

Supplementary Data

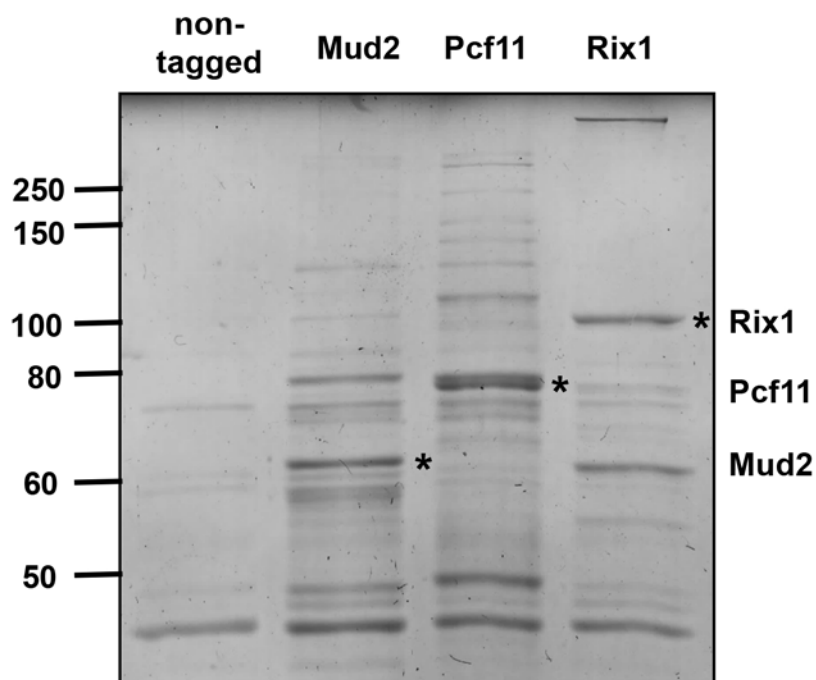
Mud2 functions in transcription by recruiting the Prp19 and TREX complexes to transcribed genes

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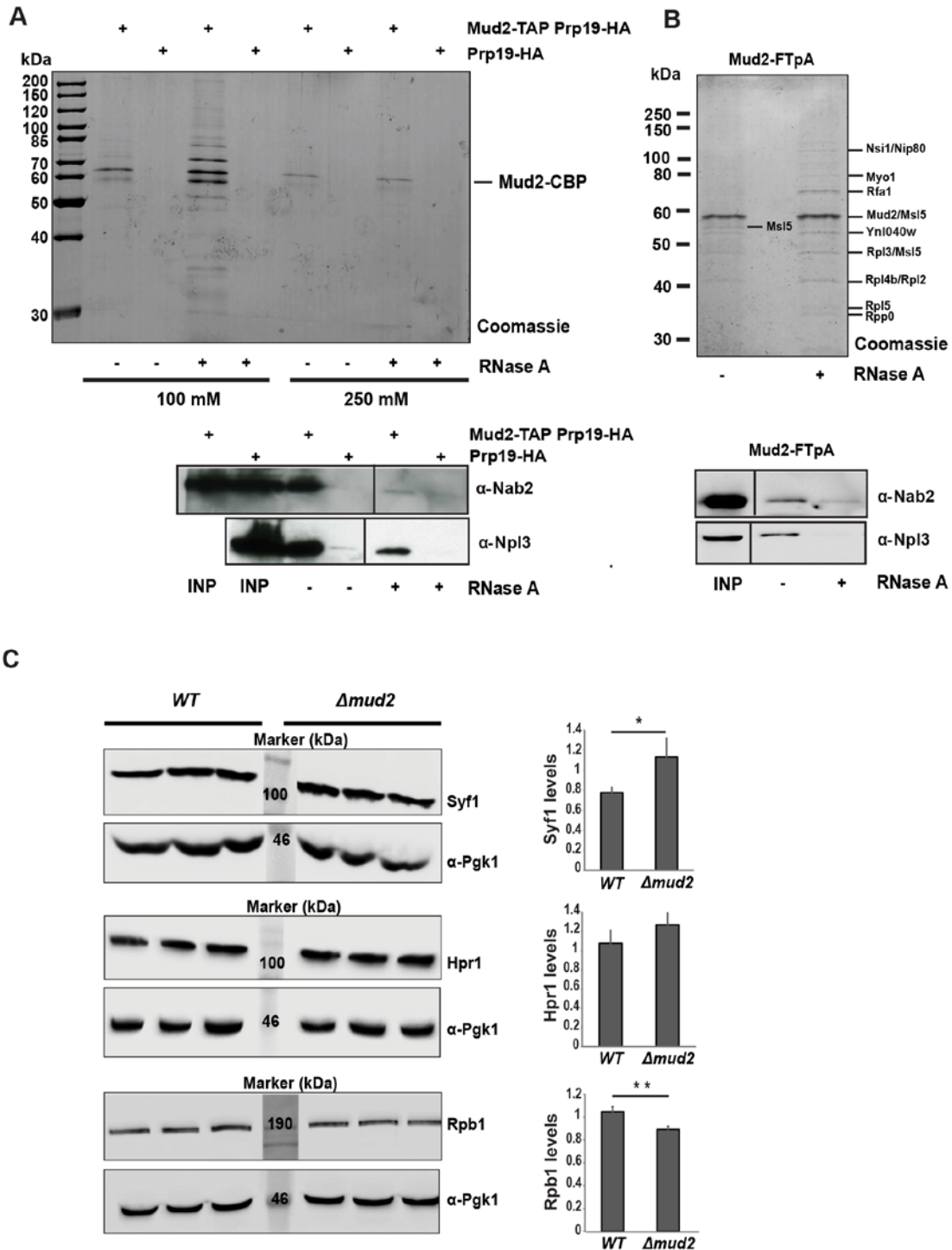
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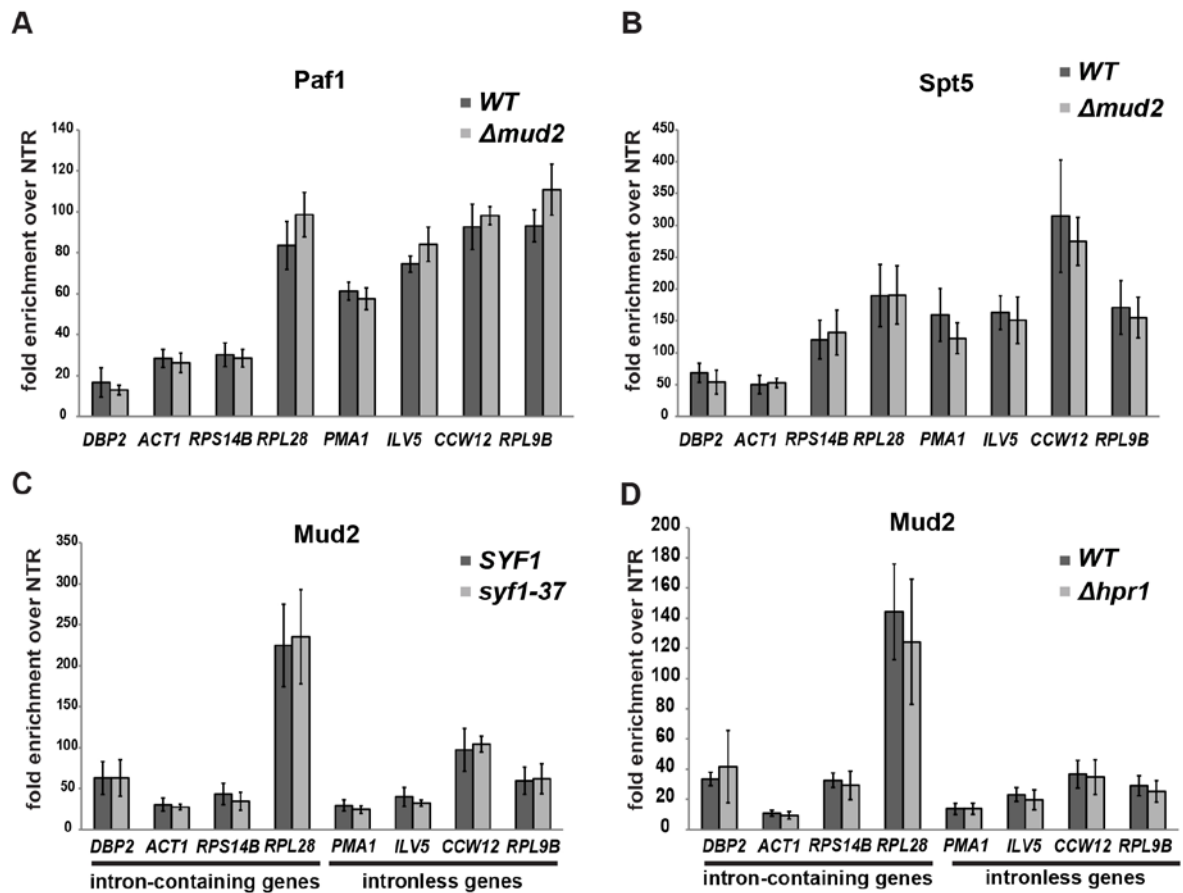
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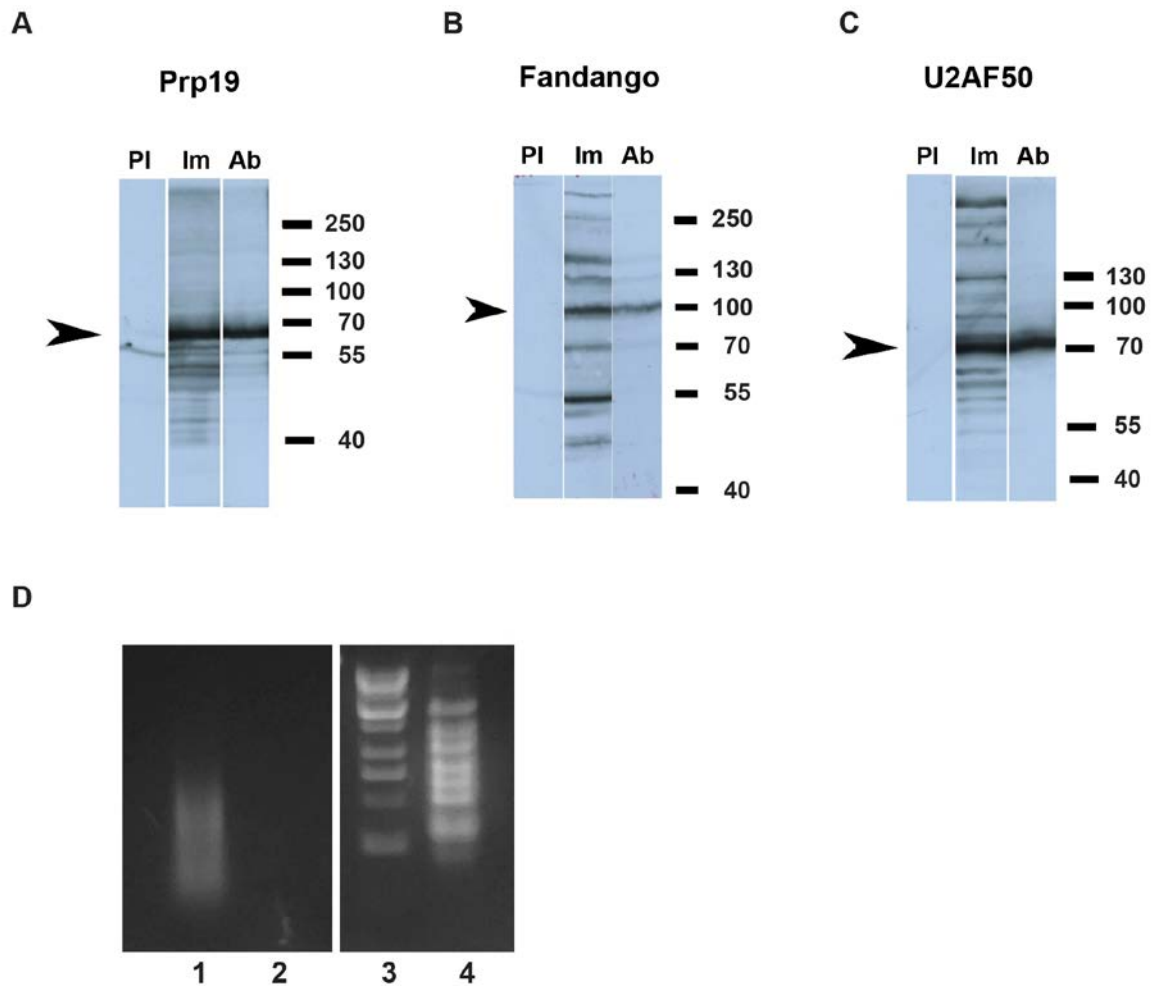
Supplementary Figure 1. Purification of Mud2, Pcf11 and the Rix1 complex from *S. cerevisiae*. Coomassie gel of the TEV-eluates from wild-type, *MUD2-TAP*, *PCF11-TAP* and *RIX1-TAP* strains purified using IgG-coupled tosylactivated Dynabeads M280, respectively, used for the CTD *in vitro* binding experiments shown in Figure 3. The purified tagged proteins are indicated by a star.



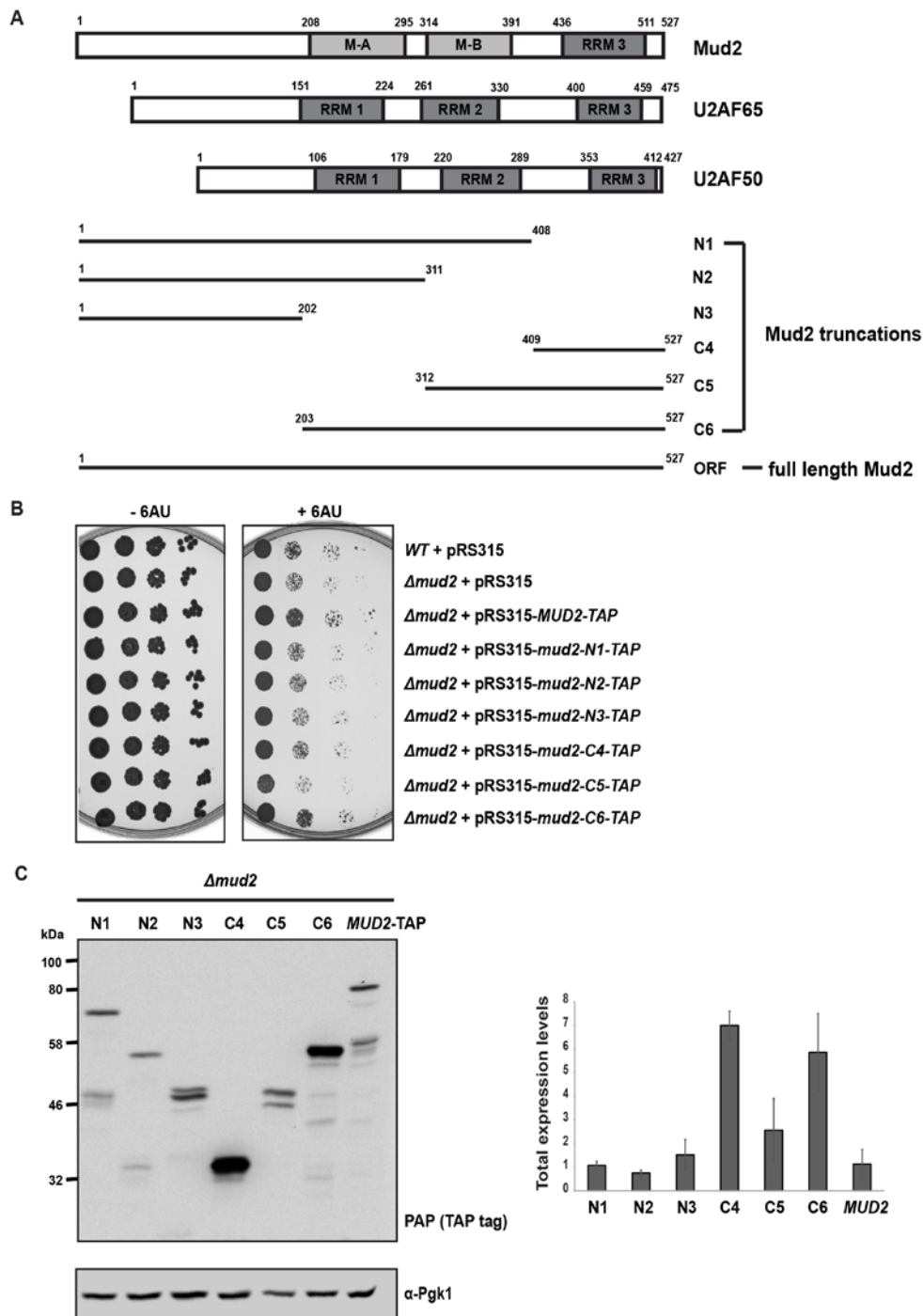
Supplementary Figure 2. (A) Upper panel: Coomassie gel of the EGTA eluates of the Mud2-TAP purifications shown in Figure 4A. Lower panel: Treatment of extracts with RNase A prior to the purification abrogates and greatly diminishes co-purification of Nab2 and Npl3 with Mud2-TAP, respectively, as assessed by Western blotting of TEV eluates. (B) Upper panel: Proteins copurifying with flag-TEV-protA (FTpA) tagged Mud2 in *S. cerevisiae* were identified by mass spectrometry. Lower panel: Treatment of extracts with RNase A prior to the purification abrogates co-purification of Nab2 and Npl3 with Mud2-FTpA as assessed by Western blotting of TEV eluates. (C) Deletion of *MUD2* causes a slight increase of total Syf1 levels and a slight decrease in Rpb1 levels, whereas Hpr1 levels are not affected. Left panel: Western blots to determine the total levels of the Prp19C subunit Syf1, the TREX subunit Hpr1 and the RNAPII subunit Rpb1 in $\Delta mud2$ cells compared to wild-type cells. The picture of the pre-stained marker in the middle of each blot was copied onto the picture of the blot detected by chemiluminescence. Right panel: Quantification of Western blots.



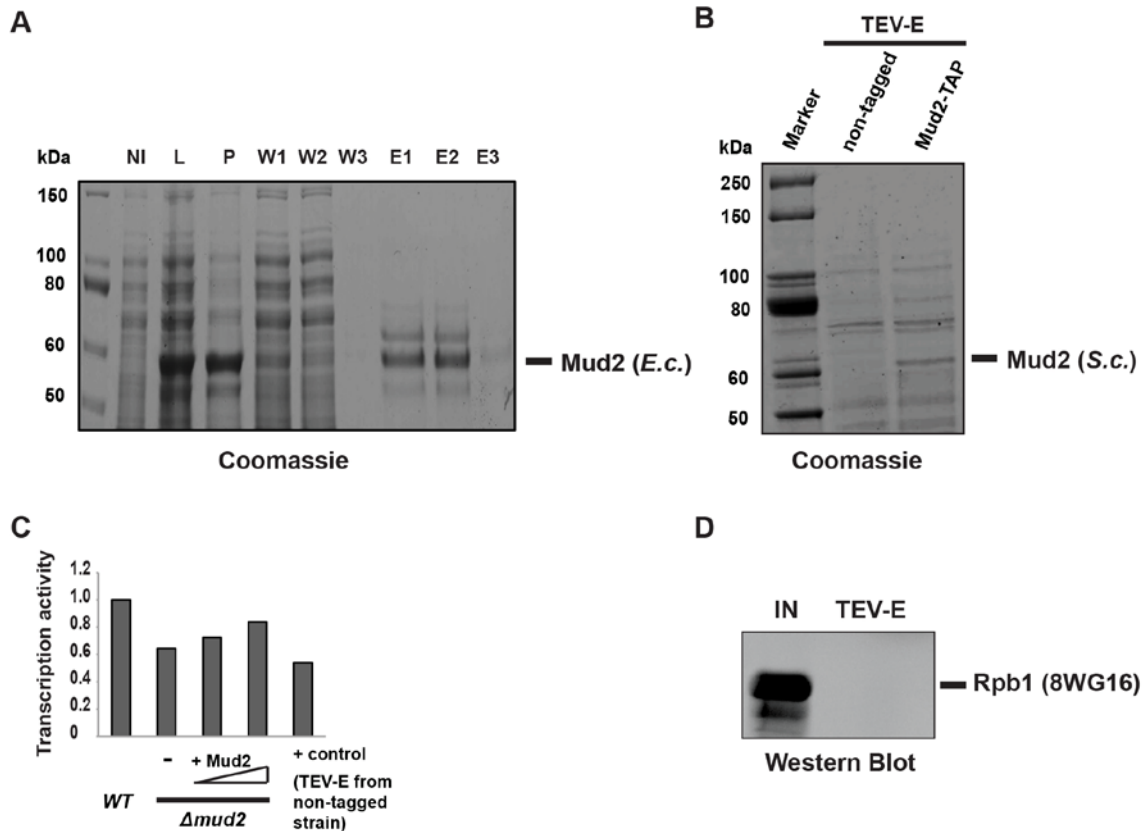
Supplementary Figure 3. Deletion of Mud2 specifically affects the occupancy of Prp19C and TREX. (A and B) The occupancy of the transcription elongation factors Paf1 (PAF complex) (A) and Spt5 (Spt4/5 complex) (B) is not changed in $\Delta mud2$ compared to wild-type cells. (C and D) The occupancy of Mud2 is not changed in *syf1-37* (C) or $\Delta hpr1$ (D) cells. ChIP experiments as in Figure 1.



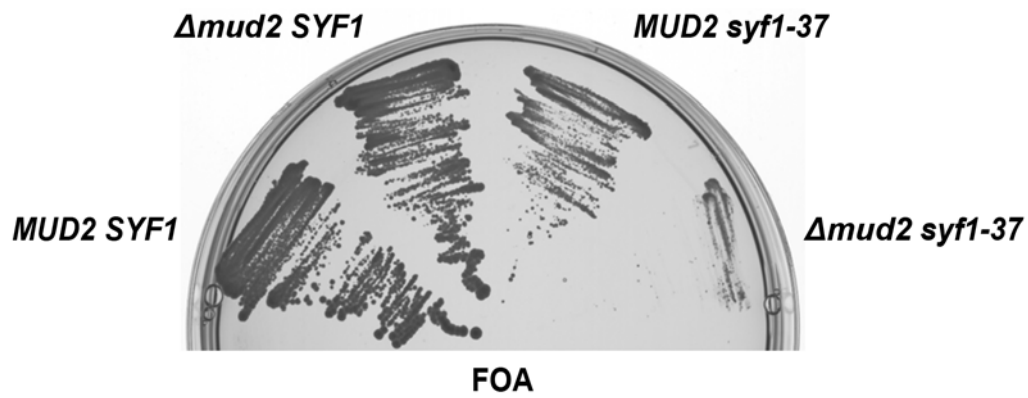
Supplementary Figure 4. Characterization of antibodies raised against the Prp19C subunits Prp19 and Fandango (Syf1) and U2AF50 (Mud2) from *Drosophila melanogaster*. **(A-C)** Western blots on nuclear extracts of S2 cells were performed using pre-immune serum (PI), immune serum (Im) and purified antibody (Abs) raised against Prp19 (A), Fandango (B) and U2AF50 (C), respectively. The band corresponding to the respective protein is indicated by an arrow. **(D)** Protein extracts from *Drosophila* S2 cells contain little RNA/DNA before and no RNA/DNA after treatment with DNase I and RNase A. Nucleic acids were purified from 10 μ l of nuclear extract used for the IP experiments treated or untreated with DNase I and RNase A and separated on a 2% agarose gel. Lane 1: untreated extract; lane 2: extract treated with DNase I and RNase A; lane 3: DNA molecular weight marker: 250, 500, 750, 1000, 1500, 2000, 3000 and 4000/5000/6000 bp; lane 4: RiboRuler high range RNA ladder (#SM1823 Thermo Scientific): 200, 500, 1000, 1500, 2000, 3000, 4000, 6000 b.



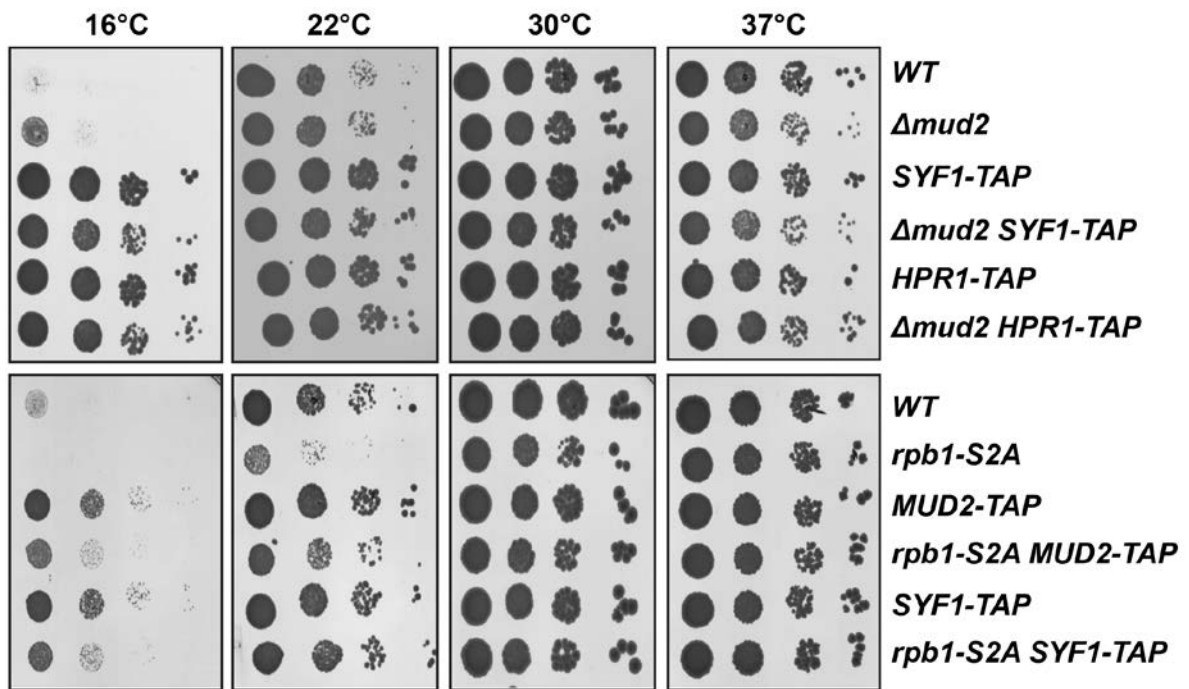
Supplementary Figure 5. The three (potential) RRMs of Mud2 are needed to complement the 6AU sensitivity phenotype of the $\Delta mud2$ strain. **(A)** Scheme showing the domain structure of Mud2 and its potential homologs U2AF65 and U2AF50 (upper panel) as well as the *MUD2* deletion mutants (lower panel). Structural alignment of Mud2 with U2AF65 and splicing factor Prp24 predicts the conserved RRM (RRM3 with high sequence identity) as well as motifs A and B (M-A and M-B) of Mud2 with low sequence identity with RRM1 and RRM2 of U2AF65. **(B)** 6AU sensitivity of the six *MUD2* deletion mutants. Ten-fold serial dilutions of wild-type (WT, RS453 background) and $\Delta mud2$ cells carrying pRS315 and $\Delta mud2$ cells expressing C-terminally TAP-tagged full-length *MUD2* or the six *MUD2* partial deletion mutants as shown in A from pRS315 were spotted on SDC(-ura) plates containing solvent (-6AU) or 50 μ g/ml 6AU (+6AU) and incubated for 2-3 days at 30°C. **(C)** Expression levels of TAP-tagged full-length Mud2 and the six deletion mutants expressed from pRS315. Western blotting against Pgk1 was used as loading control. An exemplary Western blot is shown (left panel). Quantifications of three independent experiments to determine the expression levels of the Mud2 variants normalized to the Pgk1 signal (right panel).



Supplementary Figure 6. Mud2 functions in transcription. **(A)** Coomassie gel of recombinant Mud2 purified from *E. coli* used for the add-back experiment shown in Figure 5D. NI: non-induced; L: total cell lysate; P: pellet; W1-W3: three wash steps; E1-E3: three eluted fractions of purified Mud2. **(B)** Mud2 purified from *S. cerevisiae*. Coomassie gel of the TEV eluates from a TAP purification of a wild-type (non-tagged control strain) and a *MUD2*-TAP strain under high-salt conditions (250 mM NaCl) used for the add-back experiment shown in Supplementary Figure 3C. **(C)** Add-back of Mud2 purified from *S. cerevisiae* increases the transcription activity of a $\Delta mud2$ extract. *In vitro* transcription assay as in Figure 5C with 50 and 150 ng of Mud2 purified from *S. cerevisiae* (Supplementary figure 3B) added to the transcription reaction. As negative control served the TEV eluate from a non-tagged strain (+ control). The quantification of one representative experiment of three independent experiments is shown. **(D)** RNAPII does not co-purify with Mud2 from *S. cerevisiae* under high salt conditions. TEV eluates of the Mud2-TAP purification shown in Supplementary Figure 3B were separated by SDS PAGE and the presence of RNAPII was assessed by Western blotting using antibody 8WG16.



Supplementary Figure 7. *Δmud2* and *syf1-37* are synthetically lethal. A double deletion strain of *MUD2* and *SYF1* complemented by *URA3*-plasmid encoded *SYF1* was transformed with plasmids encoding *SYF1* or *syf1-37* and encoding *MUD2* or an empty plasmid. Combination of *Δmud2* with *syf1-37* causes an sl phenotype. Transformants were streaked onto FOA-containing plates and incubated for 3 days at 30°C.



Supplementary Figure 8. The TAP-tag fused to Mud2, Syf1 and Hpr1 does not impair growth at 16°C, 22°C, 30°C or 37°C. Cells containing the TAP-tag and thus a functional *TRP1* gene grow better as reported by (1).

Supplementary Materials and Methods

Tryptic in-gel digestion of proteins

Bands of interest were excised and the proteins were digested with trypsin. Tryptic peptides were eluted from the gel slices with 1% trifluoroic acid.

Matrix-assisted laser-desorption ionization time-of-flight mass spectrometry (MALDI-TOF-MS)

MALDI-TOF-MS was performed on an Ultraflex TOF/TOF mass spectrometer (Bruker Daltonics, Bremen) equipped with a nitrogen laser and a LIFT-MS/MS facility. The instrument was operated in the positive-ion reflectron mode using 2,5-dihydroxybenzoic acid and methylendiphosphonic acid as matrix. Sum spectra consisting of 200–400 single spectra were acquired. For data processing and instrument control the Compass 1.4 software package consisting of FlexControl 4.4, FlexAnalysis 3.4.4, Sequence Editor and BioTools 3.2 was used. External calibration was performed with a peptide standard (Bruker Daltonics).

Database search

Proteins were identified by MASCOT peptide mass fingerprint search (<http://www.matrixscience.com>) using the Sprot database (version 2017_01, 553474 sequence entries; $p < 0.05$). The search was restricted to *S. cerevisiae*. For the search a mass tolerance of 75 ppm was allowed and oxidation of methionine as variable modification were used.

Supplementary Table 1. Yeast strains.

Strain name	Strain Background	Number	Genotype	Reference
wild-type	RS453	Y1	<i>MATa; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	(2)
wild-type	W303	Y8	<i>MAT a; ade2-1; his3-11, 15; ura3-1; leu2-3, 112; trp1-1; can1-100; rad5-535</i>	(3)
wild-type	BY4741	Y2657	<i>MATa; his3Δ1; leu2Δ0; met15Δ0; ura3Δ0</i>	Euroscarf
<i>MUD2 shuffle</i>	RS453	Y2868	<i>MATa; mud2::kanMX4; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+, pRS316-MUD2</i>	this study
<i>MUD2-TAP</i>	RS453	Y2869	<i>MATa; MUD2-TAP::TRP1; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	this study
GHY498	S288C	Y202	<i>Mat a; his3Δ200; his4-912Δ; lys28Δ; leu2Δ1; ura3-52; rpb1Δ187::HIS3; YCp50-RPB1</i>	(4)
<i>RPB1 shuffle</i>	cross of GHY498 and RS453	Y298	<i>MATa; rpb1Δ187::HIS3; his3; ura3-52; leu2; trp1-1; YCp50-RPB1</i>	this study
<i>RPB1 shuffle MUD2-TAP</i>	cross of GHY498 and RS453	Y2870	<i>MATa; rpb1Δ187::HIS3; MUD2-TAP::TRP1; his3; ura3-52; leu2; trp1-1; YCp50-RPB1</i>	this study

<i>RPB1</i> shuffle <i>SYF1-TAP</i>	cross of GHY498 and RS453	Y2871	<i>MATa; rpb1Δ187::HIS3; SYF1-TAP::TRP1; his3; ura3-52; leu2; trp1-1; YCp50-RPB1</i>	this study
<i>MUD2-TAP</i> <i>PRP19-HA</i>	RS453	Y2872	<i>MATa; MUD2-TAP::TRP1; PRP19-HA::HIS3MS6; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	this study
<i>PRP19-HA</i>	RS453	Y2873	<i>MATa; PRP19-HA::HIS3MX6; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	this study
Δ <i>mud2</i>	RS453	Y2874	<i>MATa; mud2::kanMX4; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	this study
<i>HPR1-TAP</i>	RS453	Y46	<i>MATa; HPR1-TAP::TRP1-KL; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	(5)
Δ <i>mud2</i> <i>HPR1-TAP</i>	RS453	Y2875	<i>MATa; mud2::kanMX4; HPR1-TAP::TRP1; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	this study
<i>SYF1-TAP</i>	RS453	Y641	<i>MAT a; SYF1-TAP::TRP1-KL; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	(6)
Δ <i>mud2</i> <i>SYF1-TAP</i>	RS453	Y2876	<i>MATa; mud2::kanMX4; SYF1-TAP::TRP1; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	this study
<i>PCF11-TAP</i>	RS453	Y713	<i>MATa; PCF11-TAP::TRP1; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	(7)
<i>RIX1-TAP</i>	RS453	Y616	<i>MATa; RIX1-TAP::TRP1; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	(7)
Δ <i>dst1</i>	RS453	Y3033	<i>MAT a; dst1:: HIS3; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	this study
Δ <i>mud2</i> Δ <i>dst1</i>	RS453	Y3034	<i>MAT a; mud2::kanMX4; dst1:: HIS3; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	this study
<i>MUD2- FTpA</i>	RS453	Y3025	<i>MAT a; MUD2-FLAG-TEV-protA::HIS3; ade2-1, his3-11,15, ura3-52, leu2-3,112, trp1-1, can1-100, GAL+</i>	this study
<i>PAF1-TAP</i>	RS453	Y2751	<i>MAT a; PAF1-TAP::TRP1; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	this study
Δ <i>mud2</i> <i>PAF1-TAP</i>	RS453	Y3026	<i>MAT a; mud2::kanMX4; PAF1-TAP::TRP1; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	this study
<i>SPT5-TAP</i>	RS453	Y533	<i>MAT a; SPT5-TAP::TRP1; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	this study
Δ <i>mud2</i> <i>SPT5-TAP</i>	RS453	Y2997	<i>MAT a; mud2::kanMX4; SPT5-TAP::TRP1; ade2-1; his3-11,15; ura3-52; leu2-3,112; trp1-1; can1-100; GAL+</i>	this study

<i>Δmud2</i> <i>SYF1 shuffle</i>	RS453	Y2980	<i>MAT a; mud2::kanMX4; syf1:: HIS3; ade2-1; his3-11, 15; ura3-52; leu2-3, 112; trp1-1; can1-100; GAL+, pRS316-SYF1</i>	this study
<i>SYF1 shuffle</i>	W303	Y2392	<i>MAT a; syf1::kanMX4; ade2-1; his3-11, 15; ura3-1; leu2-3, 112; trp1-1; can1-100; rad5-535, pRS316-SYF1</i>	this study
<i>MUD2-TAP</i> <i>SYF1 shuffle</i>	W303	Y2993	<i>MAT a; syf1::kanMX4; MUD2-TAP::TRP1; ade2-1; his3-11, 15; ura3-1; leu2-3, 112; trp1-1; can1-100; rad5-535, pRS316-SYF1</i>	this study
<i>MUD2-TAP</i>	W303	Y2990	<i>MAT a; MUD2-TAP::TRP1; ade2-1; his3-11, 15; ura3-1; leu2-3, 112; trp1-1; can1-100; rad5-535</i>	this study
<i>MUD2-TAP</i> <i>Δhpr1</i>	W303	Y2998	<i>MAT a; hpr1::HIS3; MUD2-TAP::TRP1; ade2-1; his3-11, 15; ura3-1; leu2-3, 112; trp1-1; can1-100; rad5-535</i>	this study
<i>Δmud2</i>	BY4741	Y2906	<i>MATa; mud2::kanMX4; ura3Δ0; leu2Δ0; his3Δ1; met15Δ0</i>	Euroscarf
<i>Δsnu66</i>	BY4741	Y2907	<i>MATa; snu66::kanMX4; ura3Δ0; leu2Δ0; his3Δ1; met15Δ0</i>	Euroscarf
<i>Δlea1</i>	BY4741	Y2908	<i>MATa; lea1::kanMX4; ura3Δ0; leu2Δ0; his3Δ1; met15Δ0</i>	Euroscarf
<i>Δlin1</i>	BY4741	Y2909	<i>MATa; lin1::kanMX4; ura3Δ0; leu2Δ0; his3Δ1; met15Δ0</i>	Euroscarf
<i>Δnam8</i>	BY4741	Y2910	<i>MATa; nam8::kanMX4; ura3Δ0; leu2Δ0; his3Δ1; met15Δ0</i>	Euroscarf
<i>Δmud1</i>	BY4741	Y2911	<i>MATa; mud1::kanMX4; ura3Δ0; leu2Δ0; his3Δ1; met15Δ0</i>	Euroscarf

Supplementary Table 2. Plasmids.

Plasmid	Number	Description	Reference
pBS1479	84	for C-terminal TAP-tagging of a protein by genomic integration with the <i>TRP1-KL</i> marker	(8)
pY1WT(14)	432	plasmid coding for <i>RPB1</i> with a CTD containing 14 wild-type repeats	(9)
pY1WT(9)A2(6)	434	plasmid coding for <i>RPB1</i> with a CTD containing 9 wild-type and 6 S2A repeats	(9)
pRS316-GAL10- <i>ACT1</i>	1959	<i>GAL10</i> promoter sequence in front of <i>ACT1</i> ORF in pRS316	(6)
pYM15	683	for C-terminal HA-tagging with the <i>HIS3MX6</i> marker	Euroscarf
LL279	1960	Native <i>HIS4</i> promoter ([-428] - [+24] respective to the A in the start codon), used as a template for the <i>in vitro</i> transcription assay	(10)
pRS314	6	A pBluescript-based yeast centromere vector with the <i>TRP1</i> marker	(11)
pRS316	8	A pBluescript-based yeast centromere vector with the <i>URA3</i> marker	(11)
pRS314- <i>MUD2</i>	1998	the coding sequence of <i>MUD2</i> with appr. 500 bp downstream and upstream of the ORF was amplified and cloned into pRS314	this study

pRS316- <i>MUD2</i>	1961	the coding sequence of <i>MUD2</i> with appr. 500 bp downstream and upstream of the ORF was amplified and cloned into pRS316	this study
pRS315- <i>SYF1</i>	2007	a <i>HincII</i> - <i>SpeI</i> fragment containing the <i>SYF1</i> ORF plus downstream and upstream sequences was cloned into pRS315	(6)
pRS315- <i>syf1-37</i>	2008	same as pRS315- <i>SYF1</i> but with C-terminal deletion mutant <i>syf1-37</i>	(6)
pFA6a-FTpA-HisMX6	1962	plasmid used for C-terminal Flag-TEV-protein A tagging by integration into the genome	(12)
pRS315- <i>TAP-T-ADH1</i>	787	the sequence coding for the TAP-tag from pBS1479 was amplified and cloned into pRS315- <i>T-ADH1</i>	(13)
pRS315- <i>MUD2-TAP-T-ADH1</i>	2000	Full-length <i>MUD2</i> (without the stop codon) plus 500 bp of <i>MUD2</i> promoter was cloned into pRS315- <i>TAP-T-ADH1</i>	this study
pRS315- <i>mud2-N1-TAP-T-ADH1</i>	2001	<i>mud2</i> deletion mutant N1 including 500 bp 5' of the <i>MUD2</i> ORF was cloned into pRS315- <i>TAP-T-ADH1</i>	this study
pRS315- <i>mud2-N2-TAP-T-ADH1</i>	2002	<i>mud2</i> deletion mutant N2 including 500 bp 5' of the <i>MUD2</i> ORF was cloned into pRS315- <i>TAP-T-ADH1</i>	this study
pRS315- <i>mud2-N3-TAP-T-ADH1</i>	2003	<i>mud2</i> deletion mutant N3 including 500 bp 5' of the <i>MUD2</i> ORF was cloned into pRS315- <i>TAP-T-ADH1</i>	this study
pRS315- <i>mud2-C4-TAP-T-ADH1</i>	2004	<i>mud2</i> deletion mutant C4 including 500 bp 5' of the <i>MUD2</i> ORF was cloned into pRS315- <i>TAP-T-ADH1</i>	this study
pRS315- <i>mud2-C5-TAP-T-ADH1</i>	2005	<i>mud2</i> deletion mutant C5 including 500 bp 5' of the <i>MUD2</i> ORF was cloned into pRS315- <i>TAP-T-ADH1</i>	this study
pRS315- <i>mud2-C6-TAP-T-ADH1</i>	2006	<i>mud2</i> deletion mutant C6 including 500 bp 5' of the <i>MUD2</i> ORF was cloned into pRS315- <i>TAP-T-ADH1</i>	this study

Supplementary Table 3. Primers.

Primer	Sequence (5'–3')
<i>YER</i> -for	TGCGTACAAAAAGTGTCAAGAGATT
<i>YER</i> -rev	ATGCGCAAGAAGGTGCCTAT
<i>PMA1</i> -5'-for	GTTTTTCGTCGGTCCAATTCA
<i>PMA1</i> -5'-rev	AACCGGCAGCCAAAATAGC
<i>PMA1</i> -M-for	AAATCTTGGGTGTTATGCCATGT
<i>PMA1</i> -M-rev	CCAAGTGTCTAGCTTCGCTAACAG
<i>PMA1</i> -3'-for	CAGAGCTGCTGGTCCATTCTG
<i>PMA1</i> -3'-rev	GAAGACGGCACCAGCCAAT
<i>DBP2</i> -5'-for	CCAAAGCCAATCACCCTTTC
<i>DBP2</i> -5'-rev	CAGCCTTCACTTCATTCAAACG
<i>DBP2</i> -M-for	CGTGACTGGGTTCTACAAGAGTTTAG
<i>DBP2</i> -M-rev	GGCCACATCAGTAGCAACCAT
<i>DBP2</i> -3'-for	CTTCACCGAACAAAACAAAGGTT
<i>DBP2</i> -3'-rev	TCGGGAGGAATATTTTGATTAGCT
<i>DBP2</i> -intron-for	ACGCATACATACGCTTCGTTG
<i>DBP2</i> -intron-rev	AATCTACCCTTGACAAATGCCA
<i>ACT1</i> -5'-for	TGGTATGTTCTAGCGCTTGAC
<i>ACT1</i> -5'-rev	ATCTCTCGAGCAATTGGGACC

<i>ACT1</i> -M-for	GTATTGTCACCAACTGGGACG
<i>ACT1</i> -M-rev	TCTGGGGCAACTCTCAATTCTG
<i>ACT1</i> -3'-for	TCAGAGCCCCAGAAGCTTTG
<i>ACT1</i> -3'-rev	TTGGTCAATACCGGCAGATTCT
<i>ILV5</i> -5'-for	AAGAGAACCTTTGCTTTGGC
<i>ILV5</i> -5'-rev	TTGGCTTAACGAAACGGGCA
<i>ILV5</i> -M-for	TGCCGCTCAATCAGAAACCT
<i>ILV5</i> -M-rev	GGGAGAAACCGTGGGAGAAG
<i>ILV5</i> -3'-for	TGGTACCCAATCTTCAAGAATGC
<i>ILV5</i> -3'-rev	ACCGTTCTTGGTAGATTCTGACA
<i>CCW12</i> -5'-for	ACTGTCGCTTCTATCGCCGC
<i>CCW12</i> -5'-rev	TTGGCTGACAGTAGCAGTGG
<i>CCW12</i> -M-for	CTGTCTCCCCAGCTTTGGTT
<i>CCW12</i> -M-rev	GGCACCAGGTGGTGTATTGA
<i>CCW12</i> -3'-for	TGAAGCTCCAAAGAACCACC
<i>CCW12</i> -3'-rev	AGCAGCAGCACCAGTGTAAAG
<i>RPL9B</i> -5'-for	TCCCAGAAGGTGTTACTGTCAG
<i>RPL9B</i> -5'-rev	TCAAAGTACCTCTTGGACCGAC
<i>RPL9B</i> -M-for	ACATTGTTGAAAAGGATGGTGC
<i>RPL9B</i> -M-rev	CGTTTCTGATCTTCTTGTCCACC
<i>RPL9B</i> -3'-for	AGGACGAAATCGTCTTATCTGGT
<i>RPL9B</i> -3'-rev	CAGATTTGTTGCAAGTCAGCGG
<i>RPL28</i> -5'-for	ACTAGAAAGCACAGAGGTCACG
<i>RPL28</i> -5'-rev	ACCGTCTTGGTTCTTTTCATTCC
<i>RPL28</i> -M-for	TGGTTGTAGAGAGCGCAATTATG
<i>RPL28</i> -M-rev	GGTTTTCAACTGGACATTTTATCG
<i>RPL28</i> -3'-for	TGGAAGCCAGTCTTGAACCTGG
<i>RPL28</i> -3'-rev	TTGGTCTCTCTTGTCTTCTGGGA
<i>RPS14B</i> -5'-for	ACGTGAAAGGGGTGATATCCTG
<i>RPS14B</i> -5'-rev	ACCCGATCACAGTCTCCATC
<i>RPS14B</i> -M-for	GCAGAAGTTCTGTTTACTAACAAC
<i>RPS14B</i> -M-rev	TGAGAGTTATCGCGAGCTTG
<i>RPS14B</i> -3'-for	AAGACCCCAGGACCAGGTG
<i>RPS14B</i> -3'-rev	GATACGGCCAATCCTCAAACCAG

Supplementary Table 4. Identification of proteins co-purifying with Mud2-FTpA as shown in Supplementary Figure S2B.

Lane	Accession	Name	MW [kDa]	Peptides	SC [%]	Rank	Mascot Score
A	NSI1_YEAST	RNA polymerase I termination factor OS= <i>Saccharomyces cerevisiae</i> (strain ATCC 204508 / S288c) GN=NSI1 PE=1 SV=1	66.3	20	28.2	1	55.3
A	NIP80_YEAST	Protein NIP100 OS= <i>Saccharomyces cerevisiae</i> (strain ATCC 204508 / S288c) GN=NIP100 PE=1 SV=2	100.2	17	18.0	2	49.8

B	MYO1 _YEAST	Myosin-1 OS= <i>Saccharomyces cerevisiae</i> (strain ATCC 204508 / S288c) GN=MYO1 PE=1 SV=3	223.5	41	18.7	1	55.1
C	RFA1 _YEAST	Replication factor A protein 1 OS= <i>Saccharomyces cerevisiae</i> (strain ATCC 204508 / S288c) GN=RFA1 PE=1 SV=1	70.3	24	37.4	1	83.9
D	MUD2 _YEAST	Splicing factor MUD2 OS= <i>Saccharomyces cerevisiae</i> (strain ATCC 204508 / S288c) GN=MUD2 PE=1 SV=3	60.4	30	55.2	2	127.0
D	BBP _YEAST	Branchpoint-bridging protein OS= <i>Saccharomyces cerevisiae</i> (strain ATCC 204508 / S288c) GN=MSL5 PE=1 SV=1	53.0	28	54.6	1	138.0
E	BBP _YEAST	Branchpoint-bridging protein OS= <i>Saccharomyces cerevisiae</i> (strain ATCC 204508 / S288c) GN=MSL5 PE=1 SV=1	53.0	20	49.2	1	79.8
F	AJT30041. 1	hypothetical protein H823_YJM1447N00286 [<i>Saccharomyces cerevisiae</i> YJM1447]	50.9	12	26.5	2	73.9
F	GAA26055 .1	K7_Ynl040wp [<i>Saccharomyces cerevisiae</i> Kyokai no. 7]	51.0	12	26.8	1	73.9
G	BBP _YEAST	Branchpoint-bridging protein OS= <i>Saccharomyces cerevisiae</i> (strain ATCC 204508 / S288c) GN=MSL5 PE=1 SV=1	53.0	23	44.3	2	81.7
G	RL3 _YEAST	60S ribosomal protein L3 OS= <i>Saccharomyces cerevisiae</i> (strain ATCC 204508 / S288c) GN=RPL3 PE=1 SV=4	43.7	21	47.3	1	89.0
H	EGA62708 .1	Rpl4bp [<i>Saccharomyces cerevisiae</i> FostersO]	39.1	12	41.7	2	64.3
I	RL5 _YEAST	60S ribosomal protein L5 OS= <i>Saccharomyces cerevisiae</i> (strain ATCC 204508 / S288c) GN=RPL5 PE=1 SV=4	33.7	12	55.2	2	41.6
J	RLA0 _YEAST	60S acidic ribosomal protein P0 OS= <i>Saccharomyces cerevisiae</i> (strain ATCC 204508 / S288c) GN=RPP0 PE=1 SV=2	33.7	15	43.3	1	84.0
J	AAA34974. 1	ribosomal protein L2 [<i>Saccharomyces cerevisiae</i>]	39.1	11	34.5	1	64.7

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