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| **Table S14: Flowering genes affected in the *prp4ka* mutant.** For more details, see Tables S4-S9. A3: alternative 3’ splice site; A5: alternative 5’ splice site; EI: exitron; ES: exon skipping; IR: intron retention. | | | | | |
| **Common name** | **Gene** | **Differential gene expression** | **Differential alternative splicing** | **Gain/loss of phosphorylation** | **Description** |
| AGL8/FRUITFULL | AT5G60910 | down-regulated | - | - | The MADS-box gene FRUITFULL, like SOC1, acts to promote flowering (Gu et al., 1998).  Down-regulation of *FRUITFULL* in the *prp4ka* mutant fits the delayed flowering phenotype. |
| AGL19 | AT4G22950 | up-regulated | - | - | AGL19 is able to promote flowering following vernalization independent of FLC. The expression of *AGL19* is maintained at low levels by the Polycomb-group proteins MSI1, CLF, and EMF2 (Schönrock et al., 2006). |
| AGL20/SOC1 | AT2G45660 | down-regulated | - | - | The MADS-box gene *AGL20*, also known as *SUPPRESSOR OF OVEREXPRESSION OF CONSTANS1* (*SOC1*), is critical to the control of flowering time and acts as a floral integrator by integrating signals from the photoperiod, vernalization, and autonomous floral induction pathways (Lee et al., 2000).  *SOC1* is down-regulated in *prp4ka* mutant, fitting with the delayed flowering phenotype. |
| AGL24 | AT4G24540 | down-regulated | - | - | The MADS-box gene *AGL24* is an activator of flowering in response to vernalization and *agl24* mutants are late-flowering. SOC1 and AGL24 can up-regulate each other’s expression, however, AGL24 acts in an FLC-independent manner (Michaels et al., 2003).  Like *FUL* and *SOC1*, *AGL24* is down-regulated in the *prp4ka* mutant, conforming to the late-flowering phenotype. |
| BRN1 | AT4G03110 | up-regulated | - | - | Bruno-like proteins are able to regulate flowering time by post-transcriptional regulation of *SOC1*, via binding to the 3’ UTR of *SOC1* (Kim et al., 2013).  *BRN1* is up-regulated in *prp4ka.* For *BRN2*, the intron retention AT1G03457.s1 isoform is higher expressed, whereas the protein-coding AT1G03457.P1 and AT1G03457.P2 isoforms are expressed at the wild-type levels. Function of the *BRN2* intron retention isoform is not known. Isoform IDs are according to the Arabidopsis reference transcriptome AtRTD2 (Zhang et al., 2017). |
| BRN2 | AT1G03457 | - | IR | - |
| BZR1 | AT1G75080 | - | - | loss | BRASSINAZOLE-RESISTANT1 (BZR1) binds to a putative brassinosteroid response cis-element and suppresses the expression of the autonomous pathway component *FLOWERING LOCUS D* (*FLD*). BZR1 is functionally regulated by phosphorylation (Zhang et al., 2013).  BZR1 loses phosphorylation in the *prp4ka* mutant, indicating that PRP4KA might play a role in BZR1 phosphorylation and therefore *FLD* regulation. |
| EDM2 | AT5G55390 | - | A3, IR | - | EDM2 acts as an upstream repressor of FLC by regulating its transcript levels, thereby promoting flowering (Tsuchiya and Eulgem, 2010).  The three isoforms of *EDM2* are alternatively spliced in the 5’ UTR and all potentially code for the full-length protein. The intron retention isoform AT5G55390.P1 is down-regulated in the *prp4ka* mutant, whereas the expression of the remaining two isoforms is slightly increased, resulting in equal gene expression in both the *prp4ka* mutant and wild-type plants. |
| EMF2 | AT5G51230 | - | IR | - | The Polycomb repressive complex 2 subunits CURLY LEAF, EMBRYONIC FLOWER 2, and FERTILIZATION INDEPENDENT ENDOSPERMrepress the expression of members of the *FLC*/*MAF* gene family (Jiang et al., 2008).  The intron retained in AT5G51230.ID14 isoform, resulting into the introduction of a PTC, shows a fivefold increase in expression compared to the wild-type, whereas the expression of the full-length protein-coding AT5G51230.P1 isoform slightly drops. However, the AT5G51230.P1 is still ~1.5 higher expressed than the intron retention isoform. Isoform IDs are according to the Arabidopsis reference transcriptome AtRTD2 (Zhang et al., 2017). |
| FKF1 | AT1G68050 | down-regulated | - | - | A *fkf1* mutant showed delayed flowering, both in days to flowering or leaf number at flowering, compared to the wild-type plant (Nelson et al., 2000).  *FKF1* is down-regulated in the *prp4ka* mutant to two-thirds of wild-type levels. |
| FLC | AT5G10140 | up-regulated | - | - | The MADS-box gene FLOWERING LOCUS C acts as a floral repressor to delay flowering via the repression of the floral integrators *FLOWERING TIME* (*FT*) and *SOC1* (Alexandre and Hennig, 2008).  The *prp4ka* mutant shows both up-regulation of *FLC* and down-regulation of its direct target *SOC1*, conforming to the delayed flowering phenotype of the *prp4ka* mutant plants. |
| FLK | AT3G04610 | up-regulated | IR x 2 | gain | The autonomous pathway component FLOWERING LOCUS KH DOMAIN (FLK) is one of the suppressors of *FLC* (Kim et al., 2004).  FLK is up-regulated in *prp4ka*, mainly due to the more than seven times up-regulation of the AT3G04610.P2 isoform. This isoform has a retained intron in the 5’ UTR. Retention of this intron reduces levels of FLK protein and leads to the up-regulation of FLC (Deng et al., 2010). |
| FLL4 | AT5G61920 | - | IR x 2 | - | FLOWERING LOCUS C EXPRESSOR-LIKE 4 (FLL4) is essential for the up-regulation of FLC (Lee and Amasino 2013).  The introns retained in AT5G61920.P2 and AT5G61920.P6 isoforms exhibit a slight but significant increase in *prp4ka*. Both retained introns are located in the 5’ UTR. Their effect on FLL4 protein levels is not known. |
| HUA2 | AT5G23150 | - | IR | - | HUA2 acts as a repressor of floral transition. *hua2* mutations reduce the expression of several floral repressors, including *FLC* and *MAF1/FLM* (Doyle et al., 2005).  The two *HUA2* full-length protein-coding isoforms (AT5G23150.P1 and AT5G23150.P3) are expressed at wild-type levels. The expression of the intron retained AT5G23150.P2 isoform increases to roughly the same levels as the full-length protein-coding isoforms. |
| LOV1 | AT2G02450 | - | EI | - | The *LONG VEGETATIVE PHASE 1* (*LOV1*) gene controls flowering time. An Arabidopsis mutant, *lov1-1D*, showed a late-flowering phenotype (Yoo et al., 2007).  In *prp4ka*, the expression of the exitron-spliced AT2G02450.P1 isoform is reduced, whereas the expression of the exitron-containing AT2G02450.P2 isoform remains the same. Exitron (exonic intron) is an alternatively spliced internal region of a reference protein-coding exon (Marquez et al., 2015). |
| MAF1/FLM | AT1G77080 | up-regulated | A3, A5, EI, RI x 4 | - | The Arabidopsis genome contains five MADS box genes coding for proteins highly related to *FLC*, *MADS AFFECTING FLOWERING 1* to *5* (*MAF1-MAF5*). MAF1, also known as FLOWERING LOCUS M, is a flowering inhibitor acting independently and with different flowering pathways than FLC (Scortecci et al., 2003). MAF2 to MAF5 have also been shown to inhibit flowering (Ratcliffe et al., 2003).  *MAF1/FLM*, *MAF4*, and *MAF5* are all up-regulated in *prp4ka*, which shows a delayed flowering phenotype. |
| MAF4 | AT5G65070 | up-regulated | - | - |
| MAF5 | AT5G65080 | up-regulated | IR | - |
| SPL9 | AT2G42200 | - | IR | - | Several members of the *SQUAMOSA PROMOTER BINDING LIKE* (*SPL*)gene family act in floral transition. The *SPL3*, *SPL4*, *SPL5* and *SPL9* factors directly bind to and activate transcription of *SOC1*, *APETALA1*, *LEAFY*, and *FRUITFULL* (Wang et al., 2009l; Yamaguchi et al., 2009).  In the *prp4ka* mutant, expression is slightly but significantly shifted away from the fully-spliced protein-coding AT2G42200.P1 isoform, towards the intron retention AT2G42200.JS1 isoform. Isoform IDs are according to the Arabidopsis reference transcriptome AtRTD2 (Zhang et al., 2017). |
| VIP5 | AT1G61040 | - | A5 x 2 | - | The *VERNALIZATION INDEPENDENCE* (*VIP*) gene class represses flowering, by activation of the *FLC/MAF* gene family. Loss of function of the *VIP5* and *VIP6* results in downregulation of the *FLC/MAF* family genes (Oh et al., 2005).  *VIP5* has three protein-coding isoforms (AT1G61040.P1-3), and the significantly changing A5 events occur in the 5’ UTR. The expression of the AT1G61040.P3 isoform is down-regulated in *prp4ka*, whereas the expression of the other two transcripts is slightly increased. |

Inspection of the DEG, DAS, and phosphorylation data sets revealed that a number of flowering-related genes are affected in the *prp4ka* mutant. Most notably, the floral repressor *FLC*, and its homologs *FLM*, *MAF4*, and *MAF5*, were all up-regulated in the *prp4ka* mutant. Conversely, the floral promoters *FUL*, *SOC1* and *AGL24* were all down regulated (**Table S5**). *FLK*, a suppressor of *FLC*, was up-regulated in the *prp4ka* mutant (**Table S5**), mainly due to the more than seven times up-regulation of the AT3G04610.P2 isoform with a retained intron in the 5’ UTR (**Table S6**). Retention of this intron reduces levels of FLK protein and leads to the up-regulation of *FLC* (Deng et al., 2010). The autonomous pathway flowering time locus FVE, which epigenetically represses *FLC* transcription loses phosphorylation in the *prp4ka* mutant (**Table S10**). FLC delays flowering by repressing SOC1 (Alexandre and Hennig, 2008), whereas FLM inhibits flowering in a FLC-independent manner (Scortecci et al., 2003). MAF4 and MAF5 have also been shown to inhibit flowering (Ratcliffe et al., 2003). SOC1 integrates signals from the photoperiod, vernalization, and autonomous floral induction pathways and is critical to the control of flowering time (Lee et al., 2000). Like SOC1, FUL and AGL24 act to promote flowering, whereby SOC1 and AGL24 can up-regulate each other’s expression (Gu et al., 1998; Michaels et al., 2003).

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