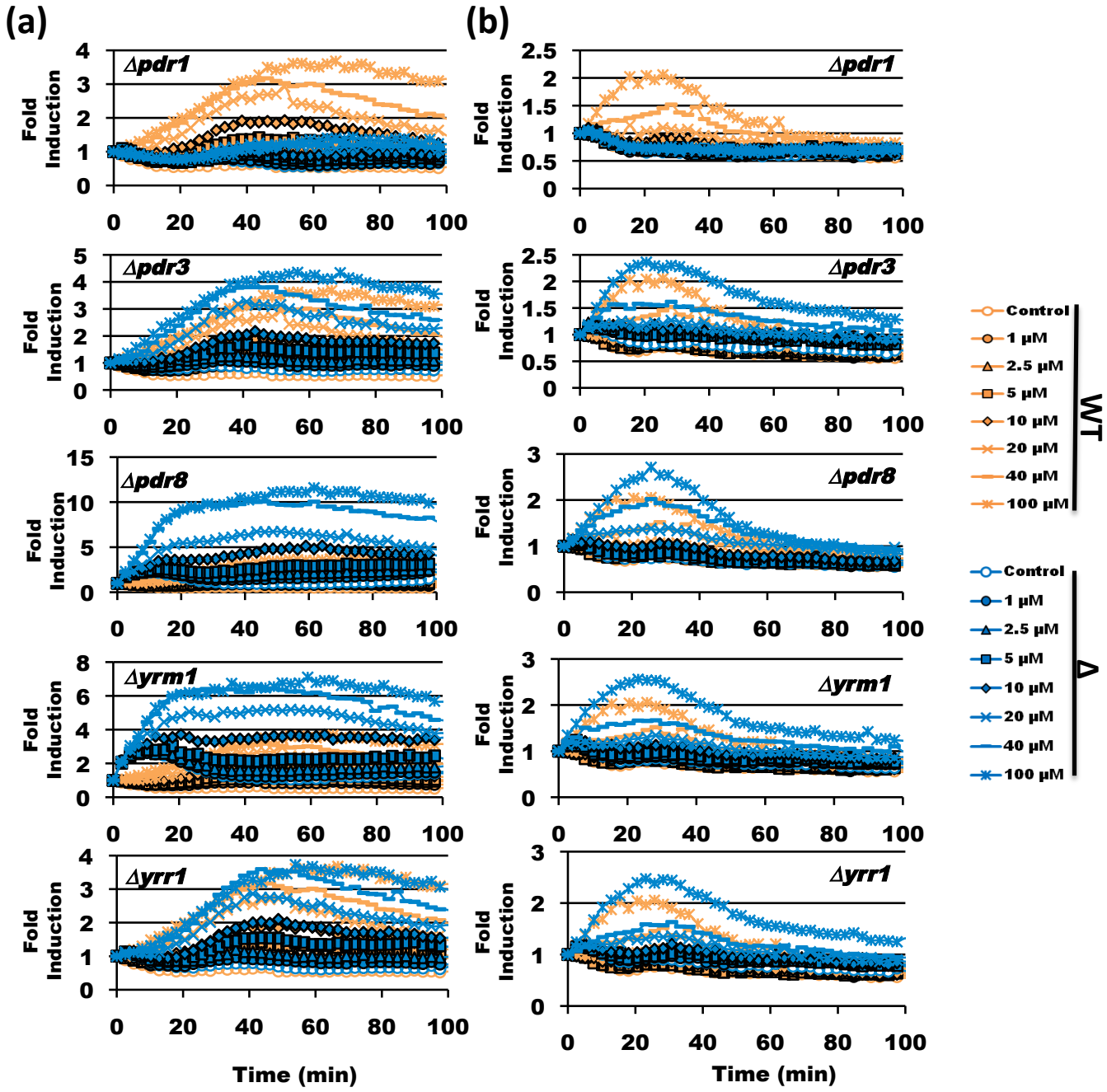


Figure S2. **The dose dependent response of Pdr TF mutants upon CIT and OTA exposure.** The indicated yeast strains were assayed with a PDRE-lucCP^{+} live cell reporter. Dose response profiles are shown upon the treatment with the indicated CIT (a) or OTA (b) concentrations. The light emission from three independent culture aliquots was continuously measured. The fold induction was calculated as described in Materials. SD was <15% throughout the experiment, but is not included in the graphs.

Table S1: Yeast strains used in this study

Name		Source
BY4741	*MATa; his3Δ1; leu2Δ0; met15Δ0; ura3Δ0*	EUROSCARF
BY4741-3xPDRE-lucCP^{+}	BY4741 with plasmid pAG413-3xPDRE-LucCP^{+}	This study
Δpdr1-3xPDRE-lucCP^{+}	BY4741 with *pdr1::KAN* with plasmid pAG413-3xPDRE-lucCP^{+}	This study
Δpdr3-3xPDRE-lucCP^{+}	BY4741 with *pdr3::KAN* with plasmid pAG413-3xPDRE-lucCP^{+}	This study
Δpdr8-3xPDRE-lucCP^{+}	BY4741 with *pdr8::KAN* with plasmid pAG413-3xPDRE-lucCP^{+}	This study
Δyrm1-3xPDRE-lucCP^{+}	BY4741 with *yrm1::KAN* with plasmid pAG413-3xPDRE-lucCP^{+}	This study
Δyrr1-3xPDRE-lucCP^{+}	BY4741 with *yrr1::KAN* with plasmid pAG413-3xPDRE-lucCP^{+}	This study
PDR5-lucCP^{+}	BY4741 with pPDR5-lucCP^{+}-CYC1T-KANMX4	This study
PDR15-lucCP^{+}	BY4741 with pPDR15-lucCP^{+}-CYC1T-KANMX4	This study
SNQ2-lucCP^{+}	BY4741 with pSNQ2-lucCP^{+}-CYC1T-KANMX4	This study
YOR1-lucCP^{+}	BY4741 with pYOR1-lucCP^{+}-CYC1T-KANMX4	This study
Δpdr1-PDR5-lucCP^{+}	BY4741 *pdr1::HIS3* with pPDR5-lucCP^{+}-CYC1T-KANMX4	This study
Δpdr1-PDR15-lucCP^{+}	BY4741 *pdr1::HIS3* with pPDR15-lucCP^{+}-CYC1T-KANMX4	This study
Δpdr1-SNQ2-lucCP^{+}	BY4741 *pdr1::HIS3* with pSNQ2-lucCP^{+}-CYC1T-KANMX4	This study
Δpdr3-PDR5-lucCP^{+}	BY4741 *pdr3::HIS3* with pPDR5-lucCP^{+}-CYC1T-KANMX4	This study
Δpdr3-PDR15-lucCP^{+}	BY4741 *pdr3::HIS3* with pPDR15-lucCP^{+}-CYC1T-KANMX4	This study
Δpdr3-SNQ2-lucCP^{+}	BY4741 *pdr3::HIS3* with pSNQ2-lucCP^{+}-CYC1T-KANMX4	This study
W303-1A	*MATa; can1-100, his3-11'15, leu2-3'112, trp1-1, ura3-1, ade2-1*	R. Serrano
Δgal4	W303-1A with *gal4::KANMX4*	This study
Δgal4 Gal4$_{DBD}$-Pdr1	W303-1A *gal4::KANMX4* with pGBKT7-ADH1p-Gal4$_{DBD}$-Pdr1-myc	This study
Δgal4 Gal4$_{DBD}$-Pdr3	W303-1A *gal4::KANMX4* with pGBKT7-ADH1p-Gal4$_{DBD}$-Pdr3-myc	This study
Δgal4 Gal4$_{DBD}$-Pdr8	W303-1A *gal4::KANMX4* with pGBKT7-ADH1p-Gal4$_{DBD}$-Pdr8-myc	This study
Δgal4 Gal4$_{DBD}$-Yrm1	W303-1A *ga14::KANMX4* with pGBKT7-ADH1p-Gal4$_{DBD}$-Yrm1-myc	This study
Δgal4 Gal4$_{DBD}$-Yrr1	W303-1A *gal4::KANMX4* with pGBKT7-ADH1p-Gal4$_{DBD}$-Yrr1-myc	This study
Δgal4 Gal4$_{DBD}$-Stb5	W303-1A *gal4::KANMX4* with pGBKT7-ADH1p-Gal4$_{DBD}$-Stb5-myc	This study
Δgal4 Gal4$_{DBD}$-Pdr1 GAL1$_{UAS}$-lucCP^{+}	W303-1A *gal4::KANMX4* with pGBKT7-ADH1p-Gal4$_{DBD}$-Pdr1-myc and pAG413-GAL1$_{UAS}$-lucCP^{+}	This study
Δgal4 Gal4$_{DBD}$-Pdr3 GAL1$_{UAS}$-lucCP^{+}	W303-1A *gal4::KANMX4* with pGBKT7-ADH1p-Gal4$_{DBD}$-Pdr3-myc and pAG413-GAL1$_{UAS}$-lucCP^{+}	This study
Δgal4 Gal4$_{DBD}$-Pdr8 GAL1$_{UAS}$-lucCP^{+}	W303-1A *gal4::KANMX4* with pGBKT7-ADH1p-Gal4$_{DBD}$-Pdr8-myc and pAG413-GAL1$_{UAS}$-lucCP^{+}	This study
Δgal4 Gal4$_{DBD}$-Yrm1 GAL1$_{UAS}$-lucCP^{+}	W303-1A *ga14::KANMX4* with pGBKT7-ADH1p-Gal4$_{DBD}$-Yrm1-myc and pAG413-GAL1$_{UAS}$-lucCP^{+}	This study
Δgal4 Gal4$_{DBD}$-Yrr1 GAL1$_{UAS}$-lucCP^{+}	W303-1A *gal4::KANMX4* with pGBKT7-ADH1p-Gal4$_{DBD}$-Yrr1-myc and pAG413-GAL1$_{UAS}$-lucCP^{+}	This study
Δgal4 Gal4$_{DBD}$-Stb5 GAL1$_{UAS}$-lucCP^{+}	W303-1A *gal4::KANMX4* with pGBKT7-ADH1p-Gal4$_{DBD}$-Stb5-myc and pAG413-GAL1$_{UAS}$-lucCP^{+}	This study
PDR1-HA	BY4741 with *PDR1*-3xHA-KANMX4	This study
PDR3-HA	BY4741 with *PDR3*-3xHA-KANMX4	This study

Table S2: Plasmid constructions used in this study.

Name	Description	Source
pUG6-lucCP^{+}-CYC1T-KAN	*AmpR, lucCP^{+}-Cyc1T-loxp-KANMX-loxp*	Pascual-Ahuir *et al.*, 2019
pAG413-CYC1Δ-lucCP^{+}	*AmpR, CEN, HIS3, pCYC1Δ-lucCP^{+}*	Rienzo *et al*., 2012
pAG413-3xPDRE-lucCP^{+}	*AmpR, CEN, HIS3*, *pCYC1Δ-3xPDRE-lucCP^{+}*	This study
pAG413-GAL1$_{UAS}$-lucCP^{+}	*AmpR, CEN, HIS3*, *pCYC1Δ-GAL1$_{UAS}$-lucCP^{+}*	This study
pGBKT7-ADH1p-Gal4$_{DBD}$-myc	*KanR, TRP1*	Clontech
pGBKT7-ADH1p-Gal4$_{DBD}$-Pdr1-myc	pGBKT7 with Pdr1 XBD	This study
pGBKT7-ADH1p-Gal4$_{DBD}$-Pdr3-myc	pGBKT7 with Pdr3 XBD	This study
pGBKT7-ADH1p-Gal4$_{DBD}$-Pdr8-myc	pGBKT7 with Pdr8 XBD	This study
pGBKT7-ADH1p-Gal4$_{DBD}$-Yrm1-myc	pGBKT7 with Yrm1 XBD	This study
pGBKT7-ADH1p-Gal4$_{DBD}$-Yrr1-myc	pGBKT7 with Yrr1 XBD	This study
pGBKT7-ADH1p-Gal4DBD-Stb5-myc	pGBKT7 with Stb5 XBD	This study

Table S3: Oligonucleotides used in this study.

Name	Sequence 5' - 3'	Description
PDR5-luc-KAN1	CTTTTAAGTTTTCGTATCCGCTCGTTCGAAAGACTTTAGACAAAA CCATGGCCGATGCTAAGAAC	Forward primer for *PDR5* replacement with lucCP$^+$
PDR5-luc-KAN2	GTCCATCTTGGTAAGTTTCTTTTCTTAACCAAATTCAAAATTCTA GCATAGGCCACTAGTGGATCTG	Reverse primer for *PDR5* replacement with lucCP$^+$
PDR5-292	GTGGTACGATATCTGTTGAACG	Forward primer to verify lucCP$^+$ integration
SNQ2-luc-KAN1	AGTGGATAGAATAACACAGCTACCAAAATACGTAAAGAGAATT CACCATGGCCGATGCTAAGAAC	Forward primer for *SNQ2* replacement with lucCP$^+$
SNQ2-luc-KAN2	AAAGGCAGATGAATGCACAAAATGTTAAGTTATCTGAAGCCCA CAGCATAGGCCACTAGTGGATCTG	Reverse primer for *SNQ2* replacement with lucCP$^+$
SNQ2-273	CAAGTTGAAGTGTTGCGAGGTC	Forward primer to verify lucCP$^+$ integration
PDR15-luc-KAN1	ACACACACACACACAAGCAAACACACTTATAATTATCAAAAACC TCCATGGCCGATGCTAAGAAC	Forward primer for *PDR15* replacement with lucCP$^+$
PDR15-luc-KAN2	TATAATAAAAAGATAATATAACTAAAAAAAAGGAAAATAACGTC AGCATAGGCCACTAGTGGATCTG	Reverse primer for *PDR15* replacement with lucCP$^+$
PDR15-278	CTGCTACTGCTGTGCGAGAC	Forward primer to verify lucCP$^+$ integration
YOR1-luc-KAN1	CTGTTTTTATATTCAAAAAGAGTAAAGCCGTTGCTATATACGAAT CCATGGCCGATGCTAAGAAC	Forward primer for *YOR1* replacement with lucCP$^+$
YOR1-luc-KAN2	CATATAAATAAATAAAAGAGAAAAATCATGCAACAAATAATATA AAGCATAGGCCACTAGTGGATCTG	Reverse primer for *YOR1* replacement with lucCP$^+$
YOR1-247	ACACATCTGTCAGAGGTAGC	Forward primer to verify lucCP$^+$ integration
LucSeqRev	GGTGATGTCCACCTCAATG	Reverse primer to verify lucCP$^+$ integration
Kan-B	GGATGTATGGGCTAAATG	Reverse primer to verify KanMX integration
BspEI-EcoRV-3xPDRE-1	CCGGCGATATCTCCGTGGATAGAATACATCCGTGGATCGCGATC ATCCGTGGAT	3xPDRE containing oligonucleotide for PDRE-lucCP$^+$ fusion
BspEI-EcoRV-3xPDRE-2	CCGGATCCACGGATGATCGCGATCCACGGATGTATTCTATCCAC GGAGATATCG	3xPDRE containing oligonucleotide for PDRE-lucCP$^+$ fusion
Gal4D1	ACGCCATCATTTTAAGAGAGGACAGAGAAGCAAGCCTCCTGAA AGCAGCTGAAGCTTCGTACGC	Forward primer for gal4::KanMX deletion
Gal4D2	CAGTTGAAGTGAACTTGCGGGGTTTTTCAGTATCTACGATTCAT TGCATAGGCCACTAGTGGATCTG	Reverse primer for gal4::KanMX deletion
Gal4Chk	TTGAGACAGCATTCGCCCAG	Forward primer to verify KanMX integration
Gal1$_{UAS}$-MunI	CCGGCAATTGTGGAAATGTAAAGAGCCCC	Forward primer for *GAL1$_{UAS}$* (-551/-336)
Gal1$_{UAS}$-Kpn2I	ATCGTCCGGAGCAGTGCGGCGCGAG	Reverse primer for *GAL1$_{UAS}$* (-551/-336)
PDR1-NcoI-N	CATGCCATGGGTGGCGCTCGAATAAAAAACC	Forward primer for Gal1$_{UAS}$-Pdr1(81-1068) fusion
PDR1-BamHI-N	CATGGATCCAAACGTATACGTTTAACTATCTGG	Reverse primer for Gal1$_{UAS}$-Pdr1(81-1068) fusion
PDR3-NcoI	CATGCCATGGATAGTTCCCAGTCTTTGCC	Forward primer for Gal1$_{UAS}$-Pdr3(53-976) fusion
PDR3-BamHI	CATGGATCCGTTTTCATAAGAAGGGATATG	Reverse primer for Gal1$_{UAS}$-Pdr3(53-976) fusion
YRM1-NcoI	CATGCCATGGAGTTATTTGGCAGTACTCCG	Forward primer for Gal1$_{UAS}$-Yrm1(73-786) fusion
YRM1-BamHI	CATGGATCCTCTACTGCGTATCAAATAA	Reverse primer for Gal1$_{UAS}$- Yrm1(73-786) fusion
YRR1-NcoI	CATGCCATGGAAAAGAAAGCTACCTATGGTCC	Forward primer for Gal1$_{UAS}$-Yrr1(93-810) fusion
YRR1-BamHI	CATGGATCCTCGACTTAGCATTAATTGTC	Reverse primer for Gal1$_{UAS}$- Yrr1(93-810) fusion
PDR8-EcoRI	CATGAATTCGAGCAAGTGCCCAATGTAG	Forward primer for Gal1$_{UAS}$-Pdr8(74-701) fusion
PDR8-BamHI	CATGGATCCATGTAAAAAAATACATCAATC	Reverse primer for Gal1$_{UAS}$-Pdr8(74-701) fusion
STB5-EcoRI	CATGAATTCAAGAAGGAGTTAGCGGATGC	Forward primer for Gal1$_{UAS}$-Stb5(59-743) fusion
STB5-BamHI	CATGGATCCTCGGTGAACATATGTCATAC	Reverse primer for Gal1$_{UAS}$-Stb5(59-743) fusion