Isolation and functional characterization of JcFT, a FLOWERING LOCUS T (FT) homologous gene from the biofuel plant Jatropha curcas

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Chaoqiong Li1,2, Li Luo3, Qiantang Fu1, Longjian Niu1,4 and Zeng-Fu Xu1*

Abstract

Background: Physic nut (Jatropha curcas L.) is a potential feedstock for biofuel production because Jatropha oil is highly suitable for the production of the biodiesel and bio-jet fuels. However, Jatropha exhibits low seed yield as a result of unreliable and poor flowering. FLOWERING LOCUS T (FT)–like genes are important flowering regulators in higher plants. To date, the flowering genes in Jatropha have not yet been identified or characterized.

Results: To better understand the genetic control of flowering in Jatropha, an FT homolog was isolated from Jatropha and designated as JcFT. Sequence analysis and phylogenetic relationship of JcFT revealed a high sequence similarity with the FT genes of Litchi chinensis, Populus nigra and other perennial plants. JcFT was expressed in all tissues of adult plants except young leaves, with the highest expression level in female flowers. Overexpression of JcFT in Arabidopsis and Jatropha using the constitutive promoter cauliflower mosaic virus 35S or the phloem-specific promoter Arabidopsis SUCROSE TRANSPORTER 2 promoter resulted in an extremely early flowering phenotype. Furthermore, several flowering genes downstream of JcFT were up-regulated in the JcFT-overexpression transgenic plant lines.

Conclusions: JcFT may encode a florigen that acts as a key regulator in flowering pathway. This study is the first to functionally characterize a flowering gene, namely, JcFT, in the biofuel plant Jatropha.

Keywords: Biofuel, Early flowering, Florigen, FLOWERING LOCUS T, Physic nut

Background

Physic nut (Jatropha curcas L.) is a perennial plant that belongs to the Euphorbiaceae family, and is monoecious with male and female flowers borne on the same plant within the same inflorescence [1]. The potential benefit of growing Jatropha as a cash crop for biofuel in tropical and sub-tropical countries is now widely recognized [2-4]. Jatropha has been propagated as a unique and potential biodiesel plant owing to its multipurpose value, high oil content, adaptability to marginal lands in a variety of agro-climatic conditions, non-competitiveness with food production, and high biomass productivity [2,5]. The oil content of Jatropha seeds and the kernels ranges from 30% to 50% and 45% to 60% by weight, respectively. Oil from Jatropha contains high levels of polyunsaturated fatty acids, and it is therefore suitable as a fuel oil [6,7]. However, the potential of Jatropha as a biofuel plant is limited by its low seed production. Despite the clear evidence of the abundant biomass generated by Jatropha, it is not indicative of high seed productivity [8]. There are too many vegetative shoots in Jatropha, which could develop into reproductive shoots under suitable conditions. It is therefore imperative to reduce undesired vegetative growth. In addition to these considerations, unreliable and poor flowering are important factors that contribute to low seed productivity in Jatropha [9]. The FLOWERING LOCUS T (FT) gene plays a crucial role in the transition from vegetative growth to flowering, which is a potent factor integrating the flowering signals. In this context, the function of JcFT, an FT homolog in Jatropha, was analyzed to improve the understanding of the flowering mechanism in Jatropha, which will be critical for the genetic improvement of this species.

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The transition from vegetative to reproductive growth in plants is regulated by both environmental and endogenous cues [10]. The genetic network of flowering has been investigated primarily in the model plant Arabidopsis, and five major genetically pathways control flowering initiation: the photoperiod, vernalization, gibberellin, autonomous and age pathways [11]. Recent advances in transgenic plants and traditional grafting studies have revealed that FT protein acts as a mobile flowering signal, whose ability to induce flowering involves long-distance transport [12,13]. The findings of many studies have helped establish the role of FT as a floral pathway integrator that respond to both environmental and endogenous flowering signals [14].

In Arabidopsis, FT is expressed in leaf phloem, and the FT protein subsequently moves to the shoot apex, where it forms a complex with the basic domain/leucine zipper protein FD. This FT/FD heterodimer activates the downstream floral meristem identity gene APETALA1 (AP1) [12,15,16]. FT-like genes have been isolated from many plants, including tomato [17], pumpkin [18], rice [19], barley [20], grape [21], apple [22], and potato [23], and the function of most FT genes is conserved [24].

In this study, we cloned and characterized the Jatropha FT homolog, JcFT. We also analyzed the function of JcFT in floral induction using transgenic Arabidopsis and Jatropha.

**Results**

**Cloning and sequence analysis of JcFT**

A combined reverse transcriptase-polymerase chain reaction (RT-PCR) and rapid-amplification of cDNA ends (RACE) strategy was used to isolate an FT-like cDNA from Jatropha. JcFT cDNA (GenBank accession no. KF113881) encoded a 176-amino-acid protein with 89%, 83%, 80%, and 78% sequences identity with Litchi chinensis LcFT [25], Citrus unshiu CiFT [26], rice Hd3a [27], and Arabidopsis FT [28], respectively. The molecular weight and isoelectric point of the deduced protein were 20.03 kDa and 6.82, respectively.

The genomic sequence of JcFT consisted of four exons, which resembles the genomic structure of other FT genes (Figure 1A). A multiple alignment was performed using the JcFT sequence and the sequences of FT homologs from other species (Figure 1B). The conserved key amino acid residue Tyr (Y) found in FT homologs was identified at position 85 of the JcFT protein (Figure 1B). JcFT also contained two highly similar sequences to Arabidopsis FT in the 14-AA stretch known as “segment B” and in the LYN triad in “segment C” [29] (Figure 1B).
A phylogenetic tree was constructed to analyze the phylogenetic relationship between JcFT and the FTs from other angiosperms (Figure 2). The analysis revealed that the JcFT protein (indicated with a red-boxed) was more closely related to the FTs of perennial woody plants such as *Litchi chinensis*, instead of annual herbaceous plants such as *Arabidopsis*.

**Expression pattern of JcFT in Jatropha**

To assess the expression pattern of JcFT in *Jatropha*, we performed a quantitative RT-PCR (qRT-PCR) analysis using the specific primers listed in Table S1. JcFT was expressed in all adult plants tissues except young leaves (Figure 3). Interestingly, JcFT was primarily expressed in the reproductive organs rather than the leaves, where expression of a florigen-encoding gene is expressed (Figure 3).

**Constitutive overexpression and phloem-specific expression of JcFT in Arabidopsis induces early flowering and complements the ft-10 mutant phenotype**

To determine whether JcFT is involved in the regulation of flowering time, JcFT cDNA driven by the constitutive cauliflower mosaic virus 35S (CaMV 35S) promoter or the phloem-specific *Arabidopsis* SUCROSE TRANSPORTER 2 (SUC2) promoter was transformed into wild-type *Arabidopsis* Columbia (WT) and ft-10 mutant plants. An empty vector was transformed into WT as a control. Transgenic plants were confirmed by RT-PCR analysis of JcFT expression (Additional file 1: Figure S1A). Twenty-four and seven independent T0 transgenic lines were generated with the 35S::JcFT construct in WT and ft-10 mutant, respectively. For most of these lines, bolting occurred significantly earlier than in WT and ft-10 plants under inductive long-day (LD) conditions (Figures 4A and 5A).

We selected four independent homozygous lines in the T2 generation to examine the phenotypes. The L1 and L9 lines were created by transforming WT with the 35S::JcFT construct, and the C1 and C7 lines harbored the construct in the ft-10 mutant background. Lines L1 and L9 bolted 8–14 days earlier and produced 6–11 fewer rosette leaves than the WT control under LD conditions, whereas no differences in bolting time were observed when comparing WT and the transgenic lines transformed with the empty vector (Figure 5A). Under non-inductive short-day (SD) conditions, all transgenic plants flowered much earlier than WT and the ft-10 mutant, both of which did not flower until 60 days after sowing in soil (Figures 4B and 5B). JcFT overexpression in *Arabidopsis* did not cause any

![Figure 2 Phylogenetic analysis of the FT homologs from different plant species.](image)
defects in flower development (Figure 4C and 4D), but it did significantly reduce vegetative growth time. Further analysis indicated that the promotion of flowering in 35S::JcFT transgenic Arabidopsis was correlated with a significant up-regulation of the flower meristem identity genes AP1 and LEAFY (LFY) (Additional file 2: Figure S1).

To determine whether FT-like genes are functionally conserved and active in vascular tissue, the phloem-specific promoter SUC2 has been used to drive the expression of FT-like genes in Arabidopsis and other species [12,30-32]. We obtained ten WT and eight ft-10 independent T0 transgenic lines harboring the SUC2::JcFT construct. The S1 and S3 lines were created by transforming WT with the SUC2::JcFT construct, and the CS1 and CS4 lines harbored the construct in the ft-10 mutant background. Similar to the observations for the 35S::JcFT transgenic lines, lines S1 and CS1 flowered much earlier than WT and ft-10, respectively. Lines S3 and CS4 flowered at approximately the same time and produced as many leaves as WT (24 days, 12 leaves) or flowered slightly earlier (Figure 5A) under LD conditions. Similar to the 35S::JcFT transgenic lines, all the SUC2::JcFT transgenic lines flowered earlier than WT and ft-10 under SD conditions (Figure 5B).

Taken together, these findings demonstrated that ectopic expression of JcFT in Arabidopsis resulted in an early flowering phenotype.

Overexpression of JcFT in Jatropha causes early flowering in vitro

The transgenic analysis in Arabidopsis suggested that JcFT could be a floral activator in Jatropha. To test whether JcFT similarly resulted in an early-flowering phenotype in Jatropha, we generated transgenic Jatropha with the 35S::JcFT construct used for Arabidopsis transformation. Mature Jatropha cotyledons were used as explants for transformation, as previously described [33]. To our surprise, flower buds initiated directly from the Agrobacterium-transformed calli after in vitro culture for seven weeks (Figure 6A and 6B), whereas the control explants never produced flower buds under the same conditions. The in vitro cultured transgenic Jatropha also produced intact inflorescences, but the inflorescences did not produce as many small flowers as wild Jatropha in the field (Figure 6C and 6D). Nevertheless, these findings demonstrate that JcFT is a powerful inducer of flowering in Jatropha.

Although flower buds were produced in vitro, most were abortive and wilted several weeks later. A few flower buds developed into flowers (Figure 7A and 7C), but these flowers also wilted. Furthermore, these in vitro flowers were abnormal; for example, the petals of the female flower could not spread (Figure 7A). By removing the sepals and petals of female flower, the pistil was made visible (Figure 7B). Compared with the wild-type female flower (Figure 7F), the stigma of transgenic female flower was shorter (Figure 7B). An abnormal in vitro hermaphrodite flower of transgenic Jatropha (Figure 7C) had six stamens with very short filaments (Figure 7D) in contrast to the normal male flower (Figure 7E, G), which has ten stamens (Figure 7H). Consequently, no regenerated transgenic plants harboring 35S::JcFT were obtained.

To determine whether JcFT overexpression in the transgenic in vitro flowering lines altered the expression of downstream flowering genes, such as SUPPRESSOR OF OVEREXPRESSION OF CONSTANS 1 (SOC1), LFY, and AP1 homologs in Jatropha [11], qRT-PCR analysis was performed with RNA extracted from apex of the 35S::JcFT transgenic and wild-type shoots cultured in vitro. As expected, the transcript levels of JcLFY, JcAPI, and JcAP3 were significantly up-regulated (Figure 8). JcSOC1 was also strongly up-regulated in the transgenic in vitro flowering lines (Figure 8), indicating that it is a target of JcFT, which is consistent with the findings that SOC1 and API are activated by the FT–FD complex in Arabidopsis [15,16,34].

Discussion

Chailakhyan [35] coined the term “florigen” to refer to the floral stimulus, but exactly what contributes florigen remains unclear. Evidence indicating that Arabidopsis FT protein acts as a long-distance signal to induce flowering was published half a decade ago [12]. Subsequent findings have led to the now widely accepted view that FT protein is the mobile flowering signal (florigen), or at the very least, a component of it [14]. In the present

**Figure 3 Expression of JcFT in various organs of three-year-old adult Jatropha.** The qRT-PCR results were obtained from two independent biological replicates and three technical replicates for each sample. The levels of detected amplicons were normalized using the amplified products of the JcActin1. The mRNA level in the root tissue was set as the standard with a value of 1.
study, we found that *JcFT* encoded an *FT* homolog in *Jatropha*, and thus represented a potential flowering activator.

*FT*-like genes have been isolated from many plants. There are two members of the *FT*-like subclade in *Arabidopsis* [36], five in Lombardy poplar [37], ten in soybean [38], three in chrysanthemum [39], thirteen in rice, and fifteen in maize [40]. In *Jatropha*, we cloned only one member of the *FT*-like subclade, and only one *FT*-like gene was identified in the whole genome sequence data of *Jatropha* [41,42]. Many transgenic plants overexpressing *FT* homologs exhibit an early flowering phenotype [22,38,39,43,44], suggesting a conserved function of *FT* homologs in flowering induction in different plant species.

Although the leaf is generally expected to be the site where a florigen gene is translated into protein [13], many *FT*-like genes are abundantly expressed in reproductive organs, such as flowers and immature siliques in *Arabidopsis* [28], flowers and pods in soybean [38], capsules in poplar [37], inflorescence axes in *Curcuma kwangsiensis* [32], and flowers and berries in grapevine [45]. In the present study, *JcFT* was mainly expressed in flowers, fruits, and seeds, with the highest expression level in female flowers, suggesting that *JcFT* may be involved in the development of reproductive organs. In fact, *FT*-like genes in various species play multifaceted roles in plant development in addition to the crucial role of *FT* homologs in flowering induction [10].

Transgenic *Arabidopsis* ectopically expressing the *JcFT* exhibited an early flowering phenotype compared with the control plants (Figures 4 and 5). Similarly, transgenic *Jatropha* overexpressing *JcFT* flowered *in vitro* during regeneration (Figure 6), which may have resulted from
the up-regulation of flowering genes downstream of *JcFT* (Figure 8). Unexpectedly, the transgenic *Jatropha* flower buds that were produced *in vitro* failed to develop normally into mature flowers. Many flower buds were abortive, and only a few developed into abnormal flowers. We supposed that the floral abnormalities and the failure of regeneration of the 35S::*JcFT* transgenic *Jatropha* plants could result from the ectopic overexpression of *JcFT* driven by the strong constitutive 35S promoter. Consistent with this hypothesis, by using a phloem-specific promoter...
SUC2, we successfully obtained SUC2::JcFT transgenic Jatropha shoots, which were grafted onto rootstocks of wild-type Jatropha seedlings. The grafted SUC2::JcFT transgenic Jatropha plants flowered earlier than did wild-type plants, and produced normal flowers (Additional file 2: Figure S2). Therefore, the production of normal transgenic Jatropha overexpressing JcFT for use in molecular breeding programs of Jatropha will likely require the use of weaker constitutive promoters [43], tissuespecific promoters [46], or inducible promoters [47] to

Figure 7 Abnormal flowers of transgenic Jatropha harboring 35S::JcFT. (A) A female flower of transgenic Jatropha cultured in vitro. (B) Pistil of a transgenic female flower. (C) An abnormal hermaphrodite flower of transgenic Jatropha cultured in vitro. (D) Abnormal stamens from an abnormal hermaphrodite flower of transgenic Jatropha shown in (C). (E) Normal female and male flowers of wild Jatropha grown in the field. (F) Pistil of a wild-type female flower. (G and H) Stamens of a wild-type male flower. Bars in (A)-(D) and (F)-(H) represent 1 mm, and bar in (E) represents 5 mm. Red arrows indicate pistils, and blue arrows indicate stamens.
confine the expression of the transgene JcFT to shoot meristems at an appropriate level. In addition, a loss of function analysis with a RNA interference construct targeted at JcFT will be necessary to determine the exact function of JcFT in Jatropha flowering.

Conclusions
The FT homolog of the biofuel plant Jatropha was isolated and characterized in the present study. JcFT is mainly expressed in the reproductive organs, including female flowers, fruits, and seeds. JcFT also induced early flowering in transgenic Arabidopsis and Jatropha, indicating that JcFT acts as a flowering promoter in Jatropha.

Materials and methods
Plant materials and growth conditions
The roots, stems, young leaves, mature leaves, flower buds, flowers, and fruits of Jatropha curcas L. were collected during the summer from the Xishuangbanna Tropical Botanical Garden of the Chinese Academy of Sciences, Mengla County, Yunnan Province in southwestern, China. Mature seeds were collected in autumn. All tissues prepared for qRT-PCR were immediately frozen in liquid N\textsubscript{2} and stored at −80°C until use.

WT Arabidopsis thaliana ecotype Columbia (Col-0), the ft-10 mutant (a gift from Dr. Tao Huang, Xiamen University), and the transgenic lines were grown in peat soil in plant growth chambers at 22 ± 2°C under a 16/8 h (light/dark) or 8/16 h (light/dark) photoperiod, with cool-white fluorescent lamps used for lighting. Transgenic plants in the T\textsubscript{2} homozygous generation were selected to examine flowering time and other phenotypes. For each genotype, ten plants were used to for characterization: the number of leaves was counted along with the number of days between sowing and when the first flower bud was visible.

Cloning of JcFT cDNA
Total RNA was extracted from the leaves of flowering Jatropha using the protocol described by Ding et al. [48] First-strand cDNA was synthesized using M-MLV-reverse transcriptase from TAKARA (Dalian, China) according to the manufacturer's instructions. To clone the conserved region of JcFT cDNA, a pair of primers, ZF632 and ZF633, was designed according to the conserved regions of FT homologs from other plant species using the Primer Premier 5 software. The PCR products were isolated, cloned into the pMD19-T simple vector (TAKARA, Dalian, China), and sequenced. The cloned sequence was used to design gene-specific primers (GSPs) to amplify the cDNA 5′ and 3′ end. The primers were listed in Table S1. First round PCR and nested amplification were performed according to the instructions provided in the SMART™ RACE cDNA Amplification Kit User Manual (Clontech). The PCR products were subsequently cloned into pMD19-T and sequenced.

The full length JcFT cDNA was obtained by PCR using the primers JcFT-F and JcFT-R, which introduced KpnI and SalI recognition sites, respectively, to facilitate the transformation of JcFT into Arabidopsis and Jatropha. The PCR products were subsequently cloned into the pMD19-T and sequenced.
Sequence and phylogenetic analyses

Sequence chromatograms were examined and edited using Chromas Version 2.23. Related sequences were identified using BLAST (http://www.ncbi.nlm.nih.gov/BLAST/). To determine the amino acid identities, sequences from the alignment were pairwise compared using DNAMAN 6.0. A phylogenetic tree based on the protein sequences was constructed using MEGA5.0 (http://www.megasoftware.net). The amino acid sequences of the PEBP family were assembled using ClustalX. A Neighbor–Joining phylogenetic tree was generated with MEGA 5.0 using the Poisson model with gamma-distributed rates and 10000 bootstrap replicates. The molecular weight and isoelectric point of the protein were analyzed on-line using ExPASy (http://web.expasy.org/compute_pi/).

Plant expression vector construction and Arabidopsis and Jatropha transformation

To construct the plant overexpression vector 35S::jcFT, the JcFT sequence was excised from the pMD19-T simple vector using the restriction enzymes KpnI and SalI and then cloned into the pOCA30 vector containing the CaMV 35S promoter and the 35S enhancer. The SUC2 promoter was obtained by PCR from Arabidopsis genomic DNA using primers SUC2-F and SUC2-R, which introduced HindIII and KpnI recognition sites, respectively. The PCR products were cloned into pMD19-T and sequenced. To construct the SUC2::jcFT plasmid, the 35S promoter of the vector containing 35S::jcFT was placed with the SUC2 promoter using the restriction enzymes HindIII and KpnI. The fidelity of the construct was confirmed by PCR and restriction digestion.

Transformation of WT Col-0 and ft-10 mutant plants with Agrobacterium strain EHA105 carrying the recombinant vector was performed using the floral dip method [49]. Transgenic seedlings were selected for kanamycin resistance and confirmed by genomic PCR and RT-PCR.

Transformation of Jatropha with Agrobacterium strain LBA4404 carrying the overexpression construct was performed according to the protocol described by Pan et al. [33].

Expression analysis by qRT-PCR

Jatropha total RNA was extracted from frozen tissue as described by Ding et al. [48] Arabidopsis total RNA was extracted from frozen tissue using TRIZol reagent (Transgene, China). First-strand cDNA was synthesized using the PrimeScript™ RT Reagent Kit with gDNA Eraser (TAKARA, Dalian, China) according to the manufacturer’s instructions. qRT-PCR was performed using SYBR® Premix Ex Taq™ II (TAKARA) on a Roche 480 Real-Time PCR Detection System (Roche Diagnostics).

The primes used for qRT-PCR are listed in Table S1. qRT-PCR was performed using two independent biological replicates and three technical replicates for each sample. Data were analyzed using the 2 -ΔΔCT method as described by Livak and Schmittgen [50]. The transcript levels of specific genes were normalized using Jatropha Actin1 or Arabidopsis Actin2.

Availability of supporting data

All the supporting data of this article are included as additional files (Additional file 1: Figure S1; Additional file 2: Figure S2; Additional file 3: Table S1).

Additional files

**Additional file 1:** Figure S1. RT-PCR analysis of flowering genes downstream of FT in WT and transgenic Arabidopsis. Arabidopsis seedlings were collected 20 days after germination. For semi-quantitative RT-PCR, 25 cycles were used for the reference gene AtActin2, and 30 cycles were used for the target genes. The qRT-PCR results were obtained from three technical replicates for each sample. Values were normalized using AtActin2 as a reference. The mRNA level in WT was set as the standard with a value of 1.

**Additional file 2:** Figure S2. Early flowering of SUC2::jcFT transgenic Jatropha. Transgenic shoot grafted onto a non-transgenic rootstock showing the precocious flowers (red oval) forty days after grafting. Red arrows indicate the graft sites.

**Additional file 3:** Table S1. Primers used in this study.

**Abbreviations**

AP1: APETALA1; FT: FLOWERING LOCUS T; LFY: LEAFY; LD: long day; SD: short day; SOC1: SUPPRESSOR OF OVEREXPRESSION OF CONSTANS 1; SUC2: sucrose transporter 2; qRT-PCR: quantitative reverse transcriptase-polymerase chain reaction.

**Competing interests**

The authors declare that they have no competing interests.

**Authors’ contributions**

CL and ZFX conceived the experiment and drafted the manuscript. LL contributed to the data processing. QF contributed to the data processing. LN collected the tissue samples. All authors read and approved the final manuscript.

**Authors’ information**

CL and LN are PhD students, LL was a master student at the time of study, and QF is an associate professor, and ZFX is a professor and head of the laboratory.

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References

1. Divakara B, Upadhyaya H, Wan S, Gowda C: Biology and genetic improvement of Jatropha curcas L: A review. Appl Energy 2010, 87:732–742.

2. Akashi K: Jatropha research: A new frontier for biofuel development. Plant Biotechnol 2012, 29:121.

3. Pua F-l, Fang Z, Zakaria S, Guo F, Chia C-h: Integration of spatial and temporal information during floral induction of Jatropha curcas and Calophyllum inophyllum for biodiesel: a review. Renew Sust Energy Rev 2011, 15:3510–3515.

4. Khalil H, Aprilia N, Bhat A, Jawaid M, Paridah M, Rudi D: Jatropha curcas L: A potential biofuel plant for sustainable environmental development. Renew Energy 2003, 28:239–248.

5. Ghosh A, Chaudhary D, Reddy M, Rao S, Chikara J, Pandya J, Patolia J, Gandhi M, Adimurthy S, Wood C, Gang, Wang T, Putterill J, Hellens RP: Homologs of FT, CEN and FD respond to developmental and environmental signals affecting growth and flowering in the perennial vine kiwifruit. New Phytol 2013, 198:732–746.

6. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

7. Pramanik K: Properties and use of Jatropha curcas L. Plant Cell Physiol 2010, 51:659–674.

8. Ghosh A, Chaudhary D, Prakash AR, Boricha G, Zala A: Paclobutrazol arrests vegetative growth and unifies unexpressed yield potential of Jatropha curcas. J Plant Growth Regul 2010, 29:307–315.

9. Pin P, Nilsson O: The multifaceted roles of FLOWERING LOCUS T in plant development. Plant Cell Environ 2013, 36:1742–1755.

10. Pin P, Nilsson O: The multifaceted roles of FLOWERING LOCUS T in plant development. Plant Cell Environ 2013, 36:1742–1755.

11. Corbesier L, Vincent C, Jang S, Fornara F, Fan Q, Searle I, Giakountis A, Vagnozzi R, Watanabe Y, Iwabuchi M, Araki T: A pair of related genes with antagonistic roles in mediating flowering signals. Science 1999, 286:1960.

12. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

13. Fujii T, Suzuki MA: Cloning and expression analysis of the FLOWERING LOCUS T (FT) homologous gene in Jatropha curcas L. using kanamycin selection. Afr J Biotechnol 2010, 9:477–481.

14. Michaels SD: Flowering time regulation produces much fruit. Curr Opin Plant Biol 2009, 12:75–80.

15. Challakhyan MK: New facts in support of the hormonal theory of plant development. CR Acad Sci URS 1936, 1936:79–83.

16. Yano K, Shimizu T, Takahashi S, Iwabuchi M, Araki T: A genomic and expression analysis of the FLOWERING LOCUS T (FT) homologous gene in Jatropha curcas L. using kanamycin selection. Afr J Biotechnol 2010, 9:477–481.

17. Danilevskaya ON, Simmons CR: A genomic and expression complement of the expanded PEBP gene family from maize. Plant Physiol 2008, 146:258–265.

18. Sato S, Hirakawa K, Ishobe S, Fukushima K, Shin-iti T: The tomato FT ortholog triggers systemic signals that regulate growth and flowering and substitute for diverse environmental stimuli. Proc Natl Acad Sci U S A 2006, 103:6398–6403.

19. Lin M-K, Belanger H, Lee Y-J, Varkony-Gasic E, Tacka K-I, Miura E, Xocorrente-Cázares B, Gendler K, Jorgensen RA, Phinney B, Lough TJ, Lucas WJ: FLOWERING LOCUS T protein may act as the long-distance floral signaling protein in the cucurbits. Plant Cell 2007, 19:1588–1596.

20. Nishiguchi M, Yano K, Shimizu T, Takahashi S, Iwabuchi M, Araki T, Miura E, Abe K: Molecular characterization of FLOWERING LOCUS T-like genes of apple Malus domestica Borkh.. Plant Cell Physiol 2010, 51:561–575.

21. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

22. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

23. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

24. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

25. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

26. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

27. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

28. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

29. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

30. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

31. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

32. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

33. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

34. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

35. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

36. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

37. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

38. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

39. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

40. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.

41. Danilevskaya ON, Simmons CR: Expression of FT-like genes of apple Malus domestica Borkh. in Arabidopsis thaliana. New Phytol 2013, 198:2873–2883.
44. Imamura T, Nakatsuka T, Higuchi A, Nishihara M, Takahashi H: The gentian orthologs of the FT/FL1 gene family control floral initiation in Gentiana. Plant Cell Physiol 2011, 52:1031–1041.

45. Sreekantan L, Thomas MR: VvFT and VvMADS8, the grapevine homologues of the floral integrators FT and SOC1, have unique expression patterns in grapevine and hasten flowering in Arabidopsis. Funct Plant Biol 2006, 33:1120–1139.

46. Tränkner C, Lehmann S, Hoenicka H, Hanke M-V, Fladung M, Lenhardt D, Dunemann F, Gau A, Schlangen K, Malony M, Flachowsky H: Over-expression of an FT-homologous gene of apple induces early flowering in annual and perennial plants. Planta 2010, 232:1309–1324.

47. Wenzel S, Flachowsky H, Hanke M-V: The Fast-track breeding approach can be improved by heat-induced expression of the FLOWERING LOCUS T genes from poplar (Populus trichocarpa) in apple (Malus x domestica Borkh.). Plant Cell Tissue Organ Cult 2013, 115:127–137.

48. Ding L-W, Sun Q-Y, Wang Z-Y, Sun Y-B, Xu Z-F: Using silica particles to isolate total RNA from plant tissues recalcitrant to extraction in guanidine thiocyanate. Anal Biochem 2008, 374:426–428.

49. Clough SJ, Bent AF: Floral dip: a simplified method for Agrobacterium-mediated transformation of Arabidopsis thaliana. Plant J 1998, 16:735–743.

50. Livak KJ, Schmittgen TD: Analysis of relative gene expression data using real-time quantitative PCR and the 2^−ΔΔCT method. Methods 2001, 25:402–408.

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