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ORIGINAL RESEARCH

A Defect in Zinc Finger Protein Double B-box 1a (DBB1a) Causes Abnormal Floral Development in *Arabidopsis*

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Abstract The double B-box (DBB) type zinc finger protein has thus far been shown to be involved in photomorphegenesis in *Arabidopsis thaliana*. Here, we show that *DBB1a* is expressed in the embryo, cytolden, and flower. Misexpression of *DBB1a* in mutant plants resulted in abnormal numbers and patterns of floral organs. We further show that *DBB1a* could regulate expression of several floral homeotic genes, including *APETALA 2*, *APETALA 3*, *PISTILLATA*, and *AGAMOUS*. Interestingly, expression of the microRNA gene *MiR172*, which is involved in organ boundary establishment, was also misregulated in the *dbb1a* mutant plants. Our study identified a previously uncharacterized role of *DDB1a* in regulation of expression of floral homeotic genes and *miR172*, which is important for understanding of floral pattern formation.

Keywords *Arabidopsis thaliana* · Double B-box type zinc finger · Flower development

Introduction

Floral organogenesis is dependent on the combinatorial action of many genes, which in turn control the expression of suites of genes required for growth, patterning, and differentiation. Molecular genetics approaches have uncovered genes that specify floral homeotic gene (Chae et al. 2008). The shoot apical meristem after the vegetative to reproductive transition is the foundation for flower initiation (Smyth et al. 1990). The flower consists of four organ types that develop in distinct floral whorls: four sepals in the first whorl, four petals in the second whorl, six stamens in the third whorl, and two fused carpels in the fourth whorl. Within each of these floral organs, a variety of tissue and cell types differentiate (Nag et al. 2007). Organ identity is specified by the overlapping activities of three classes of homeotic genes, termed A, B, and C, as predicted in the ABC model (Coen and Meyerowitz 1991). The activity of a floral homeotic gene is typically confined to two adjacent whorls, with the A class represented by *APETALA1* (*AP1*) and *APETALA2* (*AP2*) acting in whorls one and two, the B class genes *APETALA3* (*AP3*) and *PISTILATA* (*PI*) in whorls two and three, and the C class gene *AGAMOUS* (*AG*) in whorls three and four (Lohmann and Weigel 2002).

Photoperiod is one of the most important environmental signals affecting flower (Lin 2002). *GIGANTEA* (*GI*) acts between the circadian oscillator and *Constans* (*CO*) to promote flowering by increasing *CO* and *FLOWER LOCUS T* (*FT*) mRNA abundance (Mizoguchi et al. 2005), while *GI* could regulate miR172 processing (Jung et al. 2007). MiR172 regulates both floral architecture and flowering time by controlling a group of *AP2* domain-containing transcription factors. *AP1*, *AP3*, and *PI* are other cascade for flower development, while miR164 family of mircroRNAs is a role in defining the boundaries (Irish 2008). The MADS domain transcription factor AG plays a central role in floral patterning and is required not only for the specification of the two reproductive organ types, but also for termination of stem cell fate (Lohmann and Weigel 2002).

B-box domain zinc finger family proteins play an important role in light and circadian signaling in *Arabidopsis*

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(Datta et al. 2008; Kumagai et al. 2008). *CO* is the representative of B-box zinc finger proteins in *Arabidopsis* (Putterill et al. 1995; Wenkel et al. 2006). Double B-box-type zinc finger (DBB) subfamily protein which contains N-terminal zinc-binding B-boxes that are spaced by eight to fifteen amino acids, which have thus far been associated mainly to function as a regulator in photomorphogenesis and circadian signaling in *Arabidopsis*. For example, *SALT TOLERANCE*, *SALT TOLERANCE HOMOLOG2* (*STH2*), *LIGHT-REGULATED ZINC FINGER*/*STH3*, which were identified as genes transcriptional regulated by *ELONGATED HYPOCOTYL 5* (*HY5*) (Ang and Deng 1994; Datta et al. 2007; Datta et al. 2008; Kumagai et al. 2008; Nagaoka and Takano 2003). However, the biological functions and physiological roles of other double B-box type zinc finger genes in plant are scarcely talked about. Double B-box 1a (*DBB1a*) AT2G21320 is one of the DBB subfamily. *DBB1a* is involved in photomorphogenesis and circadian signaling (Kumagai et al. 2008). However, it is not clear what detailed basis is. To investigate precise functions of *DBB1a*, we constructed the *DBB1a* overexpression and RNAi transgenic plants, However, we found that misexpression of *DBB1a* resulted in floral development abnormity, while overexpression of *DBB1a* had no effect on floral development.

In plants, after the initiation of the flower primodium, but before the formation of the primordia of individual organs, the cells in the flower primordium learn their positions, and on the basis of their positional information, direct their descendants to form organ primordia in appropriate types, numbers, and places (Chae et al. 2008; Irish 2008; Smyth et al. 1990). We observed that the abnormal floral development in *DBB1a* lose-of-function mutants, however, whether *DBB1a* plays a direct or indirect role in floral development is unknown.

In this report, we identified that *DBB1* was crucial for proper development of floral organs and expressed in embryo and flower. Moreover, we also showed that *DBB1a* could regulate the transcription level of a series of genes which play fundamental role in floral development.

Materials and Methods

Plant Materials, Growth Conditions

Columbia (Col-4) was used as wild type in this study. To avoid cosuppression, the overexpression plasmids were also transformed to *SGS* mutant (Elmayan et al. 1998). *Arabidopsis thaliana* plants were grown in a controlled culture room set at 23°C with an RH of 60%. The photoperiods were 16 h of light and 8 h of dark, with white light illumination (120 $\mu mol.m^{-2}.s^{-1}$).

Plasmid Construction and Plant Transformation

The plasmids were subcloned through Gateway technology (Invitrogen, USA). The full-length coding regions (CDS) of

Table 1 Primers used for PCR and qPCR analyses

Genes	Forward (5′-3′)	Reverse (5′-3′)
AT2G21320CDNA	ACAAGTTGTACAAAAAAGCAGGC TACTATCCATCATATTTCTCTGCTG	ACCACTTTGTACAAGAAAGCTGGGT TGAGATATAACTCTGAAGAATCG
AT2G21320RNAi-1	ACAAGTTGTACAAAAAAGCAGGC TAATAGATGGTAGTTCCCTTTG	ACCACTTTGTACAAGAAAGCTGGGT TTAGTCCCAGTTGGTCAGC
AT2G21320RNAi-2	ACAAGTTGTACAAAAAAGCAGGC TATGCTGACCAACTGGGACT	ACCACTTTGTACAAGAAAGCTGGGT TCGTGATTGTTTGCGTTATTT
AT2G21320RNAi-3	ACAAGTTGTACAAAAAAGCAGGC TAAAGAGGAGGGTATTGATG	ACCACTTTGTACAAGAAAGCTGGGT TGACATTTCCCTGTAAAGATA
AT2G21320Promoter	ACAAGTTGTACAAAAAAGCAGGC TAAGGTGTGAAATGGGTTC	ACCACTTTGTACAAGAAAGCTGGGT TTTCGTCGCAGGAGCAACAG
DBB1a	TGGTACATGTTGTGGGAAGAG	CCCAGTTGGTCAGCATGATTAG
Actin7	ATCCCTCAGCACCTTCCAAC	ACAAACTCACCACCACGAAC
Actin2	CACTGTGCCAATCTACGAGGGT	CACAAACGAGGGCTGGAACAAG
AP1	GCAAGCAATGAGCCCTAAAGAG	AGTGCGGATGTGCTTAAGAGC
AP2	CGGCTCAGGATGAACCAACA	CGCCTCCGCTACCAATGT
AP3	ATGGAGGAGATTACGACTCAGTTC	TGATGTCAGAGGCAGAGGGTG
PI	GGTCTTTAGTTTGTCTTTATCAATCTGT	CAACCAATACATAGAGGTCATAAGCA
MiR164	TTGATGGAGAAGCAGGGCAC	CCATTGACGATTGCATCCTCG
MiR172	TCTGTTGATGGACGGTGGTG	GAGGGAGAGAGAGAGAGAGGGAA
GI	CAGGCTGAAAGGGAGTTGCA	CAGTGACTCAGGCCTGTCATGA

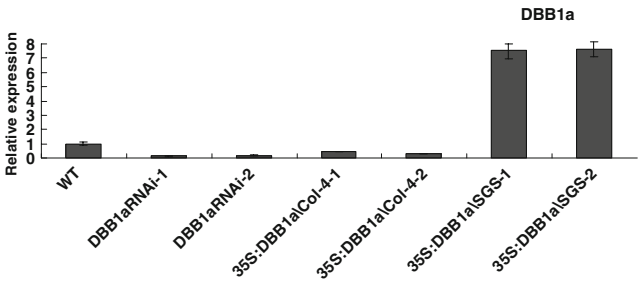

Fig. 1 Quantitative RT-PCR analyses of transcript abundance of *DBB1a*. The results of quantitative RT-PCR indicated that *DBB1a* transcript accumulation were reduced in *DBB1a-RNAi* and *35S:DBB1a/Col-4* (DBB1a-co-suppression) seedlings in relation to wild-type at 7 days after germination in continuous white light, while, overexpression transgenic lines (*35S:DBB1a\SGS*) had considerably higher levels of *DBB1a* transcripts than the wild type. The data were derived from three biological independent sets of samples, and each sample was tested in triplicate. The relative amount of *DBB1a* RNA was determined by calculating the mean of the median values for the RNA accumulation in each three biological replicates. Actin2 and actin7 mRNA levels in the samples were used to normalize the data. Data represent the means (± SD) of three independent assays

DBB1a and the segments for 447-589BP, 569-747 bp, and 709-823 bp of coding regions (CDS) of *DBB1a* were polymerase chain reaction (PCR) amplified, respectively, from a cDNA library with primer pairs PAT2G21320CDNA, P21320RNAi1, P21320RNAi2, P21320RNAi3, generated to entry vector pDONR201 by BP recombination and sequenced. The *DBB1a* CDS entry clone was generated to CaMV35S::pENSG-GFP-GW and CaMV35S::GW-Pleela expression by LR reaction respectively. The *DBB1a* RNAi entry clones were generated to pJawoh18 RNAi expression vector (Yang Meiying 2007), respectively.

The *pDBB1a::GUS* plasmid was based on plant binary vector GW::GUS. We amplified the 1,975 bp region upstream of the translational start ATG of *DBB1a*, generated to pDONR207 by BP recombination and then to GW::GUS by LR recombination. The sequence of all primers are given in Table 1.

All constructs were introduced into *Agrobacterium tumefaciens* strain GV3101 90RK by a freeze-thaw method and transformed into wild-type (Col-4) or *sgs* by the floral dip transformation method (Desfeux et al. 2000; Zhang et al. 2006).

Gene Expression Analysis by Quantitative RealTime-PCR

Total RNA was isolated using PurprepRNA kit (Invitrogen, USA) from at least three plants per sample. cDNA was prepared from 2 ug total RNA using Moloney murine leukemia virus reverse transcriptase according to the manufacturer's instruction (Promega). Real-time RT-PCR analyses were performed using Sybr Green PCR Master mix (Applied Biosystems) in Mx3000P thermal cycler (Stratagene) and data were analyzed with MxPro software (Stratagene). Data collection was performed from three

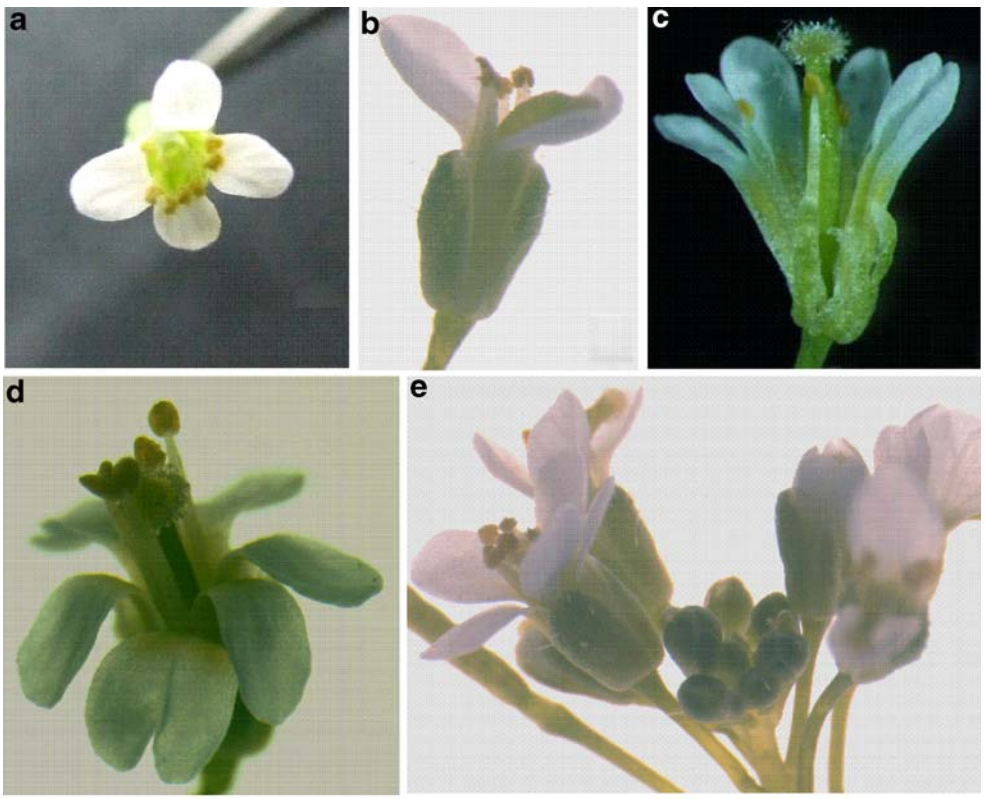

Fig. 2 Floral phenotypes of *DBB1a* loss-of-function mutant transgenic plants. Misexpression of DBB1a induced abnormal floral development. **a** showed the wild type, **b c,** and **d** showed the representative phenotypes of *DBB1a* loss-of-function transgenic plants; E indicated that flowers of less and extra petals appeared in the same inflorescence

Table 2 Petal composition of *DBB1a* flower

Number of petals	2	3	4	5	6	8	9	11	13
Percentage (%)	9	2	7	25	18	14	10	5	7

Only DBB1a flowers that externally normal in appearance were examined; n=200

independent experiments with at least three different measurements per sample. ACT2 and ACT7 were used as the internal control. The sequence of all other primers is given in Table 1.

GUS Staining

β-glucuronidase staining was performed for 2–3 h at 37°C using X-Gluc (sigma) as described previously (Richmond and Bleecker 1999).

Scanning Electron Microscopy

Inflorescences were harvested after 30 days in long-day conditions. Specimens examined were processed consulting to a previously described protocol (Ohno et al. 2004) and mounted on stubs fresh and examined at environmental scanning electron microscopy ESEM FEI quanta 200.

Results

Generation of Overexpession and Misexpression *DBB1a* Transgenic Plants

We obtained independent transgenic lines and confirmed perturbed expression of *DBB1a* in mutants by q-PCR. The results showed that overexpression transgenic lines (*35S::DBB1a\SGS*) have considerably higher levels of *DBB1a* transcripts than the wild type (Fig. 1). The *DBB1a* transcript shown in *DBB1a*RNAi and *35S::DBB1a*\Col-4 (*DBB1a* co-suppression) transgenic lines were significantly lower than that of in the wild type (Fig. 1). All the transgenic seedlings referred in this study were T3 generation seedlings.

Phenotypic Characterization of the *dbb1a* Mutant

Wild-type *Arabidopsis* flowers have, from outside to inside, four normal petal, four sepals and six stamens (Fig. 2a), however, the petal number per flower often varied from two to thirteen in *dbb1a* mutants (*DBB1a* RNAi and *DBB1a* cosuppression seedlings; Fig. 2b), while the proportion of sepal, petal and stamen was not stable, for example, when there were two petals, there were two sepals and four stamens, or there were eight petals, there were four petal

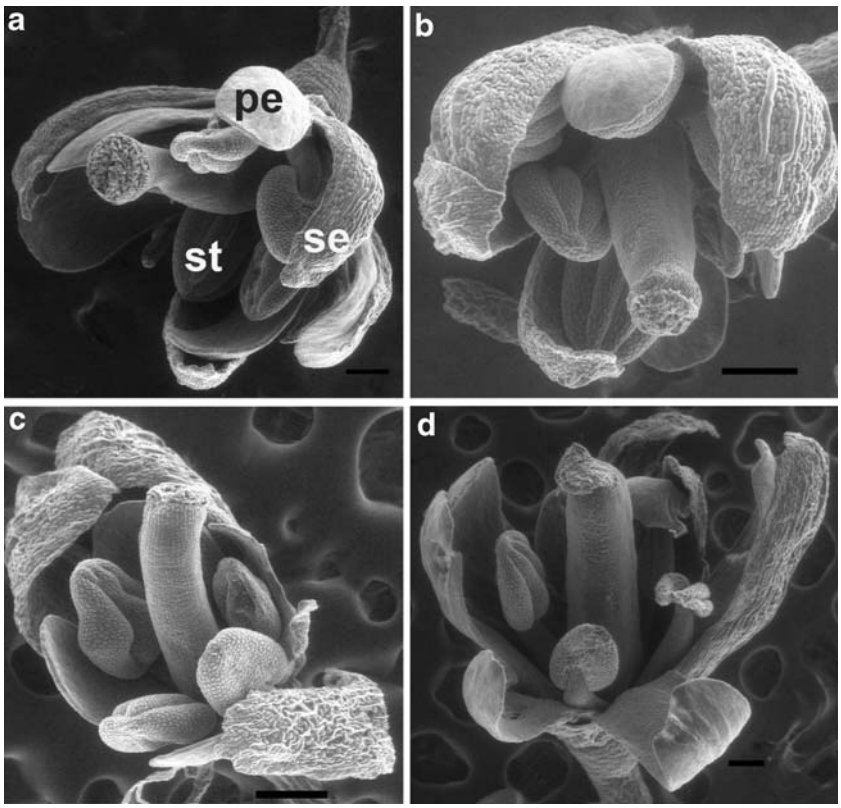

Fig. 3 Scanning electron micrographs of floral buds. **a** The floral buds of wild-types; **b**, **c**, and **d**, the floral buds of *DBB1a* loss-of-function mutant plants. The number of sepals, petals and stamens of *DBB1a* loss-of-function mutant plants were abnormal, however, the position of each floral organs were normal. The following abbreviations were used: *st* stamen, *pe* petal, and *se* sepal. The *scale bars* represent 200 μm

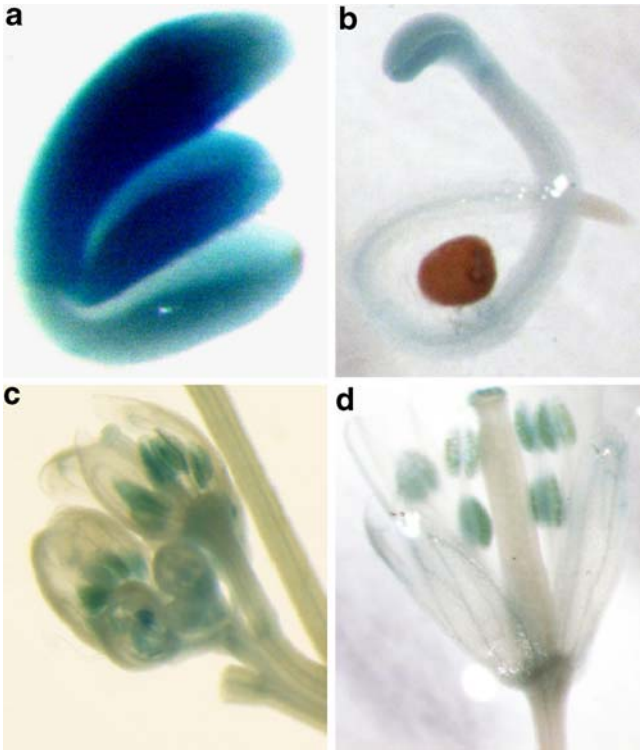

Fig. 4 The *DBB1a* promoter-driven expression of the GUS reporter gene in embryo, young seedling and young flowers. **a** *pDBB1a:GUS* embryo. GUS activity was detected at high level all over the embryo; **b** very young *pDBB1a:GUS* seedling that exhibits GUS activity in the cotyledon; **c** *pDBB1a:GUS* bud. GUS activity was detected at high levels in the stigma, node, and stamen of bud; **d** *pDBB1a::GUS* flower. GUS activity was detected at high levels in the stigma, node, and stamen of adult flower

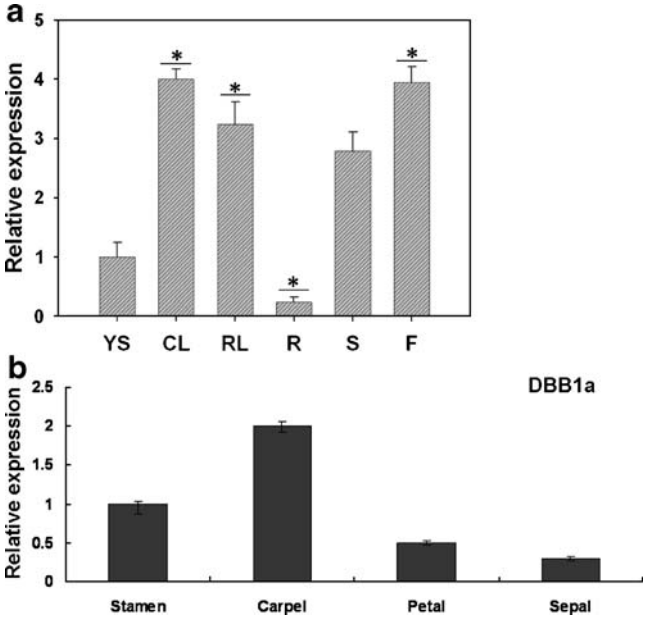

Fig. 5 **a** Comparison of *DBB1a* transcriptive levels in various mature tissues by quantitative RT-PCR. **b** Comparison of *DBB1a* transcriptive levels in various mature floral organs by quantitative RT-PCR. The level of mRNAs in each case was normalized to that of actin2 and actin7. The data were derived from three biological independent sets of samples, and each sample was tested in triplicate. The relative amount of floral homeotic genes were determined by calculating the mean of the median values for the RNA accumulation in each three biological replicates. Young seedling (*YS*), cauline leaf (*CL*), rosette leaf (*RL*), root R, silique (*S*), flower (*F*). *Asterisk* indicates significant difference at $P \leq 0.01$

and six stamens yet (Fig. 2c, d). We showed the diversity of *dbb1a* flowers through counting the number of petals. Table 2 shows the percentage of petals found in *dbb1a* flowers. However, the overexpression *DBB1a* transgenic plants were normal floral development (data not shown).

A comparison of wild-type and *dbb1a* mutants by scanning electron microscopy showed that the formation of floral organs were normal, however, the pattern and number of floral organs in the bud of *dbb1a* transgenic plants were abnormal (Fig. 3).

Expression Pattern of the *DBB1a* Gene

To gain insight into the spatial and temporal expression pattern of *DBB1a*, we constructed *pDBB1a::GUS* transgenic line. In *pDBB1a::GUS* transgenic plants, GUS activity was detected all over the embryo (Fig. 4a) and in the cotyledons of very young seedlings (Fig. 4b). In bud and flower of *pDBB1a::GUS* transgenic plants, GUS activity was obviously detected in the stigma, node, and stamen (Fig. 4c, d), and feebly detected in petal and sepal (Fig. 4b). We are confident that this line reflects the endogenous expression of *DBB1a*

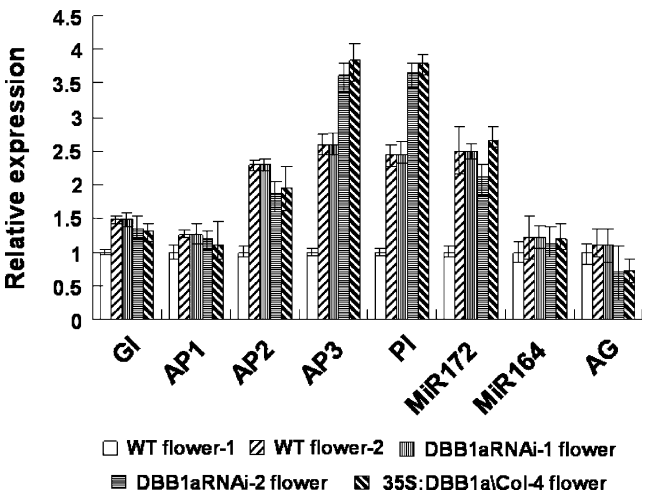

Fig. 6 Quantitative RT-PCR expression analysis of a set f flower homeotic genes in flower of different *DBB1a* genotype plants. The level of mRNAs in each case was normalized to that of actin2 and actin7. The data were derived from three biological independent sets of samples, and each sample was tested in triplicate. The relative amount of floral homeotic genes were determined by calculating the mean of the median values for the RNA accumulation in each three biological replicates

because the GUS activity pattern of *pDBB1a::GUS* transgenic plants matched the transcription expression pattern of *DBB1a* in various mature tissues as determined by q-PCR (Fig. 5).

DBB1a Modifies Transcription Expression of a Series of Flower Homeotic Genes

In order to explore the molecular mechanisms of DBB1a on floral development, the expression of flower homeotic genes were monitored. The *DBB1a* loss of function transgenic plants showed higher levels of *AP2*, *AP3*, *PI* and *MIR172*, and showed lower levels of *AG* transcripts than the wild-type plants, Moreover, the expression of *GI*, the regulator of *MIR172*, was upregulated slightly also, while the expression levels of *AP1* and *MiR164* were not measurable affected (Fig. 6). the mRNA expression levels of *MIR172* and *AP2* were upregulated in the *dbb1a* although upregulation of AP2 and miR172 are not likely coincident. Those changes were thought to be related to the *dbb1a* phenotype, in which petal number per flower often varied from two to thirteen.

Discussion and Conclusion

In this study, we have analyzed the *dbb1a* mutants, whose predominant phenotype was an abnormity in petal number in early flowers. We identified that misexpression of *DBB1a* regulated the characteristic development of floral organs, while overexpression of *DBB1a* had no effect on floral development. The temporal and spatial expression patterns of *DBB1a* showed that it expressed during very early stages of flower development. It indicated that *DBB1a* was crucial for proper development of floral organs through regulated expression of downstream genes of floral organogenesis.

Flower initiation in Arabidopsis occurs continuously in an indeterminate spiral at each floral apex (Coen and Meyerowitz 1991). During inflorescence development, flower specification depends partially upon the *FLOWERING LOCUS T* (*FY*)-*FD* (b-ZIP transcription factor) complex to directly activate *AP1* (Abe et al. 2005; Siddhartha Kanrar 2008; Wigge et al. 2005). The floral homeotic *PI* protein required for petal and stamen development, has the ability to bind directly to the promoter region of *AP1* and whose interacting partner *AP3*, regulates the transcription of *AP1* (Jens and Sundstron 2006; Krizek and Meyerowitz 1996; Lamb et al. 2002). *MIR172* has also been implicated in regulating the inner boundary of second whorl domain via the repression of *AP2* (Chen 2004; Li Zhao 2007), *AP3* and *PI* are essential for petal identity specification (Kramer et al. 1998), GI regulates *MIR172* processing rather than transcription of the *MIR172* genes. Our results showed that the number of floral organs was less or more than wild-type plants, while the transcription levels of *GI*, *MIR172*, *AP2*, *AP3* and *PI* were increased, while the transcription levels of *AG* was decreased. In previous study, *GI*, which acts earlier than *CO* and *FT* in the pathway by increasing CO and FT mRNA abundance, could regulate *MIR172* processing (Jung et al. 2007), and then *MIR172* could modify the expression of *AP2* (Chen 2004; Li Zhao 2007) but *AP2* was not only regulated by *MIR172* (Irish 2008). It is interesting that upregulation of AP2 and *MiR172* in *dbb1a*. We will get further extensive evidence to elucidate the relationship between *DBB1a* and the expression of *MiR172* by genetic or biochemistry methods in other study. Our results showed that *DBB1a* was involved in multiregulatory cascades of floral development. Despite the fact that we did not analyzed all floral homeotic genes expression in *dbb1a*, but we demonstrated that *DBB1a* could modify the transcript levels of some important floral homeotic genes, Moreover, it would be interesting to examine whether those mRNA changes in the protein levels of respective key genes in floral development. Although the target gene of *DBB1a* was unknown, the same subfamily member of *DBB*, such as *STH2*, *STH3*, were identified as a interacted protein with COP1 and HY5 (Datta et al. 2007; Datta et al. 2008). However, it is the first time to prove that double B-box protein act as a regulator of floral development. These results provide new insights into the transcriptional cascades of light signaling, which could promote flowering, but the regulation of floral development by which was still not clear. These results provide new insights into the transcriptional cascades of floral development.

We have identified that *DBB1a* was an important role in floral development. Further elucidation of the mechanism of *DBB1a* involving light signaling and floral development requires identification.

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References

Abe M, Kobayashi Y, Yamamoto S, Daimon Y, Yamaguchi A, Ikeda Y, Ichinoki H, Notaguchi M, Goto K, Araki T (2005) FD, a bZIP protein mediating signals from the floral pathway integrator FT at the shoot apex. Science 309:1052–1056

Ang LH, Deng XW (1994) Regulatory hierarchy of photomorphogenic loci: allele-specific and light-dependent interaction between the HY5 and COP1 loci. Plant Cell 6:613–628

Chae E, Tan QKG, Hill TA, Irish VF (2008) An *Arabidopsis* F-box protein acts as a transcriptional co-factor to regulate floral development. Development 135:1235–1245

Chen X (2004) A MicroRNA as a translational repressor of APETALA2 in *Arabidopsis* flower development. Science 303:2022–2025

Coen ES, Meyerowitz EM (1991) The war of the whorls: genetic interactions controlling flower development. Nature 353:31–37

Datta S, Hettiarachchi C, Johansson H, Holm M (2007) SALT TOLERANCE HOMOLOG2, a B-box protein in *Arabidopsis* that activates transcription and positively regulates light-mediated development. Plant Cell 19:3242–3255

Datta S, Johansson H, Hettiarachchi C, Irigoyen ML, Desai M, Rubio V, Holm M (2008) LZF1/SALT TOLERANCE HOMOLOG3, an *Arabidopsis* B-box protein involved in light-dependent development and gene expression, undergoes COP1-mediated ubiquitination. Plant Cell 20:2324–2338

Desfeux C, Clough SJ, Bent AF (2000) Female reproductive tissues are the primary target of *Agrobacterium*-mediated transformation by the *Arabidopsis* floral-dip method. Plant Physiol 123:895–904

Elmayan T, Balzergue S, Beon F, Bourdon V, Daubremet J, Guenet Y, Mourrain P, Palauqui J-C, Vernhettes S, Vialle T, Wostrikoff K, Vaucheret H (1998) *Arabidopsis* mutants impaired in cosuppression. Plant Cell 10:1747–1758

Irish VF (2008) The *Arabidopsis* petal: a model for plant organogenesis. Trends Plant Sci 13:430–436

Jens F, Sundstron NNKGVFI (2006) Direct regulation of the floral homeotic *APETALA1* gene by *APETALA3* and *PISTILLATA* in *Arabidopsis*. Plant J 46:593–600

Jung J-H, Seo Y-H, Seo PJ, Reyes JL, Yun J, Chua N-H, Park C-M (2007) The GIGANTEA-regulated MicroRNA172 mediates photoperiodic flowering independent of CONSTANS in *Arabidopsis*. Plant Cell 19:2736–2748

Kramer EM, Dorit RL, Irish VF (1998) Molecular evolution of genes controlling petal and stamen development: duplication and divergence within the *APETALA3* and *PISTILLATA* MADS-box gene lineages. Genetics 149:765–783

Krizek BA, Meyerowitz EM (1996) The *Arabidopsis* homeotic genes *APETALA3* and *PISTILLATA* are sufficient to provide the B class organ identity function. Development 122:11–22

Kumagai T, Ito S, Nakamichi N, Niwa Y, Murakami M, Yamashino T, Mizuno T (2008) The common function of a novel subfamily of B-box zinc finger proteins with reference to circadian-associated events in *Arabidopsis thaliana*. Biosci Biotechnol Biochem 72:1539–1549

Lamb RS, Hill TA, Tan QKG, Irish VF (2002) Regulation of *APETALA3* floral homeotic gene expression by meristem identity genes. Development 129:2079–2086

Li Zhao YKTTDXC (2007) miR172 regulates stem cell fate and defines the inner boundary of *APETALA3* and *PISTILLATA* expression domain in *Arabidopsis* floral meristems. Plant J 51:840–849

Lin C (2002) Blue light receptors and signal transduction. Plant Cell 14:S207–225

Lohmann JU, Weigel D (2002) Building Beauty: the genetic control of floral patterning. Dev Cell 2:135–142

Mizoguchi T, Wright L, Fujiwara S, Cremer F, Lee K, Onouchi H, Mouradov A, Fowler S, Kamada H, Putterill J, Coupland G (2005) Distinct roles of GIGANTEA in promoting flowering and regulating circadian rhythms in *Arabidopsis*. Plant Cell 17:2255–2270

Nag A, Yang Y, Jack T (2007) *DORNRÖSCHEN*-LIKE, an AP2 gene, is necessary for stamen emergence in *Arabidopsis*. Plant Mol Biol 65:219–232

Nagaoka S, Takano T (2003) Salt tolerance-related protein STO binds to a Myb transcription factor homologue and confers salt tolerance in *Arabidopsis*. J Exp Bot 54:2231–2237

Ohno CK, Reddy GV, Heisler MGB, Meyerowitz EM (2004) The *Arabidopsis* JAGGED gene encodes a zinc finger protein that promotes leaf tissue development. Development 131:1111–1122

Putterill J, Robson F, Lee K, Simon R, Coupland G (1995) The CONSTANS gene of *Arabidopsis* promotes flowering and encodes a protein showing similarities to zinc finger transcription factors. Cell 80:847–857

Richmond TA, Bleecker AB (1999) A Defect in {beta}-oxidation causes abnormal inflorescence development in *Arabidopsis*. Plant Cell 11:1911–1924

Siddhartha Kanrar MBBAJCHMSS (2008) Regulatory networks that function to specify flower meristems require the function of homeobox genes PENNYWISE and POUND-FOOLISH in *Arabidopsis*. Plant J 54:924–937

Smyth DR, Bowman JL, Meyerowitz EM (1990) Early flower development in *Arabidopsis*. Plant Cell 2:755–767

Wenkel S, Turck F, Singer K, Gissot L, Le Gourrierec J, Samach A, Coupland G (2006) CONSTANS and the CCAAT box binding complex share a functionally important domain and interact to regulate flowering of *Arabidopsis*. Plant Cell 18:2971–2984

Wigge PA, Kim MC, Jaeger KE, Busch W, Schmid M, Lohmann JU, Weigel D (2005) Integration of spatial and temporal information during floral induction in *Arabidopsis*. Science 309:1056–1059

Yang Meiying HHWYGQFLZX (2007) Construction of efficient gene silencing vector harboring lycopene β-cyclase gene. Mol Plant Breeding 5:43–46

Zhang X, Henriques R, Lin S-S, Niu Q-W, Chua N-H (2006) *Agrobacterium*-mediated transformation of *Arabidopsis thaliana* using the floral dip method. Nat Protocols 1:641–646