

Supplemental Tables and Figures

Table S1: *Schizosaccharomyces pombe* strains used in this study

Strain	Genotype	Figure	Source
OHI246	<i>h- leu1-32 ura4-D18 ade6-M210</i>	2, 3, 4, 5, 6, 7	K. Gould ^a
OHI408	<i>h- cwf10 2-127Δ ade6-M21X leu1-32 ura4-D18</i>	2, 3, 4, 5, 6, 7, Table 2	This work
OHI479	<i>h+ brr2-HA::kanR ade6-M210 leu1-32 ura4-D18</i>	2	Adapted from K. Gould ^a
OHI480	<i>h+ prp3-linker3XV5 cwf10 2-127Δ ade6-M21X leu1-32 ura4-D18</i>	2	This work
OHI371	<i>h+ prp3-linker3XV5</i>	2	This work
OHI482	<i>h- brr2-HA::kanR cwf10 2-127Δ ade6-M21X leu1-32 ura4-D18</i>	2, S3	This work
OHI323	<i>h- prp1-myc13X::kanR ade6-M210 leu1-32 ura4-D18</i>	2, 4	(1)
OHI439	<i>h- prp1-myc13X::kanR cwf10 2-127Δ ade6-M21X leu1-32 ura4-D18</i>	2, 4	This work
OHI448	<i>h- cwf10 2-127Δ::kanR ade6-M21X leu1-32 ura4-D18</i>	5	This work
OHI463	<i>h+ cdc28-P8</i>	5	Adapted from (2)
OHI467	<i>h+ cdc28-P8 cwf10 2-127Δ::kanR ade6-M21X ura4-D18</i>	5	This work
OHI464	<i>h+ prp10-1</i>	5	Adapted from (3)
OHI468	<i>h+ prp10-1 cwf10 2-127Δ::kanR</i>	5	This work
OHI466	<i>h+ spp42-1::kanR leu1-32</i>	5	Adapted from (4)
OHI465	<i>h+ spp41-1::kanR leu1-32 ura4-D18</i>	5	Adapted from (4)
OHI471	<i>h+ spp41-1::kanR cwf10 2-127Δ ade6-M21X leu1-32 ura4-D18</i>	5	This work
OHI322	<i>h+ prp1-4 ura4-D18</i>	5	Adapted from (3)
OHI321	<i>h+ prp4-73 leu1-32 ura4-294</i>	5	Adapted from (5)
OHI462	<i>h- prp4-73 cwf10 2-127Δ::kanR leu1-32 ura4-D18 or -294</i>	5	This work
OHI478	<i>h+ aar2::kanR ade6-M216 leu1-32 ura4-D18</i>	5	(6)
OHI481	<i>h- aar2::kanR cwf10 2-127Δ ade6-M21X leu1-</i>	5	This work

	<i>32 ura4-D18</i>		
OHI483	<i>h+ cwf10 2-23Δ ade6-M21X leu1-32 ura4-D18</i>	6	This work
OHI260	<i>h- cdc5-TAP::kanR</i>	Tables 1, 2	Adapted from (7)
OHI491	<i>h- cdc5-TAP::kanR cwf10 2-127Δ leu1-32</i>	Table 1	This work
OHI383	<i>h-/h+ cwf10+/cwf10::ura4+ ade6-M210/ade6-M216 leu1-32/leu1-32 ura4-D18/ura4-D18</i>	integrant construction	This work

^aVanderbilt University School of Medicine, Nashville, TN.

Table S2: Primers used in this study

Analysis	Name	Purpose	Primer sequence(s) (5'-3')
RT-PCR	tbp1_a F BL365	amplify first <i>tbp1</i> intron	CGCTTTACCCACCACGGCCTCGCAAG
RT-PCR	tbp1_a R BL366	amplify first <i>tbp1</i> intron	TTCTGCATTACGTGCATGTAGCGC
RT-PCR	mrps16_b F BL541	amplify second <i>mrps16</i> intron	AAAGCACCTCAAGCCAAACCTATCGA
RT-PCR	mrps16_b R BL542	amplify second <i>mrps16</i> intron	TCAAGAAACGGGTTTTTTAGGAAGCAACT
Northern	BL432	hybridize U1	ATAACTGAGTCTCCACAATTCTTG TG
Northern	BL429	hybridize U2	AGAACAGATACTACACTTGATCTAAG
Northern	BL433	hybridize U4	AAAAGTTCCTCACTGATAAGCGTAAT
Northern	BL434	hybridize U5	ACAGTCAAATTAGCACACCTTACAAA
Northern	BL435	hybridize U6	TCTTCTCTGTATCGTTTTCAATTTGAC

Table S3. Pre-mRNA splicing factors co-purifying with TAP-NTE in a *cwf10-ΔNTE* background at either 150 or 75 mM NaCl detected by 1D liquid chromatography-tandem mass spectrometry (LC-MS/MS).

	<i>S. pombe</i>	ORF number	TAP-NTE from <i>cwf10-ΔNTE</i>				<i>S. cerevisiae</i>	<i>H. sapiens</i>
			150 mM NaCl ^a		75 mM NaCl			
			I	II	I	II		
Core snRNP	Smb1	SPAC26A3.08	4 ^b	3	4	3	Smb1	SMB/B'
	Smd1	SPAC27D7.07c	1	3	3	3	Smd1	SMD1
	Smd2	SPAC2C4.03c	6	3	5	6	Smd2	SMD2
	Smd3	SPBC19C2.14	3	1	4	5	Smd3	SMD3
	Sme1	SPBC11G11.06c	1	1	2	3	Sme1	SME1
	Smf1	SPBC3E7.14	5	3	4	3	Smf1	SMF1
	Smg1	SPBC4B4.05	--- ^c	3	2	3	Smg2	SMG1
U2	Lea1	SPBC1861.08c	9	3	15	8	Lea1	U2A'
	Msl1	SPBC8D2.09c	1	---	3	1	Msl1	U2B''
	Prp10	SPAC27F1.09c	---	---	1	---	Hsh155	SF3B160
U5	Cwf10-ΔNTE	SPBC215.12	60	42	71	65	Snu114	U5-116
	Brr2	SPAC9.03c	---	---	---	----	Brr2	U5-200
	Spp42	SPAC4F8.12c	57	33	77	75	Prp8	U5-220
	Cwf17/Spf38	SPBC1289.11	9	4	16	12	UNK ^d	U5-40
NTC core	Cdc5	SPAC644.12	34	13	39	42	Cef1	CDC5
	Cwf2	SPAC3A12.11c	13	5	14	15	Cwc2	RBM22
	Cwf7	SPBC28F2.04c	10	6	14	8	Snt309	SPF27
	Cwf15	SPBC337.06c	6	1	9	7	Cwc15	AD002
	Prp5	SPBP22H7.07	14	8	22	22	Prp46	PRL1
	Prp19	SPAC29A4.08c	18	17	16	25	Prp19	PRP19
NTC-associated	Cwf3	SPBC211.02c	26	9	32	36	Syf1	SYF1
	Cwf4	SPBC31F10.11c	21	11	28	25	Cif1	CRN1
	Cwf5/Ecm2	SPCC550.02c	12	10	21	22	Ecm2/Slt11	RBM22
	Cwf12	SPBC32F12.05c	6	6	8	7	Isy1	ISY1
	Prp45	SPCC188.11	3	3	10	11	Prp45	SKIP
	Prp17	SPBC6B1.10	17	15	25	31	Prp17	hPRP17
	Syf2	SPBC3E7.13c	10	5	10	9	Syf2	SYF2
Other	Cwf11	SPBC646.02	36	17	52	50	UNK	hCWF11
	Cwf14	SPBC24C6.11	8	4	10	11	Cwc14	G10
	Cwf18	SPCP1E11.07c	6	5	8	5	UNK	MGC23918
	Cwf19	SPAC30D11.09	11	3	15	19	UNK	hCWF19L2
	Cwf21	SPAC4A8.09c	1	---	5	2	Cwc21	SRm300
	Cwf22	SPBC13E7.01	2	1	4	6	Cwc22	hCWC22
	Cyp1	SPAC57A10.03	5	1	5	5	UNK	PPIL1
	Mug161	SPAC1F3.09	---	---	---	---	YGR093W	CWF19L1
	Prp16	SPBC1711.17	---	---	---	---	Prp16	PRP16
	Prp43	SPBC16H5.10c	---	---	---	---	Prp43	hPRP43
	Prp22	SPAC10F6.02c	---	---	1	---	Prp22	hPRP22
	Saf4	SPBC18H10.10c	3	3	7	4	UNK	CCDC130

	Slu7	SPBC365.05c	---	---	---	---	Slu7	SLU7
	Sum3	SPCC1795.11	7	1	2	1	Ded1	DDX3
	unnamed	SPAC20H4.09	---	---	---	---	UNK	DHX35
	Pab1	SPAC57A7.04c	---	---	6	6	Pab1	PABC1
	Ubp10	SPBC577.07	---	---	1	4	Sad1	USP39

^aResults for purifications done at 150 mM NaCl are the same as shown in Table 2 and are included again in this table to make comparison easier. ^bPeptide counts of identified proteins in each biological replicate (I and II), ^c --- indicates no peptides identified, ^dUNK indicates that an ortholog is unknown or not present.

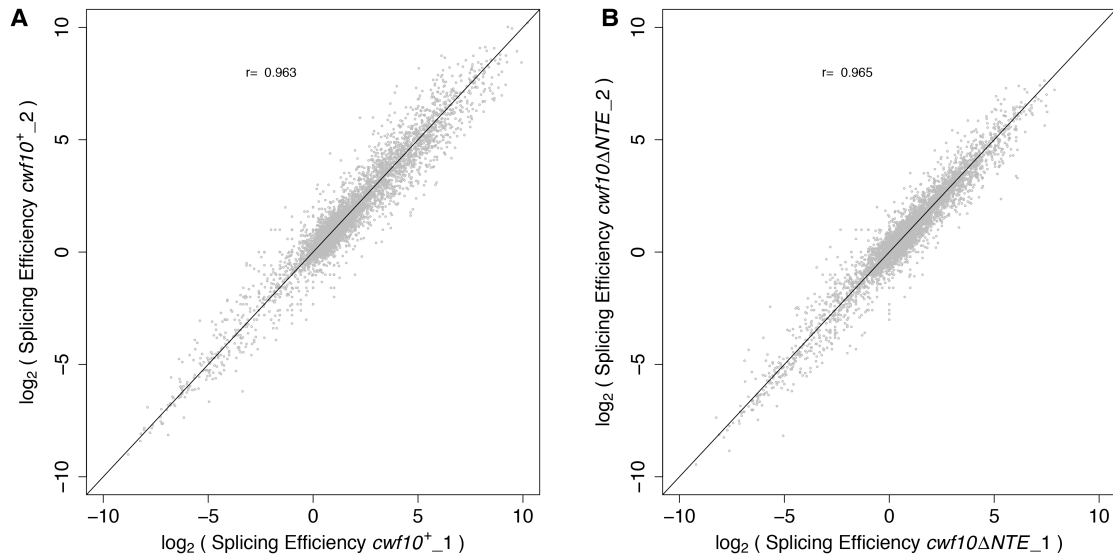


Figure S1. Reproducibility of splicing efficiency in replicates. Each point represents an intron (5,361 in total; 'r' Pearson's correlation coefficient; \log_2 scale). (A) Two wild-type replicates. (B) Two *cwf10*- Δ NTE replicates.

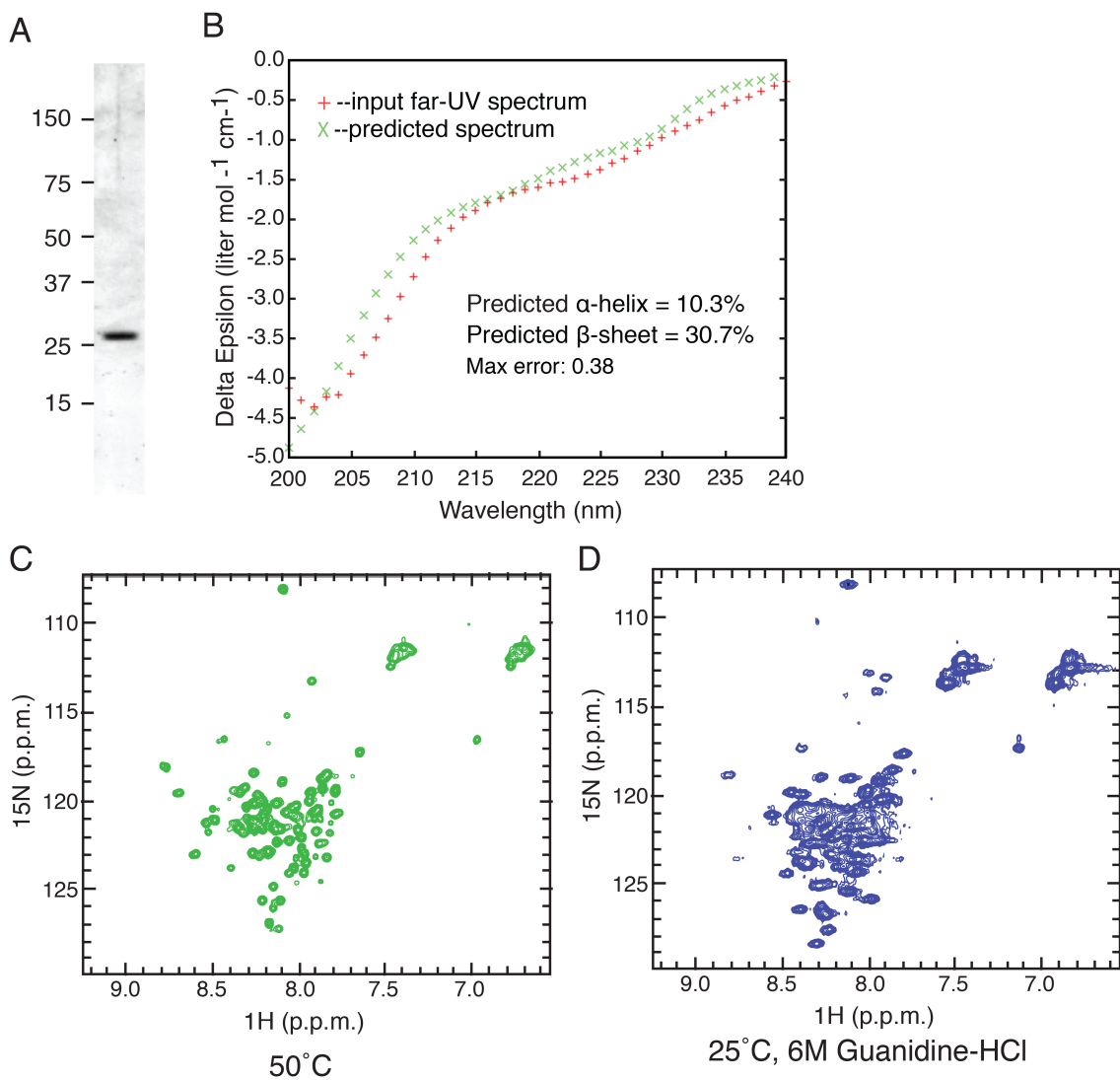


Figure S2. Analysis of recombinant Cwf10(1-135)His₆. (A) Coomassie stained SDS-PAGE gel of recombinant Cwf10(1-135)His₆. Molecular weight markers in kilodaltons are shown on left. (B) Predicted secondary structure calculated from the Far-UV spectrum seen in Fig. 6E using the program K2D2 (1). (C) ¹⁵N-¹H HSQC spectrum of Cwf10(1-135)His₆ at 50°C. (D) ¹⁵N-¹H HSQC spectrum of Cwf10(1-135)His₆ in 6 M Guanidine-HCl.

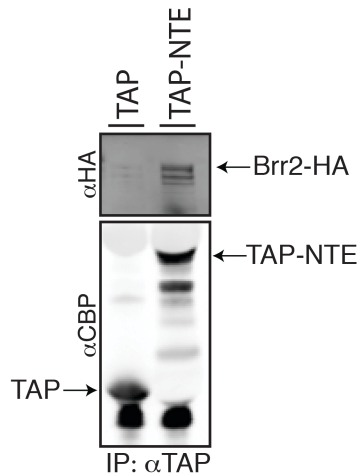


Figure S3. TAP-NTE co-immunoprecipitates Brr2-HA. *pREP41 NTAP* or *pREP41 NTAP-NTE* were expressed in *cwf10-ΔNTE brr2-HA* cells. TAP and TAP-NTE were immunoprecipitated with IgG-Sepharose beads and then immunoblotted with anti-HA antibodies. Positions of proteins on the immunoblot are labeled.

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