| FBgn ID | CG number | Subunit | U2B/SNF | SF3B5 | Ada2b-PB | Spt3 | Spt20 | ATXN7 | Ada1 | SAF6 | Sgf29 | WDA | S2 neg | CG6469 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| FBgn0050390 | CG30390 | Sgf29 | 0.00000 | 0.00082 | 0.02153 | 0.00438 | 0.01041 | 0.00300 | 0.00248 | 0.01297 | 0.14989 | 0.00178 | 0.00000 | 0.00000 |
| FBgn0030891 | CG7098 | Ada3 | 0.00000 | 0.00055 | 0.02864 | 0.00181 | 0.00563 | 0.00223 | 0.00114 | 0.00657 | 0.01179 | 0.00116 | 0.00000 | 0.00000 |
| FBgn0020388 | CG4107 | Gcn5 | 0.00000 | 0.00090 | 0.02387 | 0.00341 | 0.00965 | 0.00290 | 0.00197 | 0.00551 | 0.00483 | 0.00154 | 0.00000 | 0.00000 |
| FBgn0037555 | CG9638 | Ada2b-PB | 0.00000 | 0.00115 | 0.04834 | 0.00280 | 0.00972 | 0.00217 | 0.00133 | 0.01127 | 0.00550 | 0.00152 | 0.00000 | 0.00000 |
| FBgn0051866 | CG31866 | Ada1 | 0.00000 | 0.00062 | 0.02206 | 0.00333 | 0.00735 | 0.00176 | 0.07721 | 0.01165 | 0.00352 | 0.01053 | 0.00000 | 0.00000 |
| FBgn0031281 | CG3883 | SAF6 | 0.00000 | 0.00149 | 0.01162 | 0.00293 | 0.00662 | 0.00317 | 0.00180 | 0.01116 | 0.00202 | 0.01190 | 0.00000 | 0.00000 |
| FBgn0036374 | CG17689 | Spt20 | 0.00000 | 0.00032 | 0.00521 | 0.00351 | 0.02356 | 0.00211 | 0.00047 | 0.00570 | 0.00143 | 0.00038 | 0.00000 | 0.00000 |
| FBgn0037981 | CG3169 | Spt3 | 0.00000 | 0.00105 | 0.00907 | 0.03084 | 0.01204 | 0.00283 | 0.00171 | 0.00968 | 0.00235 | 0.00706 | 0.00000 | 0.00000 |
| FBgn0030874 | CG6506 | Spt7 | 0.00000 | 0.00211 | 0.02433 | 0.01000 | 0.01607 | 0.00766 | 0.00559 | 0.02362 | 0.00590 | 0.01333 | 0.00000 | 0.00000 |
| FBgn0026324 | CG3069 | TAF10b | 0.00000 | 0.00065 | 0.02072 | 0.00187 | 0.00106 | 0.00442 | 0.00084 | 0.00348 | 0.00071 | 0.00966 | 0.00000 | 0.00000 |
| FBgn0011290 | CG17358 | TAF12 | 0.00000 | 0.00015 | 0.00892 | 0.00192 | 0.00485 | 0.00233 | 0.00077 | 0.00218 | 0.00290 | 0.00479 | 0.00000 | 0.00000 |
| FBgn0000617 | CG6474 | TAF9 | 0.00004 | 0.00153 | 0.01622 | 0.00443 | 0.00826 | 0.00287 | 0.00155 | 0.01611 | 0.00214 | 0.01450 | 0.00000 | 0.00000 |
| FBgn0053554 | CG33554 | Tra1 (Nipped-A) | 0.00000 | 0.00073 | 0.00139 | 0.00155 | 0.01116 | 0.00286 | 0.00061 | 0.00569 | 0.00227 | 0.00051 | 0.00000 | 0.00005 |
| FBgn0039067 | CG4448 | WDA | 0.00000 | 0.00137 | 0.01583 | 0.00552 | 0.01557 | 0.00329 | 0.00190 | 0.01398 | 0.00334 | 0.07218 | 0.00000 | 0.00000 |
| FBgn0031420 | CG9866 | ATXN7 | 0.00000 | 0.00012 | 0.00229 | 0.00081 | 0.00345 | 0.02491 | 0.00038 | 0.00154 | 0.00152 | 0.00033 | 0.00000 | 0.00000 |
| FBgn0000618 | CG15191 | E(y)2 | 0.00000 | 0.00305 | 0.00452 | 0.00423 | 0.00798 | 0.00622 | 0.00000 | 0.01793 | 0.00281 | 0.00120 | 0.00000 | 0.00000 |
| FBgn0013717 | CG4166 | Nonstop | 0.00000 | 0.00024 | 0.00016 | 0.00156 | 0.00653 | 0.00365 | 0.00032 | 0.00203 | 0.00095 | 0.00019 | 0.00000 | 0.00000 |
| FBgn0036804 | CG13379 | Sgf11 | 0.00000 | 0.00048 | 0.00116 | 0.00637 | 0.02200 | 0.00918 | 0.00094 | 0.00308 | 0.00105 | 0.00062 | 0.00000 | 0.00000 |
| FBgn0040534 | CG11985 | SF3B5 (SF3b10) | 0.00651 | 0.02480 | 0.02552 | 0.00663 | 0.01168 | 0.00719 | 0.00289 | 0.02205 | 0.00729 | 0.02157 | 0.00000 | 0.00000 |
| FBgn0035162 | CG13900 | SF3B3 (SF3b130) | 0.00617 | 0.05785 | 0.03945 | 0.00890 | 0.01807 | 0.00524 | 0.00290 | 0.01690 | 0.00486 | 0.02513 | 0.00000 | 0.00010 |
| FBgn0031493 | CG3605 | SF3B2 (SF3b145) | 0.00235 | 0.00389 | 0.00000 | 0.00000 | 0.00000 | 0.00007 | 0.00000 | 0.00000 | 0.00017 | 0.00005 | 0.00000 | 0.00000 |
| FBgn0015818 | CG3780 | SF3B4 (SF3b149/Spx) | 0.01224 | 0.02799 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00007 | 0.00000 | 0.00000 | 0.00000 |
| FBgn0035692 | CG13298 | SF3B6 (SF3b14a) | 0.00915 | 0.03935 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00043 | 0.00000 | 0.00000 | 0.00000 |
| FBgn0031822 | CG9548 | PHF5A (SF3b14b) | 0.00087 | 0.00021 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00018 | 0.00000 | 0.00000 |
| FBgn0031266 | CG2807 | SF3B1 (SF3b155) | 0.00500 | 0.02121 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00050 | 0.00000 | 0.00000 | 0.00000 |
| FBgn0266917 | CG16941 | SF3A1 (SF3a120) | 0.00481 | 0.01027 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00010 | 0.00000 | 0.00000 | 0.00000 |
| FBgn0014366 | CG2925 | SF3A3 (SF3a60/noi) | 0.00806 | 0.01323 | 0.00011 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00062 | 0.00000 | 0.00000 | 0.00000 |
| FBgn0036314 | CG10754 | SF3A2 (SF3a66) | 0.00547 | 0.01992 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00010 | 0.00000 | 0.00000 | 0.00000 |
| FBgn0262601 | CG5352 | SmB | 0.02141 | 0.00440 | 0.00000 | 0.00017 | 0.00016 | 0.00000 | 0.00000 | 0.00000 | 0.00026 | 0.00000 | 0.00125 | 0.00090 |
| FBgn0261933 | CG10753 | SmD1 (snRNP69D) | 0.05591 | 0.01070 | 0.00184 | 0.00069 | 0.00050 | 0.00055 | 0.00033 | 0.00000 | 0.00208 | 0.00000 | 0.00201 | 0.00000 |
| FBgn0261789 | CG1249 | SmD2 | 0.03267 | 0.00776 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00087 | 0.00008 | 0.00105 | 0.00000 |
| FBgn0023167 | CG8427 | SmD3 | 0.06161 | 0.00110 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00017 | 0.00000 | 0.00165 | 0.00000 |
| FBgn0261790 | CG18591 | SmE | 0.06709 | 0.01285 | 0.00000 | 0.00018 | 0.00000 | 0.00000 | 0.00000 | 0.00034 | 0.00137 | 0.00000 | 0.00265 | 0.00254 |
| FBgn0000426 | CG16792 | SmF (DebB) | 0.00615 | 0.00350 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00029 | 0.00000 | 0.00000 | 0.00000 |
| FBgn0261791 | CG9742 | SmG | 0.03104 | 0.00280 | 0.00300 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00170 | 0.00000 | 0.00000 | 0.00000 |
| FBgn0033210 | CG1406 | U2A | 0.01453 | 0.01019 | 0.00000 | 0.00032 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00010 | 0.00000 | 0.00000 | 0.00000 |
| FBgn0003449 | CG4528 | U1A/U2B (snf) | 0.14041 | 0.00954 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00000 | 0.00120 | 0.00000 | 0.00000 | 0.00000 |

**Table S1: Distributed normalized spectral abundance factor (dNSAF) values for MudPIT analysis**
Relative spectral abundance of SAGA and spliceosomal subunits (rows) expressed as dNSAF (distributive normalized spectral abundance factor) in tandem FLAG-HA purifications from S2 cells using U2B, SF3B5, Ada2B-PB, Spt3, Spt20, ATXN7, Ada1, SAF6, Sgf29 and WDA as bait proteins (columns), relative to control purifications from untagged S2 cells (S2 neg) or S2 cells expressing non-specific tagged protein CG6459.

**Table S2. Genotypes used in this study**

| **Name** | **Source** | **Genotype** |
|---|---|---|
| *UAS-SF3B5* | this study | w; P{$w^{+mC}$=UAS-SF3B5}attP40 |
| *sf3b5*$^{EY12579}$ | BL21381[a] | $y^1$ $w^{67c23}$;; P{$w^{+mC}$ $y^{+mDint2}$=EPgy2}CG11985$^{EY12579}$/TM3, $Sb^1$ $Ser^1$ |
| *UAS-SF3B5; sf3b5* | this study | w; P{$w^{+mC}$ =UAS-SF3B5}attP40; P{$w^{+mC}$ $y^{+mDint2}$ = EPgy2}CG11985$^{EY12579}$/ MKRS |
| *Act-GAL4/CyO; sf3b5* | this study | w; P{$w^{+mC}$=Act5C-GAL4} 25FO1, P{$w^{+mC}$ = UAS-GFP.nls}14/ CyO; P{$w^{+mC}$ $y^{+mDint2}$ = EPgy2} CG11985$^{EY12579}$/ MKRS |
| *ey-Gal4, UAS-FLP; FRT82B, GMR-Hid* | BL5253 | $y^1$ $w^*$; P{$w^{+mC}$=GAL4-ey.H}3-8, P{$w^{+mC}$=UAS-FLP1.D}JD1; P{$ry^{+t7.2}$=neoFRT}82B, P{$w^{+mC}$=GMR-hid} SS4, l(3)CL-$R^1$/ TM2 |
| *FRT82B, Ubi-GFP* | BL32655 | $w^{1118}$;; P{$ry^{+t7.2}$ = neoFRT} 82B P{$w^{+mC}$=Ubi-GFP(S65T)nls}3R/TM6B, $Tb^1$ |
| *FRT82B, ada2B* | Dr. Mattias Mannervik (Qi et al. 2004) | $ada2B^1$, P{$ry^{+t7.2}$=neoFRT}82B/ TM3 $Sb^1$ $Ser^1$ |
| *sf3b5* | this study | $w^{1118}$;; P{$ry^{+t7.2}$=neoFRT}82B, P{$w^{+mC}$ $y^{+mDint2}$=EPgy2}CG11985$^{EY12579}$/TM6b $Tb^1$ |
| *nonstop* | BL11553 (Weake et al. 2008) | P{$ry^{+t7.2}$=PZ}$not^{02069}$ $ry^{506}$/TM3, P{$w^{+mC}$=GAL4-twi.G} 2.3, P{UAS-2xEGFP}AH2.3, $Sb^1$ $Ser^1$ |
| *sf3b5* |  | $y^1$ $w^{67c23}$;; P{$w^{+mC}$ $y^{+mDint2}$ = EPgy2} CG11985$^{EY12579}$/ TM3, P{$w^{+mC}$=GAL4-twi.G} 2.3, P{UAS-2xEGFP} AH2.3, $Sb^1$ $Ser^1$ |
| *wda* | Dr. Susan Abmayr (Guelman et al. 2006) | $wda^{11}$/TM3, P{$w^{+mC}$=GAL4-twi.G}2.3, P{UAS-2xEGFP}AH2.3, $Sb^1$ $Ser^1$ |

[a] Bloomington *Drosophila* Stock Center at Indiana University.

| | | Transcript Normalized to Rpl32 (Fig. 5b) | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | SF3B5 | WDA | Sas10 | Oda | Sap47 | Exba | Crc | CG5390 | Gp150 |
| | ANOVA (p-values) | 0.000005 | 0.000003 | 0.005600 | 0.064000 | 0.000011 | 0.000037 | 0.098100 | 0.025700 | 0.011500 |
| Tukey HSD p-values | WT v *sf3b5* | 0.001005 | 0.032619 | 0.024560 | 0.501303 | 0.001005 | 0.001005 | 0.098798 | 0.899995 | 0.260234 |
| | WT v *wda* | 0.170948 | 0.001005 | 0.005750 | 0.053814 | 0.001005 | 0.001005 | 0.217658 | 0.033333 | 0.009150 |
| | *sf3b5* v *wda* | 0.001005 | 0.001005 | 0.612784 | 0.304091 | 0.004069 | 0.157706 | 0.849764 | 0.053178 | 0.123617 |

| | Unspliced/Spliced Transcript Normalized to Rpl32 (Fig. 5c) | | | |
|---|---|---|---|---|
| | Oda | Sap47 | exba | Crc |
| t-test (p-values) | 0.939839 | 0.006769 | 0.496777 | 0.892386 |

| | Transcript Normalized to Rpl32 : WT v *sf3b5* (Fig 5d) | | | | |
|---|---|---|---|---|---|
| | SF3B5 | nonstop | e(y)2 | Atxn7 | sgf11 |
| t-test (p-values) | 0.000760 | 0.241680 | 0.102470 | 0.727390 | 0.094710 |

**Table S3: Probability of difference between wild-type and mutant transcript levels.** Shown are the *p*-values from either ANOVA with a Tukey HSD test or Student's t-test for data shown in Fig. 5b, 5c and 5d respectively. These *p*-values were analyzed using transcript levels based on a standard curve per primer set after being normalized to Rpl32. For each test, transcript levels/Rpl32 of 4 biological replicates per genotype (OregonR, *sf3b5* and *wda*) were used for each gene (columns). For the top panel, ANOVA was used to analyze all three data sets (top row), and post hoc test (Tukey HSD) was done for each pairwise combination between genotypes (bottom three rows) where OregonR is wild type (WT) for each gene (columns). For Fig 5c and 5d, student's t-test was performed comparing wild type and *sf3b5* mutants for each gene shown (columns). For all panels, numbers highlighted in red have *p*-values <0.05 suggesting a significant difference between the data sets for those genes.