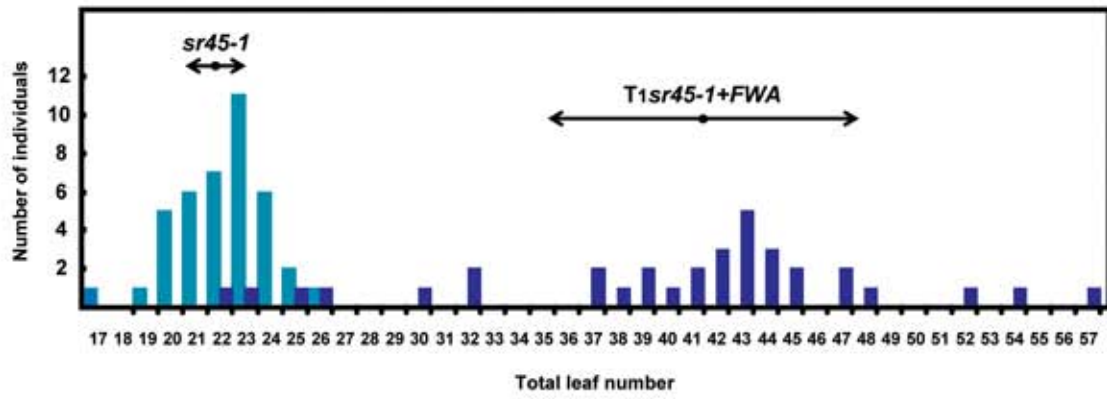
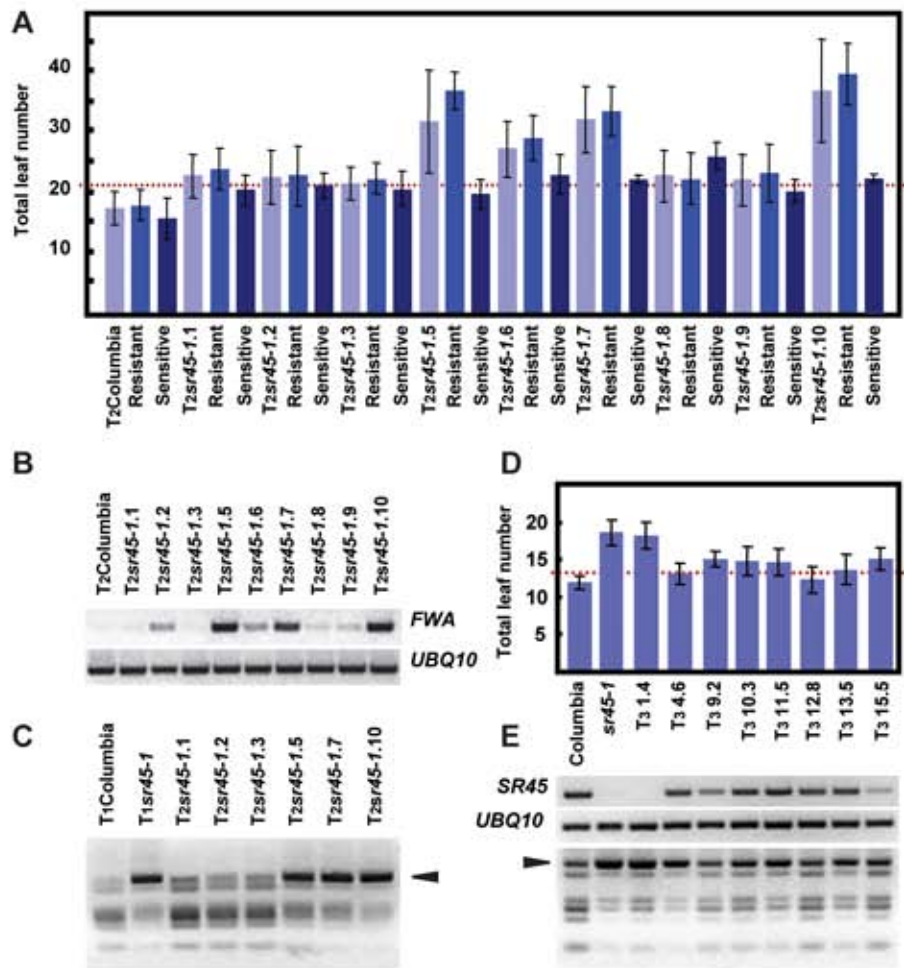


**Supplemental material to:**

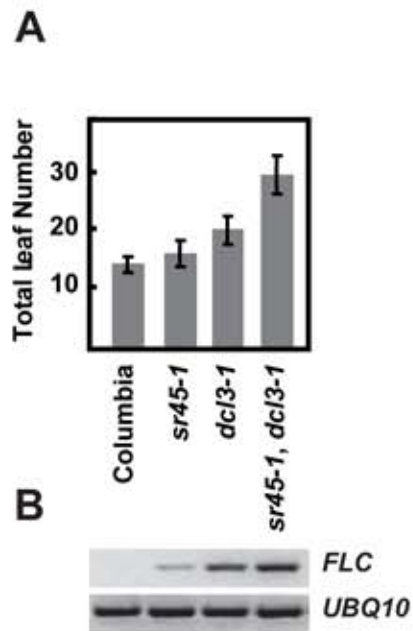
Israel Ausin, Maxim V.C. Greenberg, Carey Fei Li and Steven E. Jacobsen. *Epigenetics* 2012; 7(1); <http://dx.doi.org/10.4161/epi.7.1.18782>  
<http://www.landesbioscience.com/journals/epigenetics/article/18782>



Supplementary Figure 1. Flowering time distribution of *sr45-1* versus *sr45-1+FWA*



**Supplementary Figure 2. Confirmation of *sr45-1* de novo phenotype.** (A) Flowering time of randomly selected T<sub>2</sub> *sr45-1*+*FWA* transformants. *FWA* construct has a Basta® resistance gene as a selection marker that allows testing for the presence of *FWA* construct. Red-dotted line depicts the flowering time of untransformed *sr45-1* mutants grown under the same conditions. (B) RT-PCR showing *FWA* expression of a selection of the above-mentioned lines. *UBQ10* expression is shown as a loading control. (C) Bisulfite cutting assay, showing *FWA* methylation status in the above-mentioned lines. Genomic DNA is digested with *Bgl*III to destroy the endogenous *FWA* gene before bisulfite treatment. DNA methylation of transgenic *FWA* was assayed by PCR from bisulfite-treated DNA followed by *Cla*I digestion. CG methylation protects the *Cla*I site from bisulfite conversion. Black arrow indicates the unmethylated size. (D) Flowering time of homozygous T<sub>3</sub> *sr45-1*+*SR45* complemented lines after *FWA* transformation. (E) RT-PCR and bisulfite cutting assay showing *SR45* expression and partial restoration of methylation at *FWA*. *UBQ10* expression is shown as a loading control.



**Supplementary Figure 3. *FLC* de-repression enhancement. (A)** Flowering time of Columbia, *sr45-1*, *dcl3-1* and *sr45-1, dcl3-1* double mutant. **(B)** RT-PCR showing the expression of *FLC* in the above-mentioned lines. *UBQ10* expression is shown as a loading control.

**Supplementary Table 1: A list of known or putative spliceosome factors screened in this study.**

<b>Line</b>	<b>Insertion</b>	<b>AGI</b>	<b>Annotation</b>
99	SALK_034824	At5g37370	pre-mRNA splicing factor PRP38 family protein (SRL1)
114	SALK_040864	At1g55310	SC35-like splicing factor, 33 kD (SCL33)
118	SALK_041849	At3g13570	SC35-like splicing factor, 30a kD (SCL30a)
159	SALK_059236	At2g33440	splicing factor family protein
167	SALK_062177	At5g51300	splicing factor-related
288	SALK_125057	At1g03140	splicing factor Prp18 family protein
299	SALK_130919	At3g45950	splicing factor-related
336	SALK_147534	At3g55930	RNA splicing factor-related
357	SALK_004132	At1g16610	arginine/serine-rich protein, putative (SR45)
367	SALK_007933	At4g03430	pre-mRNA splicing factor-related
453	SALK_035759	At2g29210	splicing factor PWI domain-containing protein
518	SALK_055030	At1g60200	splicing factor PWI domain-containing protein / RNA recognition motif (RRM)-containing protein
549	SALK_063749	At4g38780	splicing factor, putative
562	SALK_069027	At5g37370	pre-mRNA splicing factor PRP38 family protein (SRL1)
589	SALK_085295	At4g38780	splicing factor, putative
591	SALK_086134	At3g06455	splicing factor-related
620	SALK_106715	At1g60200	splicing factor PWI domain-containing protein / RNA recognition motif (RRM)-containing protein
640	SALK_121879	At3g25440	group II intron splicing factor CRS1-related
662	SALK_132986	At1g09140	SF2/ASF-like splicing modulator (SRP30)
666	SALK_135024	At2g18510	pre-mRNA splicing factor, putative
667	SALK_135314	At2g29210	splicing factor PWI domain-containing protein
668	SALK_135626	At5g64270	splicing factor, putative
704	SALK_149863	At2g33440	splicing factor family protein
864	WiscDsLox289_292D17	At1g80070	splicing factor, putative
880	WiscDsLox384B10	At5g18810	SC35-like splicing factor, 28 kD (SCL28)
898	WiscDsLox246B03	At5g64200	arginine/serine-rich splicing factor SC35
899	WiscDsLox247E04	At4g02430	pre-mRNA splicing factor, putative / SR1 protein, putative
914	WiscDsLox382G12	At4g25500	arginine/serine-rich splicing factor RSP40 (RSP40)
916	WiscDsLox384G9	At4g13070	group II intron splicing factor CRS1-related

**Supplementary Table 2. Primers used in this study**

Primer	Sequence 5' to 3'
5S siRNA Probe	ATGCCAAGTTTGGCCTCACGGTCT
AGO4 RT-PCR and Northern	CAGTGCCATTTCTGTTGTTGC
AGO4 RT-PCR and Northern	TGGCGACGTTGTCTTTGAGT
<i>AtSN1</i> Chop-qPCR	TTTAAACATAAGAAGAAGTTCCTTTTTTCATCTAC
<i>AtSN1</i> Chop-qPCR	ACTTAATTAGCACTCAAATTAACAAAATAAGT
<i>AtSN1</i> siRNA Probe	ACCAACGTGTTGTTGGCCCAGTGGTAAATCTCTCAGATAGAGG
<i>FLC</i> RT-PCR	TGTGGATAGCAAGCTTGTGG
<i>FLC</i> RT-PCR	TAGTCACGGAGAGGGCAGTC
<i>FWA</i> Bisulfite	GGTTTTATATTAATATTAAGAGTTATGGGTYGAAGTTT
<i>FWA</i> Bisulfite	AACCAAATCATTCTCTAAACAAAATATAAAAAAATC
<i>FWA</i> Bisulfite Cutting	GGTTTTATTTAATGTAAATATGTATTTGATGTATT
<i>FWA</i> Bisulfite Cutting	CTAAATTAATATAACRTAATATAACATTATA
<i>FWA</i> RT-PCR	TAGTCCAGGATTGTCTGCAAAAGG
<i>FWA</i> RT-PCR	CCATTATTAACGATTTTCAGAAGAGAGA
<i>MEA-ISR</i> Bisulfite	AAAGTGGTTGTAGTTTATGAAAGGTTTTAT
<i>MEA-ISR</i> Bisulfite	CTTAAAAAATTTCAACTCATTTTTAAAAAA
<i>MEA-ISR</i> Southern Probe	AAACCTTTCGTAAGCTACAGCCACTTTGTT
<i>MEA-ISR</i> Southern Probe	TCGGATTGGTTCTTCCTACCTCTTTACCTT
<i>miR159</i> miRNA Probe	TAGAGCTCCCTTCAATCCAAA
<i>miR163</i> miRNA Probe	ATCGAAGTTGGAAGTCCTCTTCAA
<i>REP12</i> Southern probe	TCCTCTTCTCCCTCCTCTCCC
<i>REP12</i> Southern probe	TCCCACCAAGAAGCACACGC
<i>siR02</i> siRNA Probe	GTTGACCAGTCCGCCAGCCGAT
<i>Ta3</i> Southern Probe	GATCTATCTGGCCCCAGACGTAGATCTAA
<i>Ta3</i> Southern Probe	CCGGCAATCTACTATATGAGATCTTTACAA
<i>UBQ10</i> RT-PCR and Northern	GATCTTTGCCGGAAAACAATTGGAGG
<i>UBQ10</i> RT-PCR and Northern	CGACTTGTCAATTAGAAAGAAAGAGAT