

Supplementary Material

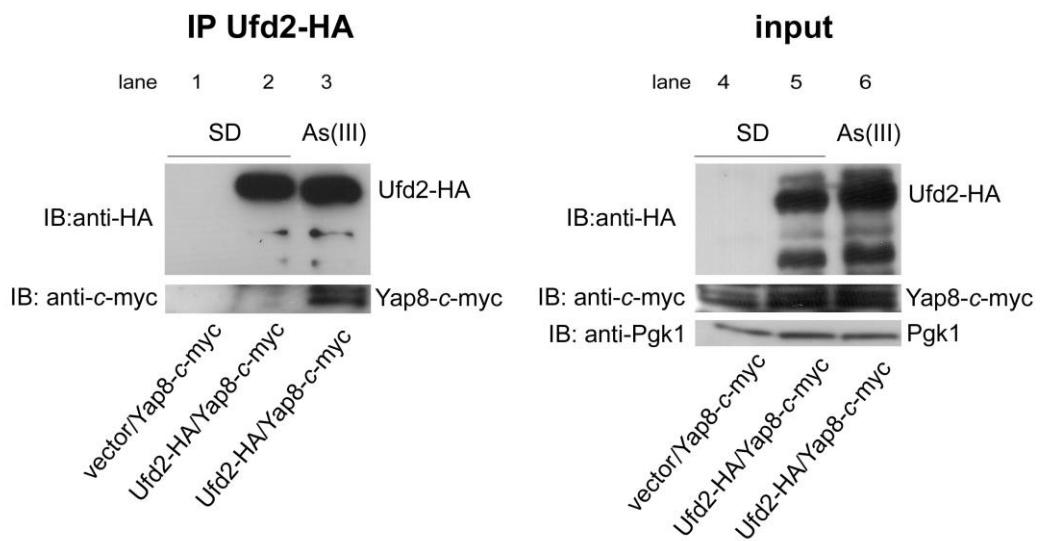


Fig. S1. Reciprocal co-immunoprecipitation assays showing arsenic-mediated interaction between Yap8 and Ufd2. BY4742 cells co-transformed with vector/*YAP8-c-myc* (lanes 1 and 4) or *UFD2-HA/YAP8-c-myc* (lanes 2, 3, 5 and 6) were exposed or not to 1.5 mM As(III) for 90 min and Ufd2-HA was immunoprecipitated with anti-HA antibody. Immunoblotting was performed using anti-HA, anti-*c-myc* and anti-Pgk1 antibodies. A representative experiment is shown. IP – immunoprecipitation; IB – immunoblotting; SD – Synthetic Defined medium, control condition.

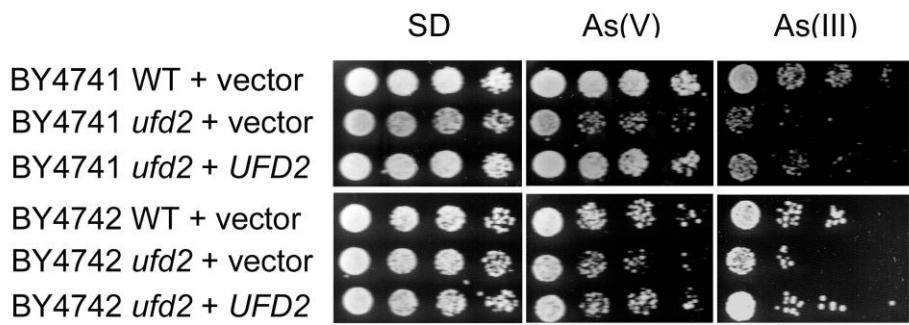


Fig. S2. As(V)- and As(III)-sensitivity phenotypes of *ufd2* are restored by expressing an episomal copy of *UFD2*. Exponential phase BY4741 and BY4742 wild type (WT) and *ufd2* cells expressing an episomal copy of *UFD2* or the respective control vector were serially diluted and spotted onto SD media supplemented or not with 2 mM As(V) or 1.5 mM As(III). Growth was recorded after 2 days incubation at 30°C. A representative experiment is shown.

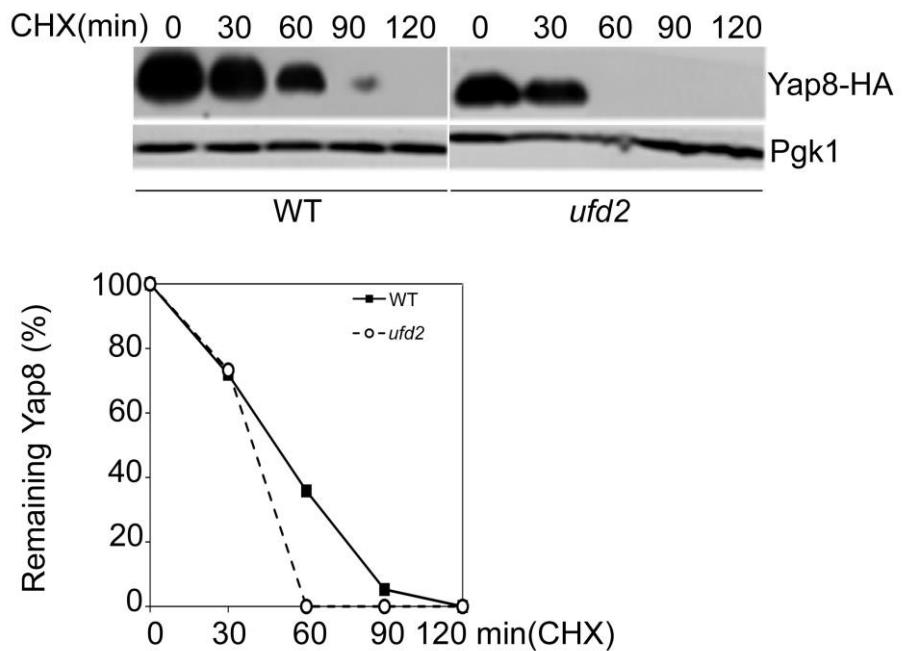


Fig. S3. Ufd2 mediates Yap8 stabilization under As(V) stress conditions. BY4742 wild type (WT) and *ufd2* mutant strains expressing Yap8-HA were pre-treated with 2 mM As(V) for 60 min, washed and subsequently treated with 0.1 mg/mL cycloheximide (CHX) up to 120 min prior to immunoblotting using anti-HA and anti-Pgk1 antibodies. The graph represents the percentage of remaining Yap8 protein after CHX addition. A representative experiment is shown.

Table S1. *Saccharomyces cerevisiae* strains used in this study

| Strain | Genotype | Reference or source |
|------------------------------|---|--------------------------------|
| BY4742 | <i>MATa his3Δ1 leu2Δ0 lys2Δ0 ura3Δ0</i> | EUROSCARF* |
| <i>ufd2</i> | <i>MATa his3Δ1 leu2Δ0 lys2Δ0 ura3Δ0</i> <i>YDL190c::kanMX4</i> | EUROSCARF |
| <i>yap8</i> | <i>MATa his3Δ1 leu2Δ0 lys2Δ0 ura3Δ0</i> <i>YPR199c::kanMX4</i> | EUROSCARF |
| <i>yap8ufd2</i> | <i>MATa his3Δ1 leu2Δ0 lys2Δ0 ura3Δ0</i> <i>YPR199c::kanMX4 YDL190c::HIS3MX4</i> | This study |
| <i>ubc4</i> | <i>MATa his3Δ1 leu2Δ0 lys2Δ0 ura3Δ0</i> <i>YBR082c::kanMX4</i> | ATCC** |
| <i>rad23</i> | <i>MATa his3Δ1 leu2Δ0 lys2Δ0 ura3Δ0</i> <i>YEL037c::kanMX4</i> | EUROSCARF |
| <i>dsk2</i> | <i>MATa his3Δ1 leu2Δ0 lys2Δ0 ura3Δ0</i> <i>YMR276w::kanMX4</i> | EUROSCARF |
| Y187 | <i>MATa ura3-52 his3-200 ade2-101 trp1-901</i> <i>leu2-3 112 gal4Δ met-gal80Δ</i> <i>URA3::GAL1UAS-GAL1TATA-lacZ MEL1</i> | Clontech Laboratories, Inc. |
| BY4741 | <i>MATa his3Δ1 leu2Δ0 met15Δ0 ura3Δ0</i> | EUROSCARF |
| <i>ufd2</i> | <i>MATa his3Δ1 leu2Δ0 met15Δ0 ura3Δ0</i> <i>YDL190C::kanMX4</i> | EUROSCARF |
| <i>Ufd2^{U-boxΔ}</i> | <i>MATa his3Δ1 leu2Δ0 met15Δ0 ura3Δ0</i> <i>YDL190C^{2568-2886 bp}::HIS3</i> | Chang Liu <i>et al.</i> (2011) |

*EUROpean *Saccharomyces Cerevisiae* ARchive for Functional analysis

** American Type Culture Collection

Table S2. Plasmids used in this study

| Name | Features | Reference or source |
|---------------------------|--|-------------------------|
| pRS416 | <i>CEN, URA3</i> | Agilent Technologies |
| pRS416- <i>YAP8-HA</i> | | Our unpublished work |
| pRS416- <i>YAP8-c-myc</i> | | Amaral et al. (2013) |
| pRS416- <i>UFD2-HA</i> | | This study |
| pGADT7-Rec | 2 μ , <i>LEU2, HA</i> | Clontech |
| pGADT7- <i>T-antigen</i> | | Clontech |
| pGADT7- <i>UFD2</i> | | This study |
| pGBK7 | 2 μ , <i>TRP1, c-myc</i> | Clontech |
| pGBK7- <i>LamC</i> | | Clontech |
| pGBK7- <i>p53</i> | | Clontech |
| pGBK7- <i>YAP8</i> | | Our unpublished work |
| YCplac111 | <i>CEN, LEU2</i> | Agilent Technologies |
| YCplac111- <i>UFD2-HA</i> | | This study |
| YEplac181 | 2 μ , <i>LEU2</i> | Agilent Technologies |
| YEplac181- <i>YAP8-HA</i> | | Our unpublished work |
| pGal- <i>MPS1-c-myc</i> | <i>Integrative, URA3, GAL1</i> ^{promoter} | Chang Liu et al. (2011) |

Table S3. Oligonucleotides used in this study

| # | Name | Sequence (5' – 3' UTR) |
|----|--|---|
| 1 | UFD2-HIS-Fw ^D | GGGAAAAGTTAACCTTGAAGTAGAACCCCTCATTCCATAGATCGTAC GCTGCAGG |
| 2 | UFD2-HIS-Rv ^D | TTGATTAGGGTCAATTGCAATTATTCTATCACTTATTAGGGAG ACCGGCAGAT |
| 3 | A1-UFD2 ^D | GGCTTGCTGGTACAATATGG |
| 4 | A4-UFD2 ^D | CCAGAGCTTGAGAAGAG |
| 5 | UFD2 400-Fw* | GAAAGGTAAAGTTGAC |
| 6 | UFD2 1300-Fw* | CTTAAACTCAAGGAC |
| 7 | UFD2 2200-Fw* | GGTAAATTAGTCAG |
| 8 | UFD2-pGADT7- Fw ^C | ACCGCCATAGAAGATATTAC |
| 9 | UFD2-pGADT7- Rv ^C | TCACTCGCTTGCTTATG |
| 10 | pRS416-SmaI- UFD2-P1000-Fw ^C | GAATTCCCTGCAGCCCATAATTCTGTTATTG |
| 11 | HA-UFD2-Rv ^C | AGCGTAATCTGGAACATCGTATGGGTACATTCACTTATTCAATTCA |
| 12 | HA-UFD2- Term500-Fw ^C | ATGTACCCATACGATGTTCCAGATTACGCTTAGAATAAAATTGCAA |
| 13 | UFD2-Term500- pRS416-SmaI-Rv ^C | ACTAGTGGATCCCCAGTGGCTGAATTGA |
| 14 | UFD2-Fw● | GACTTCCCTGTTGGATGAAG |
| 15 | UFD2-Rv● | CTCTTCCTCTTGTGGTG |
| 16 | ACR2-Fw● | AGGCAACTCAAGGCCTAAT |
| 17 | ACR2-Rv● | GAACATGCCAAGCGTTGTA |
| 18 | ACR3-Fw● | AAGAGGGTCTGGGAAGAAA |
| 19 | ACR3-Rv● | GCAATTGCCAGGGATAGTTC |
| 20 | YAP8-Fw● | AACCGCCCACATGTAACACT |
| 21 | YAP8-Rv● | TCCAACACACACTGAGAGCAG |
| 22 | ACT1-Fw● | CTATTGGTAACGAAAGATTCA |
| 23 | ACT1-Rv● | CCTTACGGACATCGACATCA |

Fw - forward; Rv - reverse

^DGene disruption and mutant confirmation, *Sequencing, ^CCloning, ●qRT-PCR