Antagonistic Regulation of Flowering Time through Distinct Regulatory Subunits of Protein Phosphatase 2A

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Abstract

Protein phosphatase 2A (PP2A) consists of three types of subunits: a catalytic (C), a scaffolding (A), and a regulatory (B) subunit. In Arabidopsis thaliana and other organisms the regulatory B subunits are divided into at least three non-related groups, B55, B' and B". Flowering time in plants mutated in B55 or B" genes were investigated in this work. The PP2A-b55α and PP2A-b55β (knockdown) lines showed earlier flowering than WT, whereas a PP2A-b'γ (knockdown) line showed late flowering. Average advancements of flowering in PP2A-b55 mutants were 3.4 days in continuous light, 6.6 days in 12 h days, and 8.2 days in 8 h days. Average delays in the PP2A-b’γ mutant line were 7.1 days in 16 h days and 4.7 days in 8 h days. Expression of marker genes of genetically distinct flowering pathways (CO, FLC, MYB33, SPL3), and the floral integrator (FT, SOC1) were tested in WT, pp2a mutants, and two known flowering time mutants elf6 and edm2. The results are compatible with B55 acting at and/or downstream of the floral integrator, in a non-identified pathway. B’ γ was involved in repression of FLC, the main flowering repressor gene. For B’ γ the results are consistent with the subunit being a component in the major autonomous flowering pathway. In conclusion PP2A is both a positive and negative regulator of flowering time, depending on the type of regulatory subunit involved.

Introduction

Protein phosphatase 2A (PP2A) is conserved among eukaryotes, and is vital for growth and development, but very little is known concerning functions of specific subunits of PP2A in plants [1–5]. PP2A complexes are composed of three different types of proteins; a catalytic (C), a scaffolding (A), and a regulatory (B) subunit. In Arabidopsis, at least seventeen regulatory B subunits are present, and these subunits are believed to be responsible for substrate specificity and cellular localization of the PP2A complexes, hence largely account for the diverse functions of PP2A [1,6–10]. In plants, the B subunits are divided into three main, non-related subgroups: B55 (also called B), B’, and B". In Arabidopsis, the B55 subgroup consists of only two members, Bα and Bβ. Our recent work related to nitrogen metabolism had brought to our attention that although the single mutant lines looked normal, these two genes are essential for survival because the pp2a-bα x pp2a-bβ double knockout was embryo lethal [11]. We decided to further study these genes in relation to growth and development, especially flowering time. Experiments with the pp2a-bα and pp2a-bβ mutants soon revealed that these mutants were early flowering. We had also noticed in introductory flowering time experiments that some pp2a mutants were late flowering. Mutants of the B' η subfamily i.e. b’ γ, b’ η [6,12] and b’ θ (unpublished data) were late flowering. Since b’ γ had the most striking phenotype in respect to flowering time, this mutant was chosen to be included in the more detailed studies to reveal underlying molecular mechanisms. B'γ provides a link between developmental regulation and and stress signaling because B’ γ also plays a key role in controlling the extent of defence reactions against different types of plant pathogens [10,12].

So far four major flowering time signalling pathways are acknowledged: the photoperiod, autonomous, vernalization, and gibberellin pathway (Figure 1) [13–15]. A more complete picture would include also pathways from ambient temperature, nitrate status, and age signals [16,17]. Genes named here refer to Arabidopsis, but orthologous are generally found in other plants studied [18–20]. The photoperiod pathway regulates flowering time in response to external signals, especially length of the photoperiod. This pathway involves for example the...
photoreceptors phytochrome and cryptochrome, clock genes, and further downstream the zinc-finger transcription factor CO (CONSTANS). CO promotes flowering through FT (FLOWERING LOCUS T) and SOC1 (SUPPRESSOR OF OVEREXPRESSION OF CONSTANS 1). FT together with FD (a bZIP transcription factor) and SOC1 up-regulate various genes, including AP1 (APETALA1), and change the vegetative meristem (VM) into an inflorescence meristem (IM) that in turn rise to a floral meristem (FM) [17]. Other pathways responding to environmental signals are the vernalization pathway and ambient temperature pathway. In the vernalization pathway, FLC (FLOWERING LOCUS C), the main flowering repressor gene, is inactivated by chromatin modifications in response to enduring cold periods. The requirement for vernalization in the “lab strains” like Columbia is, however, abolished due to a mutation in another gene (FRIGIDA) [17]. FLC also has a central role in the autonomous pathway, but external signals are not important for induction of flowering in the pathways termed autonomous [13]. A positive effect of gibberellin on flowering is seen especially in short day plants and biennial

Figure 1. A schematic model for involvement of PP2A in flowering time pathways in Arabidopsis. Expression of key genes of each pathway, CO, FLC, and MYB33 as well as FT and SOC1 of the floral integrator were tested. SPL3 and miR156, which can influence flowering by an endogenous pathway acting downstream of the floral integrator were also tested. ELF6 and EDM2 genes are known to delay and advance flowering, respectively. Mutants (knockout) lines of elf6 and edm2 were included as control lines. Genes (transcripts) tested in this work are shown in blue. Genes mutated in Arabidopsis lines tested in this work are shown in red. The work showed that PP2A-B55 was a negative regulator of flowering time possibly acting downstream of/at the floral integrator, whereas PP2A-Bγ was a positive component in flower induction acting through modulation of FLC expression.

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Table 1. Growth of WT, PP2A-b55α, and PP2A-b55β.

| Genotype    | 12 h photoperiod | 24 h photoperiod |
|-------------|------------------|------------------|
|             | Number of leaves | mean leaf weight | Number of leaves | mean leaf weight |
| WT          | 7.7 ± 0.6        | 0.13 ± 0.02      | 7.9 ± 0.7        | 0.18 ± 0.04      |
| pp2a-b55α   | 7.2 ± 0.6        | 0.12 ± 0.02      | 7.2 ± 0.5        | 0.13 ± 0.02      |
| SALK_09504  |                 |                  |                 |                  |
| pp2a-b55β   | 7.2 ± 0.6        | 0.13 ± 0.02      | 7.1 ± 0.6        | 0.18 ± 0.02      |
| SALK_062614 |                 |                  |                 |                  |

Number of leaves per plant 16 days after germination, and mean leaf fresh weight 21 days after germination. For each genotype and treatment 45 plants were scored. SE is given.

Materials and Methods

Plant material and growth

T-DNA insertion lines provided by SALK [31] and GABI-Kat [32] were obtained from NASC (The European Arabidopsis Stock Centre in Nottingham, UK). Arabidopsis mutant lines for \( ba \) (AT1g51690) were SALK_032080 (insertion in second exon) and SALK_095004 (insertion in sixth intron). Mutant lines for \( b' \) (AT1g17720) were SALK_062514 (insertion in sixth intron) and GK_290G04 (insertion in fifth exon). Mutant line for \( b'' \) were SALK_039172 (insertion in 5' UTR) and a pp2abγ line complemented by 35S-driven expression of the PP2A-Bγ gene (in SALK_039172 background) [12].

Mutant selection was done by PCR using primers for T-DNA insertion lines recommended by SALK institute website SIGnAL (http://signal.salk.edu/tdnaprimer2.html). Homozygous mutants were verified by PCR using gene specific primers [11]. Known flowering time mutants used as controls were edmi2 (AT5g55390, SALK_014520C) and elf6-3 (AT5g04240, SALK_074694C). Seeds were sown in a regular soil mixture, stratified at 4°C for 2 days, and then transferred to growth chambers with 8 h/16 h, 12 h/12 h, 16 h/8 h light/darkness or continuous light.

For testing of expression levels of flowering regulatory genes, seeds were stratified for 4 days at 4°C before placed in a 16 h light/8 h dark regimen. Shoots were harvested 10 days after germination, and generally 12 h into the photoperiod. In one experiment shoots were also harvested 8 h into the photoperiod, which confirmed the results.

Phenotyping

Plants were observed daily. Number of rosette leaves and flowering time were recorded. Flowering time was measured using two different time points: first, appearance of the floral bud (DTF2) as indicator of transition from vegetative to inflorescence meristem, and second, appearance of first open flower (DTF2) as indicator of transition from inflorescence meristem to floral meristem [33,34]. Fresh weight of leaves was measured 21 days after germination to assure that observed phenotype is not simply due to altered growth rate in mutant plants. Characterizing of flowering phenotypes was repeated at least three times and in successive generations for each mutant line to assure that observations are repeatable and phenotypes are stable during generations.

qRT-PCR

Quantitative reverse transcriptase real time PCR (qRT-PCR) was performed as previously described [35]. Total RNA was isolated using RNeasy® Plant Mini Kit (Qiagen, Chatsworth, CA), and cDNA synthesised using the High Capacity cDNA Archive Kit (Applied Biosystems). MicroRNA was isolated using mirVana miRNA isolation Kit (Invitrogen) and cDNA was made using the TaqMan MicroRNA reverse transcription kit (Applied Biosystems). Real-time PCR reactions were assayed using an ABI 7300 Fast Real-Time PCR System. The reaction volume was 25 µL containing 12.5 µL TaqMan buffer (Applied Biosystems, includes ROX as a passive reference dye), 8.75 µL H₂O, 2.5 µL cDNA and 1.25 µL primers. Primers were
predesigned TaqMan® Gene Expressions assays obtained for the following Arabidopsis thaliana genes, (Applied Biosystems identification number is given in parenthesis): FT At1g65480 (At02224075), FLC At5g10140, (At02272498), MYB33 At5g06100 (At02337117), SOC1/AGL20 AT2g45660 (At02263356), CO At5g115840, (At02200179), SPL At2g33810 (At02204412), MIR156A AT2g25095 (Assay ID000333). The Taq Man assay is based on a light signal from each transcript copy formed and allows comparing expression levels between the various genes. Standard cycling conditions (2 min at 50° C, 10 min at 95° C and 40 cycles altering between 15 s at 95° C and 1 min at 60° C) were used for product formation. Real-time PCR products were analyzed by Sequence Detection Software version 1.3. Comparative CT method for relative quantification has been used with ubiquitin At3g02540 (At02163241_g1), ACT8 At1g49240 (At02270958) and SnoR85 (Assay id 0017111) as endogenous controls. Relative quantity (RQ=2−ΔΔCT) of any gene in mutant lines was calculated relative to WT (calibrator). Expression levels are given as per cent of WT, which was set to 100%.
Results

Expression analysis had previously verified that transcripts of PP2A-B55α and PP2A-B55β genes were not detectable in the respective mutants [11] and transcript level of PP2A-B′γ was low in the pp2a-b′γ mutant plants [12]. The PP2A-B′γ complementation line contains variable levels of mRNA (2.2-fold ± 1.0-fold compared with wild-type levels) [12]. Plants were first grown in 12 h days or continuous light to test number of days to flowering (Figure 2a, b). Averaged across all four pp2a-b55 mutant lines in 12 h days, the first bud appeared 5.2 days, and the first flower 6.8 days before WT. In continuous light the first bud appeared 2.5 days and the first flower 3.4 days before WT. pp2a-b55 mutant lines were also observed in 8 and 16 h days, where the first flower appeared 8.2 and 4 days before WT, respectively (Figure 2c). The mutant line pp2a-b′γ was observed in 8 h and 16 h days, and showed that the first flower appeared 7.1 (in 8 h days) and 4.7 (in 16 h days) days later than in WT (Figure 2d). Early or late floral transition is reflected also by the formation of lower or higher number of rosette leaves prior to bolting. Compared with WT, pp2a-b55 plants formed nearly four and six rosette leaves less before bolting, under long and short day conditions, respectively (Figure 2c). In contrast, plants mutated in pp2a-b′γ formed nearly seven and five more rosette leaves before bolting compared with WT, under long and short day conditions (Figure 2d). In conclusion all pp2a-b55 mutant lines tested were early flowering, and the pp2a-b′γ mutant line was late flowering under all conditions. The pp2a-b55 mutants showed growth very similar to WT. In 12 h days or continuous light the number of leaves was the same for mutant lines and WT 21 days after sowing. In 12 h days, fresh weight of leaves was also nearly identical for mutants and WT. The pp2a-ba line showed lower fresh weight in continuous light compared with WT and pp2a-bβ (Table 1).

The results imply that the early flowering of the pp2a-b55 mutants is not caused simply by increased growth. On the contrary, these mutants showed same or slower growth compared with WT, hence the criteria for qualifying as a real flowering time mutant are strengthened [13]. Figure 3 illustrates the early flowering phenotype of pp2a-b55 and late flowering phenotype of pp2a-b′γ.

Known flowering mutants were included in the experiments to assure the relevance of the expression analysis for detecting flowering time mutants in the various signalling pathways. EDM2 has a promoting effect on flowering, and acts upstream of the floral repressor FLC, in other words EDM2 represses FLC [36]. ELF6 is an inhibitor of flowering, and delays flowering through the photoperiod pathway [37] by repressing FT [38], as well as through an autonomous pathway by effects on brassinosteroid signalling components [39]. Expression analysis of these genes showed that the late flowering mutant control, edm2, had high level of FLC expression whereas the early flowering mutant control, elf6, showed low level of FLC and high level of FT expression (Figure 4a, e). Low level of FLC transcripts in the elf6 mutant was not previously reported, but a close relative to ELF6 (REF6) was found to inhibit flowering through the autonomous pathway by acting on FLC [37]. Interestingly, in our analysis using the highly specific TaqMan assays, it was clear that FLC was expressed at a low level also in the elf6 mutant (Figure 2a). For pp2a-b55α/β mutants, expression levels of FLC and CO were the same as in WT, but there was a tendency to slightly higher expression levels of the floral integrator genes FT and SOC1. A particular microRNA, miR156, is known to promote the juvenile phase in Arabidopsis and maize and inhibit flowering [13]. Recently, a new autonomous pathway was pointed out where MIR156 and SPL3 play important roles [24]. Since these genes can act downstream of FT and FD we tested SPL3 and MIR156...
Figure 4. Expression levels of genes important in different flowering time controlling pathways and the floral integrator. Shoots were harvested ten days after germination, 12 h into the 16 h photoperiod. Gene expression was tested in WT and the mutants pp2a-bα SALK_09504, pp2a-bβ SALK_062614, pp2a-bγ, elf6 (early flowering control) and edm2 (late flowering control). Genes tested were: a) FLC, b) CO, c) MYB33, d) SOC1, e) FT and f) SPL3. Expression of established flowering pathway genes are modulated in pp2a-bγ consistent with this mutant being late flowering, whereas pp2a-b55 mutants show only minor changes in transcript levels and may act on flowering time through an unknown pathway. Data presented are means of three (except for SPL3, which had two) independent experiments of samples each containing 50 plants and assayed in triplicate. Vertical bars indicate the standard error of the mean.

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expression in the pp2a mutants. However, expression of SPL3 was not significantly altered in pp2a mutants (Figure 4f).

Expression of MIR156 was also tested (data not presented), but average deviation from WT was only 30% (down-regulated). In conclusion, the SPL3/mir156 pathway could hardly explain early flowering time in pp2a-b55 mutants. Consistent with late flowering phenomena, gene expression analysis in the pp2a-b’γ mutant line showed high levels of FLC and low levels of FT and SOC1 expression compared with WT (Figure 4). Complementation of pp2a-b’γ with the PP2A-B’γ gene under control of the 35S promoter restored WT expression of FLC, FT, and SOC1 (Figure 5), and the complemented line flowered as early as WT (Figure 3b) e.g. average flowering time was 20.3 ± 0.7 and 20.0 ± 0.7 days for WT and complemented line, respectively. The results are consistent with PP2A-B’γ promoting flowering in Arabidopsis through repression of the FLC gene.

Discussion

Multilevel control of Arabidopsis flowering time by protein phosphoregulation

A high degree of phosphorylation of proteins in the photoperiodic pathway, as well as the florigen activating complex, generally has been associated with acceleration of flowering [25,27,28], e.g. high kinase/low phosphatase activity would promote flowering. Knockout of a protein phosphatase interacting with the photoperiodic pathway or florigen activating complex, could therefore lead to earlier flowering than for WT. Although their specific protein targets are still unknown, a kinase promoting flowering, CKII, as well as a kinase that delays flowering, SNF1-like AKIN10, have been identified [40,41]. The phosphorylation status of proteins is a result of opposing kinases and phosphatases, and late flowering of the pp2a-b’γ knockdown mutant would be consistent with PP2A-B’γ acting antagonistically to a kinase that delays flowering. Furthermore, PP2A plays a role in brassinosteroid signalling, and the B’γ subunit (among other B’subunits) interacts with a key component, BZR, of the signalling pathway [7]. Impairment of brassinosteroid signalling is known to result in delayed flowering [39]. Delayed flowering in pp2a-b’γ knockdown may therefore also potentially (partly) act through changes in a brassinosteroid influenced pathway. By testing expression of selected genes of the main flowering pathways we intended to clarify which pathways PP2A subunits would target.

Photoperiod, Gibberellin and Vernalization pathways

The pp2a-b55α and pp2a-b55β mutants showed early flowering compared with wild type in continuous light, 16 h, 12 h and 8 h photoperiods (Figure 2). Mutations in the genes of the photoperiod regulatory pathway make Arabidopsis unable to sense the duration of inductive long photoperiods, resulting in altered flowering time in long days, but little effect in short days [13,37]. pp2a-b55 mutants flower earlier than wild type whatever the photoperiod, but since they are responsive to photoperiod and are able to discriminate between short and long days and still flower earlier in long days than they do in short days, mutations in the PP2A-B55α or PP2A-B55β genes
are not likely to interfere specifically with the photoperiod pathway. This was also supported by the fact that expression of CO was not altered in *pp2a-b55* mutants in comparison with WT. Flowering time of the *pp2a-b*y mutant was also perturbed (delayed) in both short and long days but the mutant is also able to discriminate between short and long days which indicates that *B* y is also not a candidate for being involved in the photoperiodic pathway. The gibberellin pathway is mainly of importance in short days [21,22], which implies that the *pp2a-b55α, pp2a-b55β*, and *pp2a-b*y mutations did not interfere specifically with this pathway since the phenotypes were pronounced regardless of length of the photoperiod. Furthermore, we did not find any large changes in expression of *MYB33*, a flowering promoting gene in the gibberellin pathway [21]. The vernalization pathway is generally inactivated through the FRIGIDA mutation in the *Arabidopsis Columbia* line, therefore the vernalization pathway is not a candidate for being involved in our experiments [13].

**Autonomous pathway**

In the autonomous pathway the flowering inhibitor FLC plays an important role. In the late flowering control mutant *edm2* expression of *FLC* was clearly enhanced in agreement with the autonomous pathway being involved (Figure 4a) [36]. This confirmed that our marker genes and expression analysis would reveal flowering time mutants of this pathway. Since expression analysis of *FLC* in the *pp2a-b55a* and *pp2a-b55β* mutants did not show any deviations from expression in WT, the classical autonomous pathway is unlikely to account for early flowering of the *pp2a-b55* mutants. However, increased phosphorylation status of the FLC protein itself still represents one possible mechanism of PP2A effects and cannot be completely ruled out, although enhanced *FT* transcript level should follow inactivation of FLC. FT transcripts, however, showed only a very moderate increase in *pp2a-b55*. In the *pp2a-b*y mutant, transcript level of the FLC gene was three-fold higher in WT, which implies that PP2A-B'y plays an important role in the repression of *FLC*, and knockdown of *PP2A-B'y* results in a late flowering phenotype. The results are consistent with PP2A-B'y being a component of the autonomous pathway upstream of *FLC*. *FLC* is epigenetically regulated [13] and a PP2A scaffolding subunit was recently found to interact with histone deacetylase HDA14 [42]. In further work it will be interesting to explore if PP2A-B'y is part of a histone modifying complex.

In conclusion, based on observations with different photoperiods and the fact that the vernalization pathway is not important in the Columbia line, three of the main flowering pathways, photoperiod, vernalization, and gibberellin pathway, are not likely to be targets for PP2A-B55 or PP2A-B'y. PP2A-B55 may target components at the downstream level of floral integrators through an unknown pathway, but targeting of different pathways should not be excluded. PP2A-B*y functions in the autonomous pathways by repressing the main flowering inhibitor *FLC*. The results show that PP2A acts both as a positive and negative regulator of flowering, depending on the regulatory B subunit involved.

**Author Contributions**

Conceived and designed the experiments: BH DN SK CL. Performed the experiments: BH DN. Analyzed the data: BH DN. Wrote the manuscript: BH CL.

**References**

1. Farkas I, Dombrádi V, Miskei M, Szabados L, Koncz C (2007) Arabidopsis PPP family of serine/threonine phosphatases. Trends Plant Sci 12: 169-176. doi: 10.1016/j.tplants.2007.03.003. PubMed: 17368808.
2. Jonassen EM, Heidari B, Nemie-Feyissa D, Matre P, Lillo C (2011) Protein phosphatase 2A regulatory subunits are starting to reveal their pattern of novel protein phosphatases revealed by analysis of protein phosphatase 2A beta subfamily members. Planta 230: 935-945. doi: 10.1007/s00225-009-0998-z. PubMed: 19676260.
3. Normandeau B, Sondhi R, Qiu X, Chappell J, Wieringa B et al. (2009) Diversity in subcellular targeting of the Protein phosphatase 2A beta family subunits. Planta 230: 935-945. doi: 10.1007/s00225-009-0998-z. PubMed: 19676260.
4. Tang W, Yuan M, Wang R, Yang Y, Wang C et al. (2011) PP2A activates brassinosteroid-responsive gene expression and plant growth by dephosphorylating BZR1. Nat Cell Biol 13: 124-131. doi: 10.1038/ncb2151. PubMed: 21258370.
5. Camilleri C, Azimzadeh J, Pastuglia M, Bellini C, Grandjean O et al. (2002) The Arabidopsis TONNEAU2 gene encodes a putative novel protein phosphatase 2A regulatory subunit essential for the control of the cortical cytoskeleton. Plant Cell 14: 833-845. doi: 10.1105/tpc.010402. PubMed: 11971138.
6. Castro Marín I, Loefl I, Bartetzko L, Searle I, Coupland G et al. (2011) Nitrate regulates floral induction in Arabidopsis, acting independently of
light, gibberellin and autonomous pathways. Planta 233: 539-552. doi: 10.1007/s00425-010-1316-5. PubMed: 20434991

17. Forma F, de Montaigu A, Coupland G (2010) SnapShot: Control of flowering in Arabidopsis. Cell 141: 3: 518-528. doi: 10.1016/j.cell.2010.05.014. PubMed: 20685730.

18. Andersson CH, Jensen CS, Petersen K (2004) Similar genetic switch systems might integrate the floral inductive pathways in dicots and monocots. Trends Plant Sci 9: 105-107. doi: 10.1016/j.tplants.2004.01.002. PubMed: 15058273.

19. Higgins JA, Bailey L, Laurie DA (2010) Comparative genomics of flowering time pathways using Brachypodium distachyon as a model for the temperate grasses. PLOS ONE 5: e10065. doi: 10.1371/journal.pone.0010065. PubMed: 20419097.

20. Izawa T, Takahashi Y, Yano M (2003) Comparative biology comes into bloom: genomic and genetic comparison of flowering pathways in rice and Arabidopsis. Curr Opin Plant Biol 6: 113-120. doi: 10.1016/S1369-5266(03)00014-1. PubMed: 12667866.

21. Achar P, Herr A, Baulcombe DC, Harberd NP (2004) Modulation of floral development by a gibberellin-regulated microRNA. Development 131: 3357-3365. doi: 10.1242/dev.01206. PubMed: 15226253.

22. Gocal GF, Sheldon CC, Gubler F, Moritz T, Bagnall DJ et al. (2001) GAMYB-like genes, flowering, and gibberellin signaling in Arabidopsis. Plant Physiol 127: 1692-1693. doi: 10.1104/pp.001044. PubMed: 11743113.

23. Yang L, Conway SR, Poethig RS (2011) Vegetative phase change is mediated by a leaf-derived signal that represses the transcription of miR156. Development 138: 245-249. doi: 10.1242/dev.01206. PubMed: 21113723.

24. Wang JW, Czech B, Weigel D (2009) miR156-regulated SPL genes affect multiple developmental and stress-responsive pathways in Arabidopsis thaliana. Plant Cell 138: 738-749. doi: 10.1105/tpc.005306. PubMed: 12468722.

25. Yang L, Conway SR, Poethig RS (2011) Vegetative phase change is mediated by a leaf-derived signal that represses the transcription of miR156. Development 138: 245-249. doi: 10.1242/dev.01206. PubMed: 21113723.

26. Kishida M, Hagiwara S, Kohno F, Nishimura K, Ohnishi H et al. (2004) A pair of homologous jumonji/zinc-finger-class transcription factor genes affect multiple developmental and stress-responsive pathways in Arabidopsis. Plant J 71: 3043-3056. doi: 10.1111/j.1365-313X.2004.025353.x. PubMed: 15377760.

27. Tsai AY, Gazzarrini S (2012) AKIN10 and FUSCA3 interact to control lateral organ development and phase transitions in Arabidopsis. Plant J 71: 263-272. doi: 10.1111/j.1365-313X.2012.04984.x. PubMed: 22404109.