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In silico analysis of three-amino acid loop extension (TALE) proteins in *Phaseolus vulgaris* L. and investigation of their transcript levels in response to drought stress

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Abstract: Drought represents a major global threat to the environment, thus leading to food insecurity. Transcription factors (TFs) have a significant role in regulating the expression of stress-related genes. Three-amino acid loop extension (*TALE*) genes, a type of TF, are known to regulate important biological processes like the maintenance of shoot apical meristems and root formation in plants. In this study, 35 *TALE* genes were identified and categorized into KNOX-like homeodomain (KNOX) and BLH/BELL-like subfamilies in *Phaseolus vulgaris* according to the phylogenetic analysis. The detailed characterization of the *PvTALE* genes using in silico analyses further showed that segmental duplications predominate in the evolution of TALE subfamilies and greatly contribute to the large-scale amplification of the gene family. Gene duplications, syntenic relationships, and the nonsynonymous/synonymous substitution ratios suggest intense purifying selection pressure during the evolution of *PvTALE*s. Furthermore, it was discovered that *PvTALE*s are regulated by a number of miRNAs and cis-elements, which play important roles in stress response signaling pathways in plants. The *PvTALE* genes showed tissue specific expression patterns and were differentially expressed in response to drought stress. Proline levels were measured in different tissues of *P. vulgaris* and were found to be accumulated under drought stress. Based on the obtained findings, it could be concluded that *PvTALE* genes may contribute to plant growth and drought stress resistance. The basis of the roles and resistance mechanisms of the PvTALE gene family were clarified with this first complete genome-wide study of its kind in *P. vulgaris*.

Key words: Common bean (*P. vulgaris* L.), expression profiling, genome-wide analysis, TALE gene family, transcriptomic analysis

1. Introduction

Homeobox genes are important transcription factors (TFs) that assist the formation of many structures during early embryonic development in many organisms (Duboule, 1994; Nam and Nei, 2005). Homeobox genes are represented by a large family of genes, and they all share a DNA binding domain known as a homeodomain (HD) with 60 amino acids (aa) (Desplan et al., 1988; Mukherjee et al., 2009). The two primary categories of HDs are atypical and typical, which are both determined by a conserved amino acid sequence of HDs and the existence of other distinguishing motifs (Burglin, 1994). Accordingly, the three-amino acid loop extension (*TALE*) genes are known to be atypical HDs (Bürglin, 1997; Chen et al., 2003).

A total of 14 homeobox gene families [Zinc finger-HD (PLINC), PINTOX, PHD, BLH, NDX (Nodulin homeobox genes), KNOX (KNOTTED1-like homeobox), SAWADEE, HD-leucine zipper (HD-ZIP) I to IV, DDT, WUSCHEL-like homeobox (WOX), and Luminidependens (LDs)] have previously been identified according to their homeobox domain sequences, until today (Que et al., 2022). *TALE* genes differ from other homeobox genes because they have an extra three residues between the first and second helix in their protein structure (Duboule, 1994; Bürglin, 1995). They are known to control the development of all eukaryotic lineages and play important roles in biological processes including root formation and maintenance of shoot apical meristem in plants (Bürglin, 1997).

Based on the protein sequence and evolutionary events, TALE proteins are known to be divided into two subfamilies, as the KNOTTED-like homeodomain (KNOX) and the BEL1-like homeodomain (BLH/BELL) subfamilies, and they both function as heterodimers and regulate the transcription of target genes (Lee et al., 2008). On the basis of sequence similarity and the gene expression pattern, the KNOX family is further divided into three groups as Class I (STM, BP/KNAT1, KNAT2, and KNAT6), Class II (KNAT3, KNAT4, and KNAT5), and Class III (KNATM). Each of these groups can interact with one another to modify the actions of other TALE members (Kimura et al., 2008). The roots, stems, leaves,

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and flowers of angiosperms are known to express various members of the KNOX gene family (Guo et al., 2022). *KNOX* gene overexpression in tobacco and rice lines was found to cause aberrant leaf phenotypes, according to previous studies (Tamaoki et al., 1997; Nagasaki et al., 2001). Additionally, the metabolic activities and signaling pathways of phytohormones, including cytokinins, gibberellins, auxins, brassinosteroids, and abscisic acid, were found to be regulated by *KNOX* genes (Becker et al., 2002; Hay and Tsiantis, 2010).

Members of the BELL subfamily, which is the second subfamily of the TALE proteins, include those whose functions are known (RPL/BLH9, PNF/BLH8, ATH1, SAW1/BLH2, SAW2/BLH8, and BEL1) and those whose functions are still unknown (BLH1, BLH3, BLH5, BLH6, BLH7, BLH10, and BLH11) (Hamant and Pautot, 2010). Members of the BELL subfamily with clearly defined roles are particularly recognized to play significant roles in controlling flower development. The *KNOX* and *BELL* genes can interact with different proteins and thus can participate in various other plant functions (Smith et al., 2002). Several KNOX proteins, including Shoot Meristemless (STM), KNAT1, and BELL1 proteins, have been found to interact with one another to trigger the transcription of specific genes in *A. thaliana*. In conclusion, *TALE* genes have been found to control many developmental processes by either themselves or forming heterodimers throughout the life cycle of a plant (Arnaud and Pautot, 2014).

It is known that exposure to drought stress in plants activates stress protection systems at biochemical, physiological, and molecular levels (Capell et al., 2004). The accumulation of metabolites such as proline, which is the most frequently synthesized osmotic protectant, is one of the effective biochemical responses of plants to drought stress. In a previous study by Kayak et al. (2023), they reported that the proline accumulation was significantly found to support the plant growth under stress conditions. Moreover, Çatav et al. (2022) stated that proline has important roles in the removal of reactive oxygen species and maintenance of cellular homeostasis in plants. In addition to its antioxidant role, proline is also involved in the maintenance of macromolecule stability with its role as an amino acid. It has been determined that proline significantly alleviates the harmful effects of drought stress by increasing the activity of antioxidant enzymes, and thus provides stability to proteins and cell membranes by scavenging free radicals (Ali et al., 2013).

In this study, *TALE* genes were determined and characterization and bioinformatics analyses were carried out for them in *P. vulgaris* for the first time. Following this, a proline assay was conducted and the mRNA levels of the *PvTALE* genes were quantified via reverse transcription-quantitative polymerase chain reaction (RT-qPCR), taking into account the publicly available RNAseq data under drought stress.

2.Materials and methods

2.1.Identification of the TALE proteins in P. *vulgaris*

The peptide sequences of the TALE proteins in *P. vulgaris* were obtained from Phytozome v13[1] via Pfam code search (Pfam IDs: PF07526-Associated with HOX, PF05920-Homeobox KN domain, PF03790-KNOX1 domain, PF03791-KNOX2 domain, PF03789-ELK domain, and PF05920-Homeobox KN domain) against a reference genome, namely *Phaseolus vulgaris* v2.1 (Phytozome genome ID: 442: National Center for Biotechnology Information (NCBI) taxonomy ID: 3885 JGIAP: 1352627; Goodstein et al., 2012). The redundant sequences were then determined and eliminated using a decrease redundancy[2] tool. Following this, the HMMER[3] tool was used to search for TALE domains in the identified PvTALE proteins, and NCBI Basic Local Alignment Search Tool-Primer (BLASTP)[4] analysis was carried out to find sequences similar to the PvTALE proteins derived from the Phytozome v13 database. The physicochemical characteristics of the identified PvTALE proteins were determined using the ProtParam[5] Tool and the cellular localizations of these proteins were predicted using the WoLF PSORT[6]: Protein Subcellular Localization Prediction Tool. Moreover, nuclear export signals (NES) and nuclear localization signals (NLS) of the PvTALE proteins were defined through the NLSdb[7] database (Tables S1 and S2).

2.2. Phylogenetic, protein motif, and gene structure analyses of the PvTALE gene family members

The peptide sequences of the PvTALE proteins were aligned using the ClustalW algorithm and phylogenetic trees were then drawn based on the maximum likelihood (ML) approach in Molecular Evolutionary Genetics

[1]Goodstein et al. (2012). Phytozome v13 [online]. Website https://phytozome-next.jgi.doe.gov/ [accessed 14 March 2023].

[2]Barlow (2001). Decrease Redundancy [online]. Website https://web.expasy.org/decrease_redundancy/ [accessed 14 March 2023].

[3]Potter et al. (2018). HMMER [online]. Website https://www.ebi.ac.uk/Tools/hmmer/ [accessed 14 March 2023].

[4]Mount (2007). NCBI BLASTP [online]. Website https://blast.ncbi.nlm.nih.gov/Blast.cgi?PAGE=Proteins [accessed 14 March 2023].

[5]ProtParam E (2017). ProtParam [online]. Website https://web.expasy.org/protparam/ [accessed 14 March 2023].

[6]Horton et al. (2006). WoLF PSORT [online]. Website https://wolfpsort.hgc.jp/ [accessed 14 March 2023].

[7]Nair et al. (2003). NLSdb [online]. Website https://ngdc.cncb.ac.cn/databasecommons/database/id/5672 [accessed 14 March 2023].

Analysis (MEGA) v11.[8] The ML approach was used since it is known to be the most realistic and accurate statistical approach for generating phylogenetic trees using peptide sequences according to Tamura et al. (2011). Interactive tree of life (iTOL)[9] was employed to display the final phylogenetic tree.

The Multiple Expectation Maximizations for Motif Elicitation (MEME)[10] tool was utilized for determining the conserved motifs in PvTALE proteins. The physical structures of *PvTALE* genes were analyzed using the Gene Structure Display Server v2.0 (GSDS).[11] The obtained data were finally visualized using Tbtools.[12]

2.3. Evolution of the duplications and localization analyses of the *PvTALE* genes on the chromosomes

The Phytozome v13 database was used to find the placement of the *PvTALE* genes on the chromosomes and the obtained data were then visualized using TBtools. *PvTALE* gene duplications and orthologs were determined using the Phytozome v13 database and TBtools; the Phytozome v13 database was used to determine the genetic relationship between *P. vulgaris* and 5 other species (*Arabidopsis thaliana, Glycine max, Oryza sativa, Solanum lycopersicum*, and *Vigna unguiculata)*. The genome files (FASTA) and genomic annotation files (GFF3) of the *TALE* gene families from related plant species were downloaded, and the collinearity analysis was performed using the Text Merge for MCScanX function in TBtools. Finally, the results were visualized using the Multiple Synteny Plot function in TBtools. Moreover, the nonsynonymous/synonymous substitution (Ka/Ks) ratios were estimated using TBtools (Chen et al., 2020).

2.4. MicroRNAs (miRNA) and cis-element analyses of *PvTALE* genes

The cis-elements in the promoter region (1.5 kb genomic sequence) of the *PvTALE* genes were scanned using PlantCARE[13] and TBtools was then used to visualize the obtained data (Chen et al., 2020). miRBase v21.0[14] was used to download all of the plant miRNA sequences and the miRNA prediction was carried out using the psRNATarget server[15] (Zhang, 2005). The common bean expressed sequenced tags from the NCBI database were BLASTX[16]-searched against these in silico-predicted *PvTALE* miRNA targets using an $1e^{-10}$ threshold. Finally, a graph showing the *PvTALE* genes targeted by the miRNAs was drawn using Cytoscape v3.9.1.[17]

2.5.Determination of the transcript levels of the *PvTALE* genes in different plant tissues and under drought stress using RNAseq data

The data (transcript fragments per kilobase per million base pairs sequenced, FPKM), which show the transcript levels of the *PvTALE* genes in various plant tissues at different developmental stages, were downloaded from the Phytozome v13 database. The log base 2 (log2) transformed FPKM values were used to draw a heat map through CIMMiner.[18]

Publicly available RNAseq data with accession numbers of SRR8284481 (drought stress-treated leaf) and SRR8284480 (untreated leaf, control) were used to evaluate the transcript levels of the *PvTALE* genes in response to drought stress, as previously suggested by Aygören et al. (2023). Next, the heat map, which shows the mRNA levels of the *PvTALE* genes during drought stress and the control leaves, was generated using CIMMiner.

2.6 Plant growth, stress treatment, and proline analysis

The Traditional Regional Agricultural Research Institute in Eskişehir, Türkiye, provided the seeds of the research material, which is a Turkish common bean cultivar, namely Zulbiye. It is a drought-susceptible cultivar according to previous studies (Güler et al., 2012; Büyük and Aras, 2017; Kabay et al., 2017; Okay et al., 2024). To comprehensively analyze the mRNA changes in the *PvTALE* genes in response to drought stress, a drought-susceptible cultivar was selected in which stress could be sensed at earlier stages and drought related symptoms could be more obvious.

The seeds of Zulbiye cv. were germinated following the surface sterilization for 5 min in a solution including 5% (v/v) hypochlorite (Büyük et al., 2016; İnal et al., 2017). The sterilized seeds were planted in pots and then the seeded

[8]Tamura (2021). MEGA11 [software]. Website https://www.megasoftware.net/ [accessed 15 April 2023].

[9]Letunic and Bork (2021). iTOL v5 [online]. Website https://itol.embl.de [accessed 15 April 2023].

[10]Bailey (2015). MEME [online]. Website https://meme-suite.org/meme/ [accessed 16 April 2023].

[11]Guo (2007). GSDS [online]. Website http://gsds.gao-lab.org [accessed 16 April 2023].

[12]Chen (2018). TBtools [software]. Website https://cj-chen.github.io/tbtools/ [accessed 16 April 2023].

[13]Rombauts (1999). PlantCARE [online]. Website https://bioinformatics.psb.ugent.be/webtools/plantcare/html/ [accessed 07 March 2023].

[14]Griffiths (2006). miRBase v21.0 [online]. Website https://mirbase.org/ [accessed 08 March 2023].

[15]Zhang (2005). psRNA Target Server [online]. Website https://www.zhaolab.org/psRNATarget/ [accessed 08 March 2023].

[16]NCBI Resource Coordinators. (2016). BLASTX [online]. Website https://blast.ncbi.nlm.nih.gov/Blast.cgi?PROGRAM=blastx&PAGE_TYPE=BlastSearch&LINK_LOC=blasthome [accessed 08 March 2023].

[17]Kohl (2011). Cytoscape 3.9.1. [online]. Website https://cytoscape.org/ [accessed 30 April 2023].

[18]Zeeberg (2004). CIMMiner [online]. Website https://discover.nci.nih.gov/cimminer/home.do [accessed 23 June 2023].

pots were then moved to a growth chamber with set at 25 °C, 70% relative humidity, and a flux of 250 mmol m^{-2} s^{-1} of photosynthetic photons. The shelf positions of the pots in the growth chamber were changed daily in order to allow the plants to get light uniformly. At the V3 (first trifoliate leaf, day 8 after planting) stage of the Zulbiye cv. development, the plants were separated into two groups as the control and stress groups. After which, the control group continued to be watered; however, water was withheld for the stress group until signs and symptoms of drought were observed in the growth, height, and weight of the plants. Throughout the experiment, the pot weights were measured daily so the experiment could be stopped at 20% water loss. The stress group reached to 20% water loss at the end of the day 16 with drought symptoms such as decreases in height, and having curved and pale leaves because of water losses. However, the control group was pot weights stayed at 80% or above throughout the experiment. On day 16 of the experiment, the plants were harvested for further analyses (three biological replicates were taken from tissues of the drought stress and control groups). The proline assay was performed in accordance with the method of Bates et al. (1973).

2.7 RNA isolation, cDNA synthesis, and qPCR analyses

The Favorgen Total Plant RNA Isolation Mini Kit (Biotech Corp., Ping-Tung, Taiwan) was used to extract the RNAs from the harvested plant materials. The extracted RNAs were then checked with agarose gel electrophoresis (1.6%) and a NanoDrop Lite spectrophotometer (NanoDrop Technologies, Wilmington, DE, USA) in terms of quality and quantity. For the cDNA synthesis, the Transcriptor First Strand cDNA Synthesis Kit (Roche Diagnostics, Basel, Basel-Stadt, Switzerland) was used. The qPCR primers were designed using the PrimerQuest Tool.[19] The qPCR reactions were done using iTaq Universal SYBR Green Supermix (Bio-Rad, Hercules, CA, USA) according to the recommendations by Büyük et al. (2019). A Mic qPCR instrument (Biomolecular Systems, Upper Coomera, Queensland, Australia) was used for the RT-qPCR assays. The actin gene was selected as a reference gene and the raw qPCR data were normalized using the $2^{-\Delta\Delta CT}$ formula (Livak and Schmittgen, 2001). GraphPad Prism 9[20] was used for the statistical analyses. Two-way ANOVA and Fisher's least significant difference test were applied at a significance level of 0.05.

3. Results

3.1 Characteristics of the PvTALE gene family members in *P. vulgaris*

In this study, 35 *PvTALE* genes were identified in the *P. vulgaris* genome, which were named from *PvTALE-01* to *PvTALE-35* according to their chromosomal positions. Additionally, some of the fundamental characteristics of the proteins encoded by these *PvTALE* genes were identified, such as the protein length (aa, amino acid), molecular weight (MW), isoelectric point (pI), and subcellular localization (Table S1). Accordingly, the lengths, molecular weights, and pI values of the PvTALE proteins ranged from 127 (PvTALE-12) to 785 aa (PvTALE-20), from 14149.01 (PvTALE-12) to 87184.43 Da (PvTALE-20), and from 4.77 (PvTALE-18) to 8.47 (PvTALE-19), respectively. The subcellular localization analyses indicated the presence of all PvTALE proteins in the nuclear region of the cell with one exception, in which the PvTALE-12 protein was detected in the mitochondria (Table S1).

3.2 Phylogenetic analysis of the PvTALE proteins

The peptide sequences of TALE proteins from *P. vulgaris* (35 TALE proteins), *A. thaliana* (22 TALE proteins), and *O. sativa* (29 TALE proteins) were aligned using the ClustalW algorithm and a phylogenetic tree was then drawn based on the ML approach in MEGA11. A total of 86 TALE proteins from the specified plant species were classified into two subfamilies, BELL1-like and KNOX, based on domain differences. According to the phylogenetic tree, the BELL1-like subfamily included 19 TALE proteins from *P. vulgaris*, 13 TALE proteins from *A. thaliana*, and 13 TALE proteins from *O. sativa*. On the other hand, the KNOX subfamily harbored 16 TALE proteins from *P. vulgaris*, nine TALE proteins from *A. thaliana*, and 16 TALE proteins from *O. sativa* (Figure 1).

3.3. Gene structure and conserved protein motifs of the PvTALEs

To understand the structure of the PvTALE proteins, 10 protein motifs were evaluated. Accordingly, the PvTALE proteins were classified into two subfamilies including subgroups with distinct domains, such as BEL1-like, KNOX1, KNOX2, and KNOX3 (Figure 2). Each of these clusters had similar motif patterns. As a result of the domain analysis, all members of the BEL1-like subfamily were found to include both POX and Homeodomain_KN domains. On the other hand, members of the KNOX subfamily with KNOX-I, KNOX-II, and KNOX-III subgroups were found to have ELK and Homeobox_KN domains in addition to their conserved KNOX-I and II domains (Table S3).

The MEME online tool was used to perform motif scanning, which helped to better demonstrate the conserved motif patterns of the PvTALE members. PvTALE members from the same subfamily were found to have similar motif compositions. All of the BEL1-like members contained Motifs 1, 2, 3, and 10; however, all of the KNOX members contained Motifs 5 and 6 (Figure 2). The KNOX1 subgroup contained

[19] PrimerQuest Tool [online]. Website https://www.idtdna.com/pages/tools/primerquest [accessed 18 June 2022].

[20] Prism (2020). GraphPad Prism [software]. Website https://www.graphpad.com/features [accessed 30 July 2023].

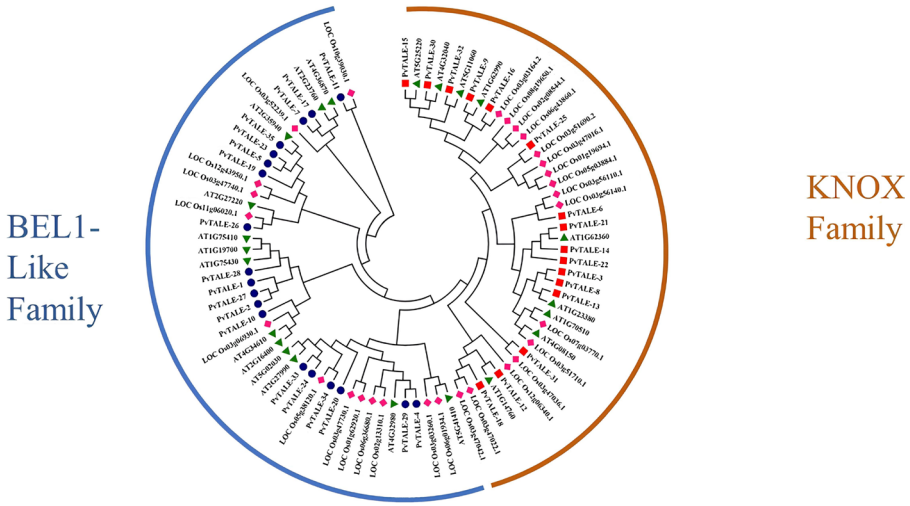

Figure 1. Phylogenetic tree based on the peptide sequences of 86 TALE proteins from *P. vulgaris*, *A. thaliana*, and *O. sativa*.

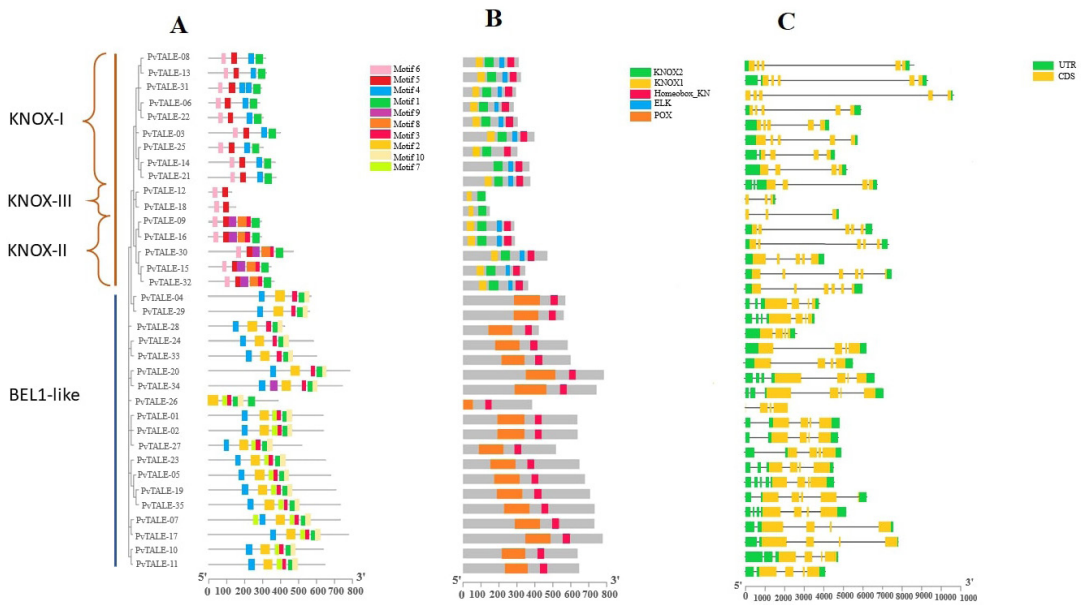

Figure 2. Motifs, conserved domains, and gene structures of the PvTALEs. **A.** Motif composition of the PvTALE proteins. **B.** Conserved domains of the PvTALE proteins. **C.** Gene structures of the *PvTALE* genes. Parts indicated by the lines are introns, while the yellow boxes are exons.

Motifs 1, 4, 5, and 6; however, the KNOX2 subgroup had Motifs 1, 3, 5, 6, 8, and 9. Additionally, the KNOX3 subgroup only contained Motifs 5 and 6 (Figure 2).

Gene structure analyses of the *PvTALE* genes were done using the Gene Structure Display database (Hu et al., 2015). Accordingly, the *PvTALE* genes were found to contain 4–6 exons and 4–7 introns. Similar gene structures were identified among *PvTALE* genes belonging to the same subfamily. Notably, all genes in the BEL1-like subfamily included four exons (Figure 2).

3.4. Chromosomal distribution of the *PvTALE* genes

All of the *P. vulgaris* chromosomes contained *PvTALE* genes in varying numbers (Figure 3). Chr09 was the chromosome with the most *PvTALE* genes (n = 7), while Chr04, Chr08, and Chr10 contained the fewest *PvTALE* genes (n = 1) (Figure 3).

3.5. Identification of the *PvTALE* gene duplications and orthologs

Segmental and tandem gene duplications have great importance in the formation of gene families (Cannon et al., 2004). According to Holub (2001), tandem duplications

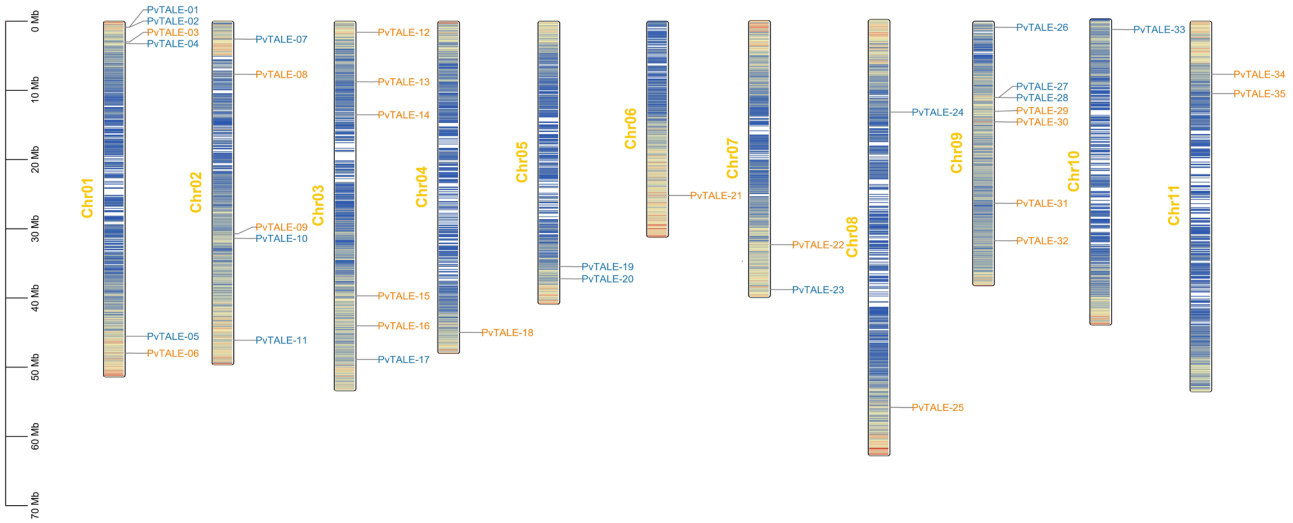

Figure 3. Distribution of the *PvTALE* genes on 11 *P. vulgaris* chromosomes. The Mb (Megabases) was used to measure the size of the chromosomes. The names of the genes belonging to the KNOX and BEL1-like subfamilies are shown in orange and blue, respectively.

occur as a result of gene clustering in areas smaller than 200 kb on the same chromosome of an organism. In this study, only one tandem duplication event was detected between the *PvTALE-10* and *PvTALE-11* genes, which were located in Chr02 (Figure 4). Segmental duplications are the duplications of gene clusters between different chromosomes (Leister, 2004; Panchy et al., 2016). In this study, six segmentally duplicated gene pairs *(PvTALE-24/PvTALE-33, PvTALE-16/PvTALE-09, PvTALE-15/PvTALE-32, PvTALE-15/PvTALE-29, PvTALE-22/PvTALE-06,* and *PvTALE-23/PvTALE-05)* were detected (Figure 4). Therefore, the segmental duplication events were found to contribute more to the expansion of the TALE gene family than tandem duplications in *P. vulgaris*. Some of the details of detected in the gene duplication events between the *PvTALE* genes are provided in Table S4.

The *TALE* genes from three dicot (*Arabidopsis thaliana*, *Vigna unguiculata*, and *Solanum lycopersicum*) and two monocot (*Glycine max* and *Oryza sativa*) plant species were also compared to understand the evolution of *TALE* genes between different plant species. Accordingly, a total of 23, 48, 27, 65, and 14 *PvTALE* orthologous genes were identified in *A. thaliana*, *V. unguiculata*, *S. lycopersicum*, *G. max*, and *O. sativa*, respectively (Figure 5). The Ka/Ks ratios of these orthologous *TALE* gene pairs between *P. vulgaris* and other plant species were calculated, and different Ka/Ks ratios were observed. The majority of the detected orthologous *TALE* gene pairs had Ka/Ks ratios of less than 1, demonstrating that the PvTALE superfamily might have been subjected to intense purifying selection pressure during the evolution of *P. vulgaris* (Table S5).

3.6. Cis-element analysis of the *PvTALE* genes

Cis-elements are essential for controlling gene transcription processes (Wang et al., 2017). Therefore, the promoter regions of the *PvTALE* genes were analyzed using a 1500 upstream sequence of each gene to define the cis-elements. As a result, the cis-elements found in the promoter regions of the *PvTALE* genes were grouped into the following categories: gibberellin-sensitive, light-sensitive, abscisic acid-sensitive, drought-inducible, auxin-sensitive, defense and stress response, seed-specific regulation, MeJA sensitive, circadian control, anaerobic induction, and salicylic acid-sensitive cis-elements (Figure 6, Table S6). The detected cis-elements showed that *PvTALE* genes might play important roles in a variety of biological processes including hormonal regulation and abiotic stress response. The promoter sequences of the *PvTALE* genes and detected cis-elements are provided in Table S6.

3.7. Transcript levels of the *PvTALE* genes in different tissues of *P. vulgaris* via RNAseq data analysis

The mRNA levels of the *PvTALE* genes in the flower buds, flowers, nodules, and roots were obtained from the Phytozome database. The downloaded fragments per kilobase of transcript per million mapped reads values were then transformed into log2 prior to generating the heatmap using TBtools (Figure 7). According to the heat map, the *PvTALE* genes were expressed at varying levels in different parts of *P. vulgaris*. *PvTALE* genes belong to different subfamilies based on their phylogeny generally showed differential expression patterns. The *PvTALE-12* and *PvTALE-18* genes from the KNOX-III subgroup showed the lowest expression levels, whereas all of the KNOX-II members revealed high expression levels in

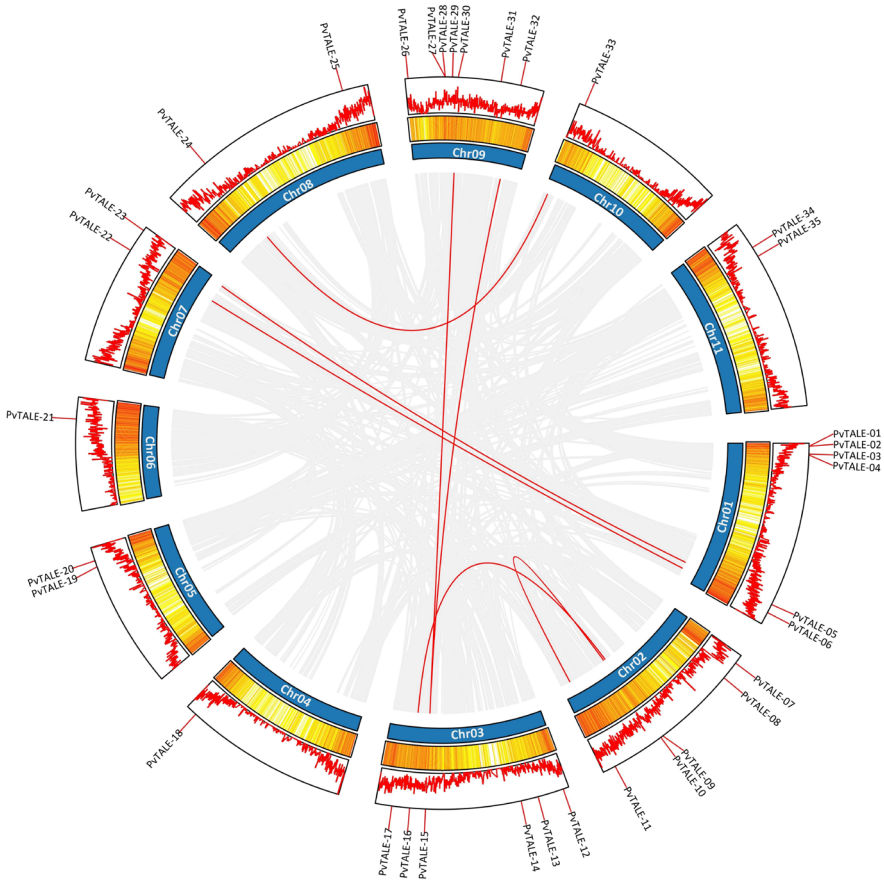

Figure 4. Circos display and chromosomal localization of the *PvTALE* genes. Gray lines display all of the synteny blocks in the *P. vulgaris* genome, whereas red lines depict the duplicated *PvTALE* gene pairs.

almost all tissues of *P. vulgaris*. Moreover, members of the KNOX-I subgroup displayed relatively low expression levels in almost all plant tissues (Figure 7). Gene pairs with similar expression levels were located in the same clade of the phylogenetic tree, such as the *PvTALE-04/PvTALE-29*, *PvTALE-24/PvTALE-33*, *PvTALE-01/02/27*, *PvTALE-05/19/35*, *PvTALE-07/17*, and *PvTALE-10/11* gene pairs (Figures 1 and 7).

3.8. Expression analysis of the *PvTALE* genes in *P. vulgaris* during drought stress through RNAseq and RT-qPCR

Using previously published RNA-seq data in the NCBI database, the expression levels of the *PvTALE* genes in response to drought stress in the leaves of *P. vulgaris* were retrieved. Then, a heatmap which shows the differential expression levels of the *PvTALE* genes under drought stress was drawn using log2 normalized RPKM values and is shown in Figure 8.

According to Figure 8, the KNOX-III subgroup genes showed comparatively low expression levels in contrast to the KNOX-II subgroup genes with high expression levels in response to drought stress. In the KNOX-I subgroup, the *PvTALE-03*, *PvTALE-06*, *PvTALE-21*, and *PvTALE-22* genes displayed relatively high expression levels; however, the *PvTALE-08*, *PvTALE-13*, *PvTALE-14*, *PvTALE-31*, and *PvTALE-25* genes showed low expression levels against drought stress (Figure 8). The *PvTALE-01*, *PvTALE-02*, *PvTALE-05*, *PvTALE-07*, *PvTALE-10*, *PvTALE-11*, *PvTALE-17*, *PvTALE-19*, *PvTALE-23*, *PvTALE-24*, *PvTALE-26*, *PvTALE-27*, *PvTALE-33*, and *PvTALE-35* genes from the BEL1-like subfamily exhibited high expression levels, while only the *PvTALE-04*, *PvTALE-20*, *PvTALE-28*, *PvTALE-29*, and *PvTALE-34* genes revealed low expression levels in response to drought. As a result, the most of the *PvTALE* genes were differentially expressed in response to drought stress in *P. vulgaris*, according to the analyzed RNAseq data with Sequence Read Archive accession numbers of SRR8284481 (drought stress-leaf-*P. vulgaris*) and SRR8284480 (untreated control, leaf, *P. vulgaris*) in the NCBI database (Figure 8).

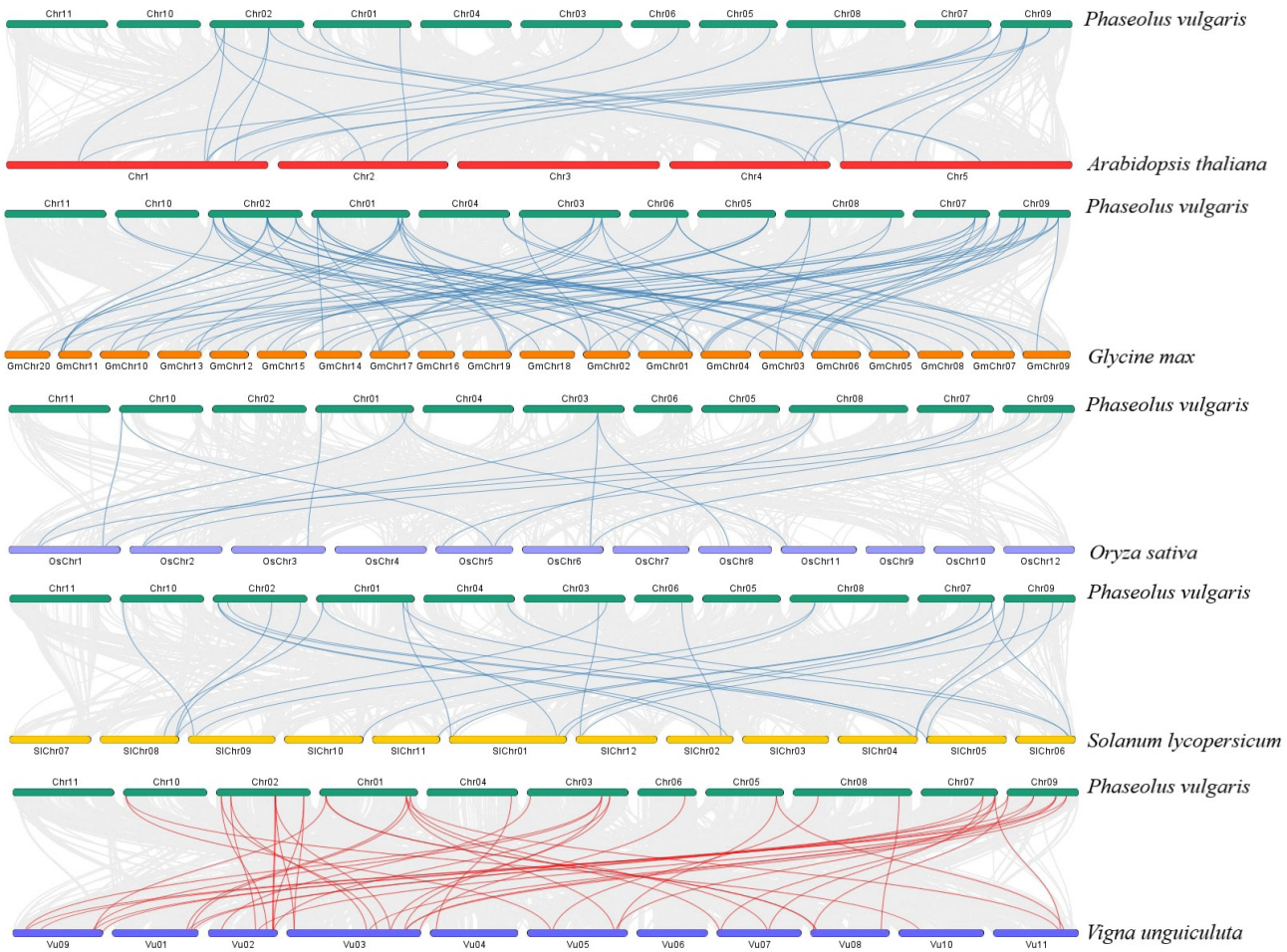

Figure 5. Syntenic relations of the *TALE* genes from *P. vulgaris* and five other plant species. The gray areas in the background show the collinear areas in *P. vulgaris* and five other plant species, while the blue lines show the syntenic orthologous gene pairs.

3.9. miRNAs targeting the *PvTALE* genes

Using the PmiREN[2121] online tool, mature-miRNA sequences from *P. vulgaris* were obtained, and the miRNAs targeting the *PvTALE* genes were examined using the psRNATarget online tool (Table S7). Additionally, Cytoscape v3.9.1 was used to display the associations between the plant miRNAs and *PvTALE* genes (Figure 9). Accordingly, 20 different miRNAs (Pvu-miR5570, Pvu-miR2118, Pvu-miR172, Pvu-miRN2587, Pvu-miRN2590, Pvu-miR395, Pvu-miR159, Pvu-miR319, Pvu-miR160, Pvu-miR1514, Pvu-miRN2588, Pvu-miRN2594, Pvu-miR397, Pvu-miRN2599, Pvu-miR171, Pvu-miR169, Pvu-miRN2591, Pvu-miRN2600, Pvu-miR396, and Pvu-miR4415) were found to target *PvTALE* genes (Figure 9).

3.10. Proline analysis in *P. vulgaris* under drought stress

In higher plants, proline accumulation is an indicator of a disrupted physiological state brought on by biotic or abiotic stress conditions (Hosseinifard et al., 2022). In this study, the proline levels were measured and determined to be significantly increased in the leaf, stem, and root tissues of *P. vulgaris* under drought stress. This increase was more pronounced in the stem tissue than in the leaf and root tissues under drought stress compared to the control (Figure 10).

3.11. RT-qPCR analysis of *PvTALE* genes during drought stress

Ten representative *PvTALE* genes were chosen for the RT-qPCR assays according to the following criteria: 1) genes with relatively high expression levels in the studied plant parts according to the RNAseq data and from different PvTALE subfamilies or subgroups, 2) genes that showed differential expressions in response to drought stress according to the RNAseq data, and 3) genes harboring cis-elements that were shown to be stress responsive

[21] PmiREN tool [online]. Website https://pmiren.com/ [accessed 18 June 2022].

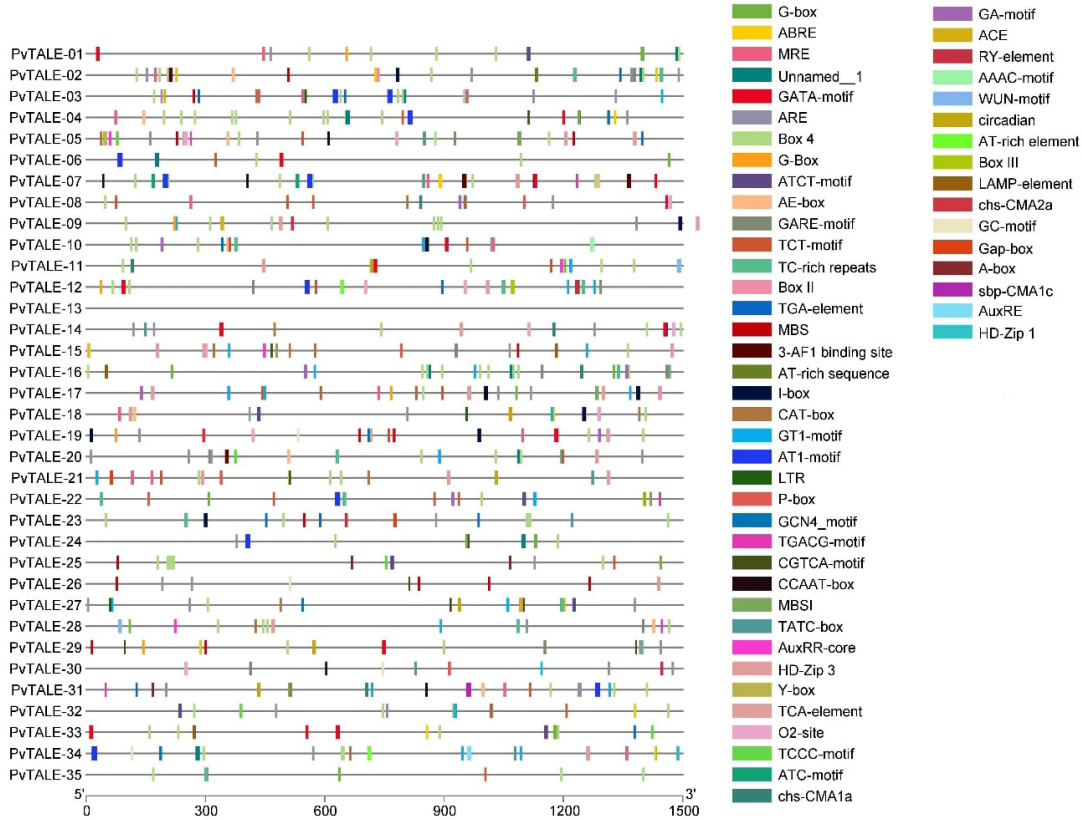

Figure 6. Distribution of the cis-elements in the upstream regions of the *PvTALE* genes spanning 1.5 kb. The colors represent the cis-elements given on the right side.

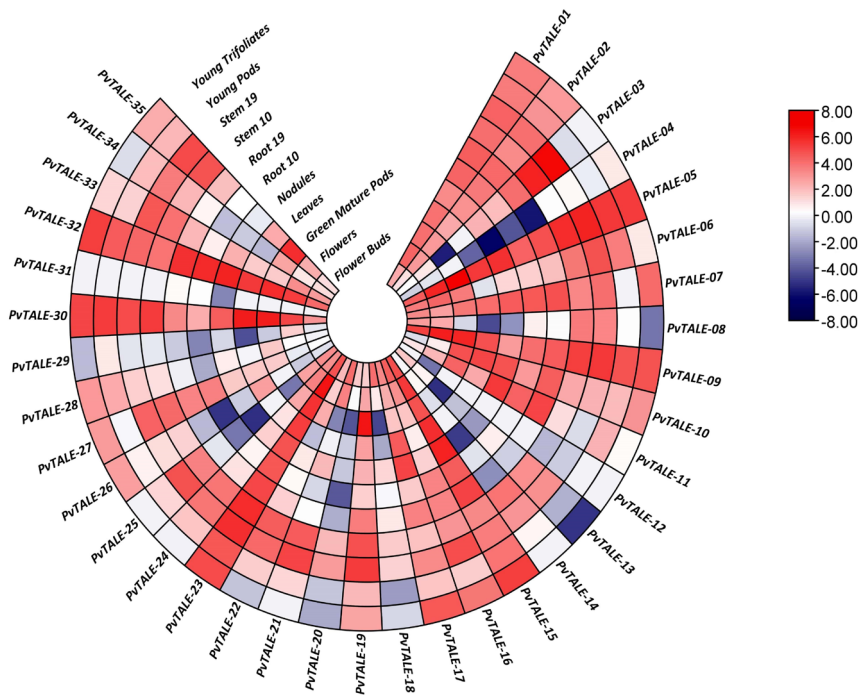

Figure 7. Differential gene expression levels of the *PvTALE* genes in different parts of *P. vulgaris*.

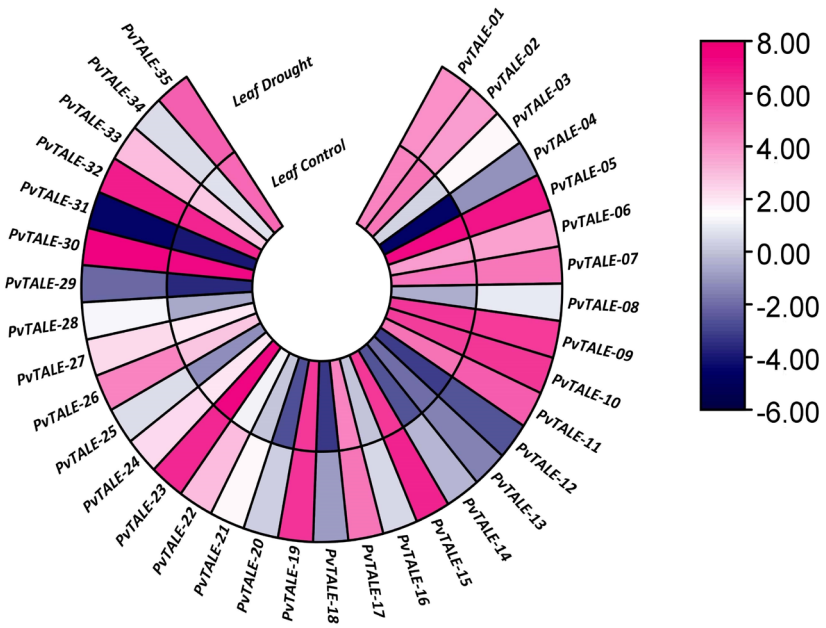

Figure 8. Differential gene expression levels of the *PvTALE* genes in response to drought stress in *P. vulgaris* based on the transcriptome RNAseq analysis.

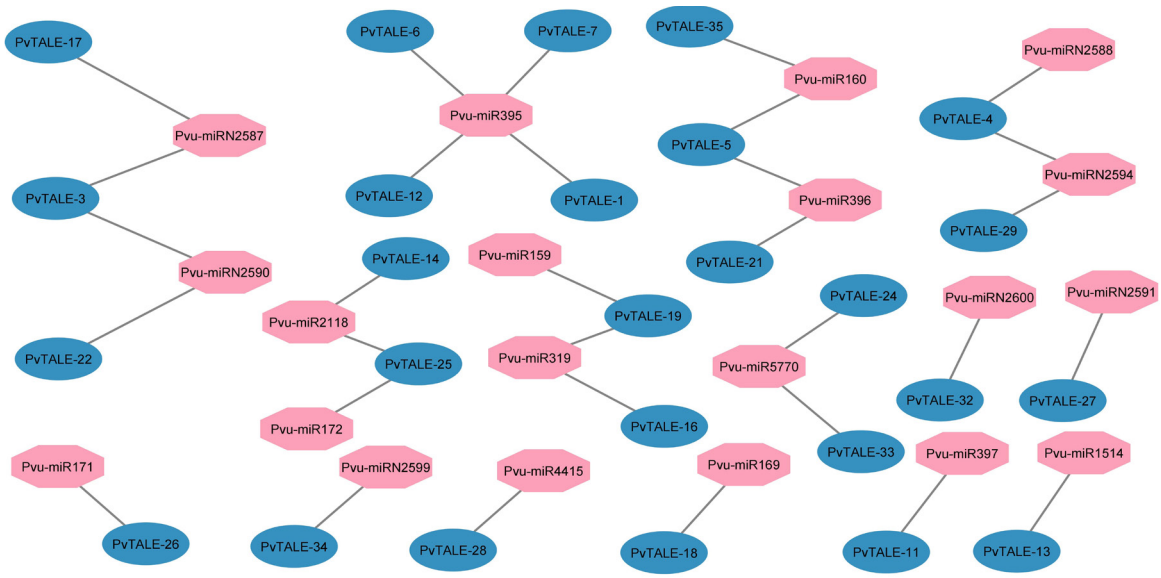

Figure 9. miRNAs targeting the *PvTALE* genes. Pink represents the miRNAs, while green represents the targeted gene.

according to the literature. In this regard, four genes *(PvTALE-06, PvTALE-16, PvTALE-22,* and *PvTALE-30)* from the KNOX subfamily and six genes *(PvTALE-10, PvTALE-20, PvTALE-27, PvTALE-28, PvTALE-29,* and *PvTALE-33)* from the BEL1-like subfamily were selected for RT-qPCR assays. The primers for those genes were designed and gene expression patterns were evaluated through RT-qPCR in the leaf, stem, and root tissues of *P. vulgaris* under drought stress. The primer sequences are provided in Table S8.

According to the RT-qPCR results, the *PvTALE-06, PvTALE-16, PvTALE-27, PvTALE-28* and *PvTALE-29* genes were upregulated in the plant leaves; however, they were down-regulated in the stem and root tissues of *P. vulgaris*. Additionally, the *PvTALE-10* and *PvTALE-30* genes were down-regulated in all tissues of *P. vulgaris* under drought stress (Figure 11). The transcript level of the *PvTALE-20* gene was increased in the leaf, stem, and root tissues; however, a statistically significant increase was only observed in the stem tissue of *P. vulgaris* under

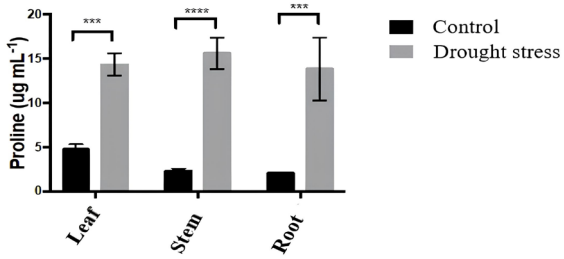

Figure 10. Proline levels in the leaves, stems, and roots of *P. vulgaris* under drought stress. Asterisks in the graph (**p < 0.01, ***p < 0.001, ****p < 0.0001) indicate the statistical significance level of the data.

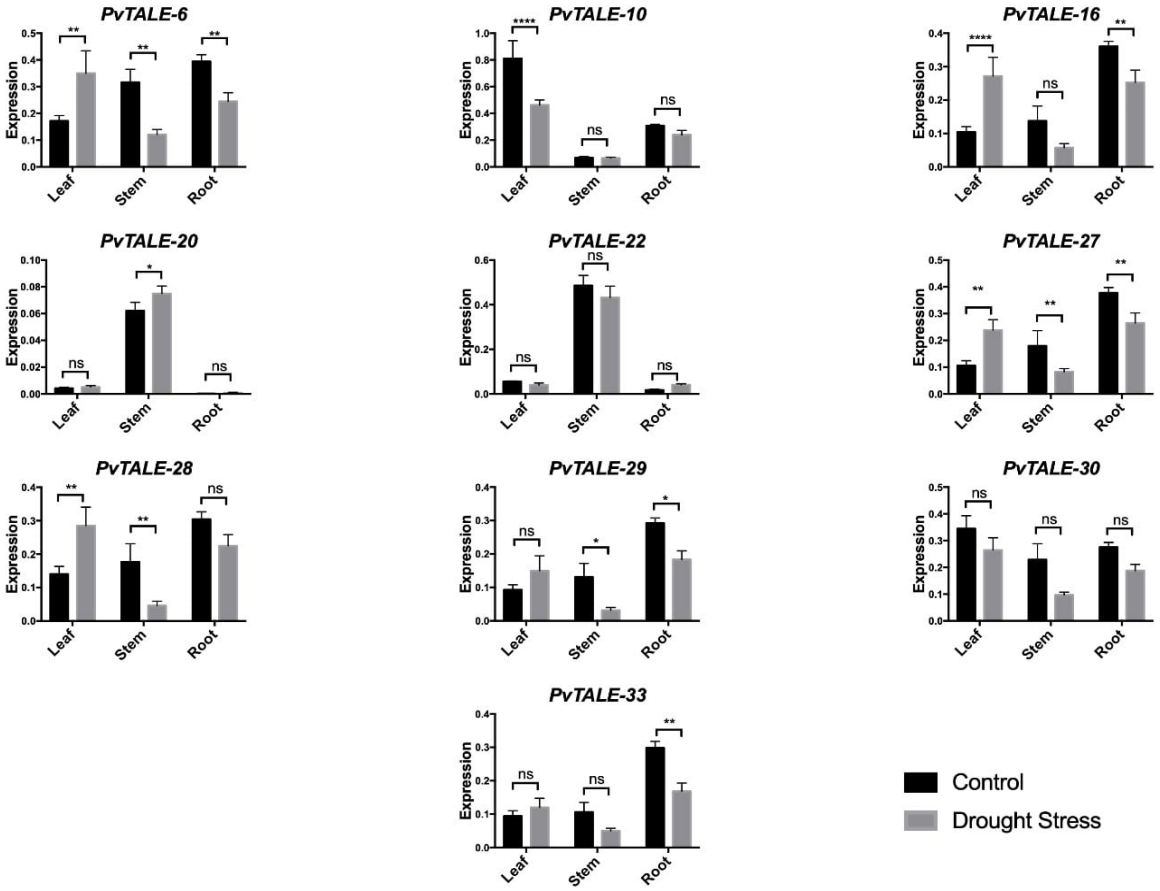

Figure 11. mRNA levels of the *PvTALE* genes in *P. vulgaris* against drought stress. Asterisks in the graph (*p < 0.05, **p < 0.01, ***p < 0.001, ****p < 0.0001, ns = nonsignificant) indicate the statistical significance level of the data (normalization was done with the *PvActin* gene and the vertical bars in the graph show the standard deviation).

drought stress. The expression level of the *PvTALE-33* gene was decreased in the stem and root tissues of *P. vulgaris*; however, only the decrease in the root tissue was statistically significant (Figure 11). Compared to the RNAseq data analyzed, the expression pattern of the selected *PvTALE* genes based on RT-qPCR were almost the same, except for the *PvTALE-10* and *PvTALE-28* genes in the *P. vulgaris* leaves (Figures 8 and 11). In conclusion, the studied *PvTALE* genes showed differential expression patterns in response to drought stress in *P. vulgaris*, suggesting their potential roles in defense or signaling mechanisms under stress conditions.

4. Discussion

TALE genes are widely dispersed throughout the genomes of plants and are crucial for plant development, growth, and stress responses (Han et al., 2022). The whole genomes of some plant species have been found to contain members of the TALE family due to recent, rapid breakthroughs in biotechnology and bioinformatics. However, in other plant species, there is still a need for the genome-wide discovery and characterization of TALE family members.

The number of TALE superfamily members was found to vary by species according to recent research. In the current study, more *TALE* genes (35 genes) were found in *P. vulgaris* than those identified in *A. thaliana* (22 genes) (Hamant and Pautot, 2010) and fern (22 genes) (Ruiz-Estévez et al., 2017). The number of *TALE* genes in *P. vulgaris* was the same as those in poplar (35 genes) (Zhao et al., 2019); however, it was less than those in walnut (69 genes) (Guo et al. 2022), cotton (94 genes) (Ma et al., 2019), and soybean (68 genes) (Wang et al., 2021a).

Herein, the protein lengths and molecular weights of the BEL-1 like subfamily members were higher than those in the KNOX subfamily in *P. vulgaris*. Previous studies also reported similar results for *G. max* (Wang et al., 2021a), *G. arboreum*, *G. raimondii*, *G. barbadense*, and *G. hirsutum* (Ma et al., 2019), and *P. Trichocarpa* genomes (Hou et al., 2021). It has been well established that introns contribute to the evolution of genomes (William Roy and Gilbert, 2006). In the present study, all of the *PvTALE* genes had introns with varying numbers. Remarkably, each of the PvTALE subfamilies and subgroups had similar gene structures and similar findings were already reported for *TALE* genes from *G. max* by Wang et al. (2021a).

Finding the subcellular localization of proteins is important since the protein activities are largely governed by where they are located inside the cells (Wang et al., 2021b). In the current study, the subcellular localization analyses indicated the presence of all PvTALE proteins in the nuclear region with one exception, in which the PvTALE-12 protein was located in the mitochondria. This protein was declared to have a role in the electron transport system in plants according to the information provided in the Uniprot database[2222] (Horton et al., 2006).

The BELL and KNOX proteins can combine to form heterodimers to change their positions within the cell. Increased DNA-binding specificity and affinity have been shown by KNOX/BELL heterodimerization, which is also associated with the nuclear import of plant TALE proteins (Rutjens et al., 2009; Zhao et al., 2019). Recently, Wang et al. (2021) found that 64 out of 68 TALE proteins were located in the nuclear region of *G. max* cells (Wang et al., 2021a). In a previous study by Han et al. (2022), most of the TALE proteins were also located in the nucleus of *T. aestivum* cells. Additionally, Han et al. (2022) also indicated that TALE proteins may be located in different cellular components like the mitochondria since they participate in various metabolic and developmental processes in plants (Han et al., 2022). The findings of these studies on TALE proteins were similar to the findings in the present study.

Different gene families have expanded as a result of the genome evolution process in plants, which frequently occurs via segmental and tandem duplications (Guo et al., 2022). The majority of *PvTALE* genes were discovered to undergone segmental duplication events according to the gene duplication analysis, highlighting that segmental duplications might play important roles in the expansion of the PvTALE gene family. Similar to the current findings, expansion of the TALE gene family in *Glycine max* (Wang et al., 2021a), *Triticum aestivum* (Han et al., 2022), and *Juglans regia* L. (Guo et al., 2022) was also found to have originated from segmental duplication events. In the present study, the *TALE* genes of three dicotyledons (*Arabidopsis thaliana*, *Vigna unguiculata*, and *Solanum lycopersicum*) and two monocotyledons (*Glycine max* and *Oryza sativa*) were used for the synteny analysis. A total of 177 *PvTALE* orthologous genes were identified in the *A. thaliana*, *V. unguiculata*, *S. lycopersicum*, *G. max*, and *O. sativa* genomes (Figure 5). Higher syntenic relationships were shown to be present between the *P. vulgaris* and *G. max* (soybean) and *P. vulgaris* and *V. unguiculata* genomes.

Many aspects of plant life are now understood to be regulated by miRNAs. Although only discovered in plants less than 20 years ago, previous studies have demonstrated that miRNAs are essential controllers of developmental processes such leaf morphogenesis, vegetative phase shift, blooming timing, and responsiveness to environmental signals (Li and Zhang, 2016; Millar, 2020; Sonkar et al., 2023). In the current study, Pvu-miR2118 and Pvu-miR159 were found to target the *PvTALE-25*, *PvTALE-14* and *PvTALE-19* genes in the *P. vulgaris* genome. These miRNAs were demonstrated to play important roles in drought and salt stress response in *P. vulgaris* by Arenas-Huertero et al. (2009).

Pvu-miR172, which targeted the *PvTALE-25* gene, is also another miRNA that has been reported to have crucial roles in stress response in *P. vulgaris* (Simon et al., 2009). Pvu-miR160 targeting the *PvTALE-5* and *PvTALE-35* genes was shown to respond to P deficiency and other abiotic stressors in *P. vulgaris* by Valdés-López et al. (2010). MiR395 and miR169, which targeted the *PvTALE-01, PvTALE-06,* and *PvTALE-18* genes in the present study, were previously shown to be involved in the P, S, and Cu homeostasis mechanism in *Brassica napus* L. (Buhtz et al., 2008). Pvu-miR319, which targeted the *PvTALE-16* and *PvTALE-19* genes, was previously described to respond to abiotic stresses in *Arabidopsis thaliana* and rice plants (Liu et al., 2019). Pvu-miR397, which targeted the *PvTALE-11* gene, was strongly inhibited in the root and leaf

[22] UniProt Consortium (2019). Uniprot database [online]. Website https://www.uniprot.org/ [accessed 14 March 2023].

tissues of *P. vulgaris* in response to manganese deficiency. Moreover, Pvu-miR1514, which targeted the *PvTALE-13* gene, has been shown to play a significant role in response to P, N, Fe, and Mn deficiency in *P. vulgaris* (Valdés-López et al., 2010). Pvu-miR171, which targeted the *PvTALE-26* in the current study, is another miRNA that was reported to respond to abiotic stress in common bean by Laskar et al. (2021). Hence, it could be concluded that almost all miRNAs targeting *PvTALE* genes were miRNAs that were previously identified as key players of stress response, especially drought, in several plant species including *P. vulgaris*.

Abiotic stressors including low or high temperatures, water shortages or surpluses, high salinity, heavy metals, and ultraviolet radiation, are harmful to plant growth and development and thus, have a significant negative impact on agricultural yields globally (He et al., 2018). *TALE* genes have been associated with abiotic stress responses in *G. max* (Wang et al., 2021a), *P. trichocarpa* (Hou et al., 2021), *T. aestivum* (Han et al., 2022), and *C. sinensis* genomes (Peng et al., 2022). Hao et al. (2021) suggested that *TALE* genes might play an important role in abiotic stress response through hormonal regulatory pathways. Wang et al. (2021a) discovered some cis-elements responsible for hormonal regulation in the promoters of *TALE* genes in soybean. *TALE* genes including these cis-elements were also found to play important roles in salt and drought stress responses. In another study conducted by Han et al. (2022), it was reported that the overexpression of *TaKNOX11-A*, the KNOX-like gene in *Triticum aestivum*, led to increased proline and decreased malondialdehyde content, and thereby enhanced the salt and drought tolerance in *A. thaliana*. Similar to their findings, significantly increased levels of proline were observed in the leaves, stems, and roots of *P. vulgaris* under drought stress in the current study.

Maize and sorghum are the main crops consumed in Africa and worldwide, and both are negatively affected by drought stress. However, sorghum has better drought tolerance than maize. A study by Ali et al. (2023) examined the protein levels associated with drought tolerance between maize and sorghum. According to their findings, drought resulted in higher proline levels in the roots of sorghum than in maize. In another study by Shan et al. (2018), the proline levels were significantly increased in the leaves of cassava plant in response to drought stress. Therefore, the current study investigated the proline levels in different tissues of *P. vulgaris* in response to drought stress and increases in the proline levels were also observed, in accordance with the findings of previous works. The increased levels of proline also showed that the plants were under stress and their stress response mechanisms were reacting against it.

In the present study, RT-qPCR analyses were performed to examine the mRNA levels of the *PvTALE* genes in different tissues of *P. vulgaris* under drought stress. The RT-qPCR data revealed that the *PvTALE-06*, *PvTALE-16*, *PvTALE-27*, and *PvTALE-28* genes were upregulated in response to drought stress in the leaves in contrast to the stem tissues of *P. vulgaris* (Figure 11). In a previous study by Wang et al. (2021a), it was also found that *GmTALE* genes were upregulated in response to drought and salt stress and the upregulations were more pronounced in the leaf tissues than the stem tissues of *G. max*.

5. Conclusion

In this study, 35 TALE proteins were identified and bioinformatically characterized for the first time in the *P. vulgaris* genome. The identified PvTALE proteins were categorized into BEL1-like and KNOX subfamilies, which were subsequently classified into KNOX-I, KNOX-II and KNOX-III subgroups. PvTALEs belonging to the same subfamily according to the phylogenetic tree shared similarities in the gene structures, protein motifs, conserved domains, and biological functions. The gene duplication analysis showed that the segmental duplications contributed to the evolution of the TALE gene family more than the tandem duplications in *P. vulgaris*. The *PvTALE* genes were found to have possible functions in plant developmental processes and abiotic stress responses in *P. vulgaris* through cis-elements, transcriptome data and RT-qPCR analyses. As a result, this study may have paved the way for future functional investigations on *PvTALE* genes, which could help to enhance the genetic makeup of *P. vulgaris* and make them more resilient to drought challenges.

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Availability of data and materials

All relevant data are provided within this manuscript and the supplementary file.

Conflict of interest

The authors declare that there are no conflicts of interest.

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Contribution of authors

İB conceived, planned, and oversaw the experiments. İB, FNA, and TK carried out the experiments for the bioinformatic analysis. İB, FNA, and AO carried out the experiments for the qRT-PCR analysis. İB, FNA, AO, and ESA analyzed and integrated the datasets and drafted the manuscript. ESA critically read and contributed to improving the manuscript.

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Supplementary Data

Table S1. A catalog *Phaseolus vulgaris* TALE gene family members.

ID	*Phaseolus vulgaris* genomic database identifier	Physical position on *P. vulgaris* genome			Protein length (aa)	pI
		Chromosome	Start position (bp)	End position (bp)		
PvTALE-01	Phvul.001G011000.1.v2.1	1	850225	855005	637	5.66
PvTALE-02	Phvul.001G011100.1.v2.1	1	861317	866004	638	5.84
PvTALE-03	Phvul.001G032200.1.v2.1	1	2986014	2991724	398	6.47
PvTALE-04	Phvul.001G034200.1.v2.1	1	3219786	3223533	570	5.81
PvTALE-05	Phvul.001G195800.1.v2.1	1	45522289	45526794	679	6.61
PvTALE-06	Phvul.001G224900.1.v2.1	1	47970467	47976339	283	6.14
PvTALE-07	Phvul.002G024800.1.v2.1	2	2580242	2587781	732	6.69
PvTALE-08	Phvul.002G064100.2.v2.1	2	7665196	7674753	311	5.16
PvTALE-09	Phvul.002G153300.2.v2.1	2	30701843	30708406	286	6.31
PvTALE-10	Phvul.002G159000.1.v2.1	2	31374897	31379625	638	6.08
PvTALE-11	Phvul.002G292000.1.v2.1	2	46088200	46092273	647	6.25
PvTALE-12	Phvul.003G016200.1.v2.1	3	1594869	1596388	127	5.09
PvTALE-13	Phvul.003G062500.1.v2.1	3	8739342	8750416	323	5.38
PvTALE-14	Phvul.003G081800.2.v2.1	3	13510855	13515994	370	6.52
PvTALE-15	Phvul.003G174900.1.v2.1	3	39677400	39684849	347	5.53
PvTALE-16	Phvul.003G213500.1.v2.1	3	44001710	44008951	289	6.20
PvTALE-17	Phvul.003G250500.1.v2.1	3	48873540	48881342	778	6.78
PvTALE-18	Phvul.004G148100.1.v2.1	4	44969689	44974427	149	4.77
PvTALE-19	Phvul.005G119300.3.v2.1	5	35459461	35465739	708	8.47
PvTALE-20	Phvul.005G132700.1.v2.1	5	37216524	37223084	785	6.37
PvTALE-21	Phvul.006G145800.1.v2.1	6	25180451	25187118	375	6.35
PvTALE-22	Phvul.007G201400.1.v2.1	7	32412462	32416692	305	5.16
PvTALE-23	Phvul.007G267900.1.v2.1	7	38901726	38906220	649	7.81
PvTALE-24	Phvul.008G114300.1.v2.1	8	13367165	13373327	583	6.92
PvTALE-25	Phvul.008G212100.1.v2.1	8	56038144	56042705	303	6.72
PvTALE-26	Phvul.009G004801.1.v2.1	9	864844	866980	385	6.12
PvTALE-27	Phvul.009G060400.1.v2.1	9	11028917	11033808	518	5.81
PvTALE-28	Phvul.009G060500.1.v2.1	9	11035125	11037662	422	6.23
PvTALE-29	Phvul.009G077100.1.v2.1	9	13001459	13004978	561	5.91
PvTALE-30	Phvul.009G090500.3.v2.1	9	14537220	14541202	470	5.98
PvTALE-31	Phvul.009G176750.1.v2.1	9	26285148	26295693	295	6.07
PvTALE-32	Phvul.009G209500.1.v2.1	9	31707622	31713542	363	5.43
PvTALE-33	Phvul.010G011900.1.v2.1	10	1529445	1534930	600	6.45
PvTALE-34	Phvul.011G082200.1.v2.1	11	7654972	7661970	744	6.42
PvTALE-35	Phvul.011G099100.1.v2.1	11	10442746	10447885	733	7.24

Table S1. A catalog *Phaseolus vulgaris* TALE gene family members (continued).

ID	Molecular weight (Da)	Instability index	Stable or unstable	Aliphatic index	GRAVY	NCBI accession number	Predictable subcellular location
PvTALE-01	70612.78	50.61	Unstable	74.73	–0.586	XP_007160715.1	Nuclear
PvTALE-02	70828.03	48.96	Unstable	75.06	–0.593	XP_007160717.1	Nuclear
PvTALE-03	45705.70	55.57	Unstable	53.72	–1.043	XP_007160965.1	Nuclear
PvTALE-04	62929.32	49.80	Unstable	79.65	–0.387	XP_007160994.1	Nuclear
PvTALE-05	73757.83	45.29	Unstable	64.55	–0.628	XP_007162972.1	Nuclear
PvTALE-06	32533.09	54.57	Unstable	70.00	–0.736	XP_007163321.1	Nuclear
PvTALE-07	79344.54	48.24	Unstable	62.69	–0.575	XP_007156875.1	Nuclear
PvTALE-08	35175.66	41.96	Unstable	68.42	–0.684	XP_007157365.1	Nuclear
PvTALE-09	32577.89	55.73	Unstable	85.24	–0.597	XP_007158449.1	Nuclear
PvTALE-10	71157.37	46.69	Unstable	63.42	–0.690	XP_007158519.1	Nuclear
PvTALE-11	72328.57	51.48	Unstable	63.89	–0.695	XP_007160096.1	Nuclear
PvTALE-12	14149.01	39.87	Stable	80.79	–0.507	XP_007153209.1	Mitochondria
PvTALE-13	36627.60	55.01	Unstable	74.06	–0.557	XP_007153755.1	Nuclear
PvTALE-14	41655.56	43.06	Unstablet	60.38	–0.789	XP_007153990.1	Nuclear
PvTALE-15	39724.16	52.94	Unstable	70.03	–0.853	XP_007155117.1	Nuclear
PvTALE-16	32833.05	54.07	Unstable	80.03	–0.608	XP_007155574.1	Nuclear
PvTALE-17	84399.36	54.87	Unstable	57.49	–0.691	XP_007156003.1	Nuclear
PvTALE-18	17167.37	73.36	Unstable	72.08	–0.809	XP_007152658.1	Nuclear
PvTALE-19	76554.01	44.47	Unstable	68.11	–0.609	XP_007150021.1	Nuclear
PvTALE-20	87184.43	49.25	Unstable	68.79	–0.614	XP_007150172.1	Nuclear
PvTALE-21	42000.00	39.41	Stable	59.33	–0.737	XP_007147684.1	Nuclear
PvTALE-22	34897.45	49.79	Unstable	78.98	–0.581	XP_007145002.1	Nuclear
PvTALE-23	71444.79	44.89	Unstable	65.53	–0.641	XP_007145789.1	Nuclear
PvTALE-24	64784.67	45.76	Unstable	73.91	–0.548	XP_007140461.1	Nuclear
PvTALE-25	34809.65	41.32	Unstable	66.67	–0.666	XP_007141628.1	Nuclear
PvTALE-26	42435.44	35.77	Stable	68.10	–0.529	XP_007135942.1	Nuclear
PvTALE-27	58337.29	44.90	Unstable	73.96	–0.621	XP_007136630.1	Nuclear
PvTALE-28	47463.78	48.24	Unstable	81.85	–0.415	XP_007136632.1	Nuclear
PvTALE-29	62509.35	50.24	Unstable	81.27	–0.371	XP_007136819.1	Nuclear
PvTALE-30	52754.12	41.14	Unstable	78.09	–0.607	XP_007136985.1	Nuclear
PvTALE-31	33700.15	34.74	Stable	72.07	–0.643	XP_007138056.1	Nuclear
PvTALE-32	40978.58	55.82	Unstable	70.96	–0.777	XP_007138444.1	Nuclear
PvTALE-33	66159.90	48.67	Unstable	71.30	–0.525	XP_007134014.1	Nuclear
PvTALE-34	82211.47	48.38	Unstable	64.38	–0.634	XP_007132286.1	Nuclear
PvTALE-35	79675.34	45.13	Unstable	64.17	–0.642	XP_007132496.1	Nuclear

Table S2. Results obtained from NLSdb database (a database of nuclear localization signals and of nuclear proteins).

Query	Signal	Signal type	Start	End	ConfNuc	ConfFam	Annotation type	Origin
PvTALE-13	RKRHW	NLS	297	301	4	3	By expert	Q09353
PvTALE-14	KRKKGK	NLS	276	281	3	3	Potential	In silico mutagenesis
PvTALE-14	RKRHW	NLS	330	334	4	3	By expert	Q09353
PvTALE-16	KSKRKR	NLS	280	285	3	3	Potential	In silico mutagenesis
PvTALE-21	KRKKGK	NLS	280	285	3	3	Potential	In silico mutagenesis
PvTALE-21	MKKRKK	NLS	278	283	2	2	Potential	In silico mutagenesis
PvTALE-21	RKRHW	NLS	334	338	4	3	By expert	Q09353
PvTALE-22	KRKKGK	NLS	223	228	3	3	Potential	In silico mutagenesis
PvTALE-22	LKKRKK	NLS	221	226	4	3	Potential	In silico mutagenesis
PvTALE-22	LKKRKKG	NLS	221	227	1	1	By expert	O55070
PvTALE-22	RKRHW	NLS	277	281	4	3	By expert	Q09353
PvTALE-25	KKKKNG	NLS	209	214	2	2	Potential	In silico mutagenesis
PvTALE-25	RKRHW	NLS	264	268	4	3	By expert	Q09353
PvTALE-3	RKRHW	NLS	357	361	4	3	By expert	Q09353
PvTALE-30	KSKRKR	NLS	411	416	3	3	Potential	In silico mutagenesis
PvTALE-31	RKRHW	NLS	271	275	4	3	By expert	Q09353
PvTALE-33	QGRKK	NLS	277	281	4	4	Potential	In silico mutagenesis
PvTALE-35	SPKRPR	NLS	582	587	3	3	Potential	In silico mutagenesis
PvTALE-6	RKRHW	NLS	252	256	4	3	By expert	Q09353
PvTALE-8	RKRHW	NLS	285	289	4	3	By expert	Q09353
PvTALE-9	KSKRKR	NLS	280	285	3	3	Potential	In silico mutagenesis

ConfNuc (confidence score for matching nuclear proteins) and ConfFam (confidence score for matching protein families). NLSdb: ConfNuc indicates the number of protein sequence in the NLS database that match the enquiry. ConfFam shows the protein families in which NLS motifs matching the query in the NLS database are found.

Table S3. Conserved domains of PvTALE proteins.

PvTALE subfamily	Gene name	Chromosome	Conserved domain
BEL1-like	PvTALE-01	1	Homeobox_KN, POX
BEL1-like	PvTALE-02	1	Homeobox_KN, POX
BEL1-like	PvTALE-04	1	Homeobox_KN, POX
BEL1-like	PvTALE-05	1	Homeobox_KN, POX
BEL1-like	PvTALE-07	2	Homeobox_KN, POX
BEL1-like	PvTALE-10	2	Homeobox_KN, POX
BEL1-like	PvTALE-11	2	Homeobox_KN, POX
BEL1-like	PvTALE-17	3	Homeobox_KN, POX
BEL1-like	PvTALE-19	5	Homeobox_KN, POX
BEL1-like	PvTALE-20	5	Homeobox_KN, POX
BEL1-like	PvTALE-23	7	Homeobox_KN, POX
BEL1-like	PvTALE-24	8	Homeobox_KN, POX
BEL1-like	PvTALE-26	9	Homeobox_KN, POX
BEL1-like	PvTALE-27	9	Homeobox_KN, POX
BEL1-like	PvTALE-28	9	Homeobox_KN, POX
BEL1-like	PvTALE-33	10	Homeobox_KN, POX
BEL1-like	PvTALE-34	11	Homeobox_KN, POX
BEL1-like	PvTALE-35	11	Homeobox_KN, POX
BEL1-like	PvTALE-29	9	Homeobox_KN, POX
KNOX-Class II	PvTALE-03	1	ELK, Homeobox_KN, KNOX1, KNOX2

Table S3. (Continued.)

KNOX-Class II	PvTALE-06	1	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class II	PvTALE-08	2	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class I	PvTALE-09	2	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class II	PvTALE-13	3	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class II	PvTALE-14	3	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class I	PvTALE-15	3	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class I	PvTALE-16	3	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class II	PvTALE-21	6	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class II	PvTALE-22	7	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class II	PvTALE-25	8	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class I	PvTALE-30	9	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class II	PvTALE-31	9	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class I	PvTALE-32	9	ELK, Homeobox_KN, KNOX1, KNOX2
KNOX-Class III	PvTALE-12	3	KNOX1, KNOX2
KNOX-Class III	PvTALE-18	4	KNOX1, KNOX2

Table S4. Gene duplication events between *PvTALE* genes.

Gene name	**Gene name**	**Ka**	**Ks**	**Ka/Ks**	**Type**
PvTALE-01	*PvTALE-26*	0.381080887	1.84826094	0.206183487	Segmental
PvTALE-01	*PvTALE-27*	0.13835999	0.83967835	0.164777369	Segmental
PvTALE-04	*PvTALE-29*	0.192015142	0.61731138	0.311050707	Segmental
PvTALE-05	*PvTALE-19*	0.30429328	2.44652393	0.124377807	Segmental
PvTALE-05	*PvTALE-23*	0.141976938	0.83659835	0.169707408	Segmental
PvTALE-05	*PvTALE-35*	0.339263556	2.0917369	0.16219227	Segmental
PvTALE-06	*PvTALE-22*	0.185816579	0.62313188	0.298197837	Segmental
PvTALE-07	*PvTALE-17*	0.143262912	0.90162448	0.158894213	Segmental
PvTALE-07	*PvTALE-27*	0.571400466	NaN	NaN	Segmental
PvTALE-08	*PvTALE-13*	0.132163153	0.92913122	0.142243797	Segmental
PvTALE-09	*PvTALE-16*	0.058228208	0.75024494	0.077612264	Segmental
PvTALE-10	*PvTALE-11*	0.120388814	0.7560827	0.159227045	Tandem
PvTALE-13	*PvTALE-31*	0.350863202	3.08604279	0.113693563	Segmental
PvTALE-14	*PvTALE-21*	0.124085722	0.67306417	0.184359425	Segmental
PvTALE-15	*PvTALE-30*	0.187200532	1.85491457	0.100921376	Segmental
PvTALE-15	*PvTALE-32*	0.089302801	0.89610783	0.099656312	Segmental
PvTALE-19	*PvTALE-23*	0.325440659	2.54219388	0.128015673	Segmental
PvTALE-19	*PvTALE-35*	0.18726472	0.98362348	0.190382524	Segmental
PvTALE-20	*PvTALE-34*	0.222012389	0.83220691	0.266775471	Segmental
PvTALE-23	*PvTALE-35*	0.335112756	1.85923772	0.180242016	Segmental
PvTALE-24	*PvTALE-33*	0.173301073	0.77078995	0.224835667	Segmental

NaN = no result.

Table S5. Orthologous gene pairs of *Phaseolus vulgaris* with *Arabidopsis thaliana (AtTALE)*, *Vigna unguiculata (VuTALE)*, *Solanum lycopersicum(SlTALE)*, *Glycine max(GmTALE)* and *Oryza sativa(OsTALE).*

Gene name	***AtTALE* gene ID**	**Ka**	**Ks**	**Ka/Ks**
PvTALE-09	*AT1G62990.1*	0.102661018	NaN	NaN
PvTALE-08	*AT1G23380.2*	0.201003497	NaN	NaN
PvTALE-08	*AT1G62360.1*	0.455370205	NaN	NaN
PvTALE-09	*AT1G70510.1*	0.767573023	NaN	NaN
PvTALE-07	*AT2G23760.1*	0.298070949	NaN	NaN
PvTALE-07	*AT4G36870.1*	0.310943506	4.29494786431731	0.072397504
PvTALE-11	*AT5G41410.1*	0.379077182	2.81626543584072	0.134602789
PvTALE-10	*AT5G41410.1*	0.421594053	NaN	NaN
PvTALE-05	*AT2G35940.1*	0.299416964	4.02156400831134	0.074452865
PvTALE-04	*AT4G32980.1*	0.411302204	1.70859359656791	0.240725592
PvTALE-16	*AT1G62990.1*	0.149652879	NaN	NaN
PvTALE-21	*AT1G62360.1*	0.230891219	2.50749427577259	0.092080457
PvTALE-20	*AT2G27990.1*	0.595211745	NaN	NaN
PvTALE-24	*AT5G02030.1*	0.351716296	3.95547047628264	0.088918954
PvTALE-23	*AT2G35940.1*	0.291119745	NaN	NaN
PvTALE-31	*AT1G23380.2*	0.350291937	1.88916877373378	0.185421198
PvTALE-31	*AT1G70510.1*	0.384762514	2.41711399421692	0.15918261
PvTALE-26	*AT1G75410.1*	0.358909987	2.92885024247058	0.122542963
PvTALE-26	*AT2G16400.1*	0.327615191	NaN	NaN
PvTALE-30	*AT4G32040.1*	0.226940379	NaN	NaN
PvTALE-26	*AT4G34610.1*	0.313866818	NaN	NaN
PvTALE-30	*AT5G25220.1*	0.121362972	5.77894685614624	0.02100088
PvTALE-30	*AT5G11060.1*	0.108355609	1.81671504444188	0.059643701
Gene name	***VuTALE* gene ID**	**Ka**	**Ks**	**Ka/Ks**
PvTALE-33	*Vigun05g112800.1*	0.181285	0.749591872	0.241845
PvTALE-33	*Vigun10g059300.1*	0.022841	0.15848182	0.144122
PvTALE-09	*Vigun02g172100.1*	0.007561	0.204032384	0.037056
PvTALE-10	*Vigun02g177500.1*	0.025948	0.154691149	0.167742
PvTALE-11	*Vigun02g177500.1*	0.1276	0.814718853	0.156619
PvTALE-07	*Vigun02g137100.1*	0.023353	0.126920222	0.183997
PvTALE-08	*Vigun02g079500.1*	0.008325	0.165933446	0.050169
PvTALE-09	*Vigun03g310400.1*	0.080372	0.794479439	0.101163
PvTALE-08	*Vigun03g234200.1*	0.127213	0.96242085	0.13218
PvTALE-11	*Vigun03g042200.1*	0.02672	0.161401173	0.16555
PvTALE-07	*Vigun03g270000.1*	0.140669	0.90306899	0.155768
PvTALE-10	*Vigun03g042200.1*	0.137972	0.791226749	0.174377
PvTALE-05	*Vigun01g180200.1*	0.016417	0.196023573	0.08375
PvTALE-06	*Vigun01g209400.1*	0.047343	0.220957136	0.214264
PvTALE-05	*Vigun05g231800.1*	0.28731	2.618316407	0.109731
PvTALE-06	*Vigun07g139600.1*	0.176768	0.760531761	0.232427
PvTALE-05	*Vigun07g014500.1*	0.14292	0.897992703	0.159155
PvTALE-03	*Vigun08g034600.1*	0.027947	0.169669705	0.164712
PvTALE-04	*Vigun08g036800.1*	0.030058	0.109074365	0.275573
PvTALE-04	*Vigun09g225900.1*	0.19392	0.647990968	0.299263
PvTALE-05	*Vigun11g121700.1*	0.332605	2.151828278	0.154568
PvTALE-18	*Vigun04g034500.1*	0.105383	0.139939467	0.753059
PvTALE-12	*Vigun02g017500.1*	0.102756	0.248323577	0.413798

Table S5. (Continued.)

PvTALE-16	*Vigun02g172100.1*	0.055939	0.89724021	0.062345
PvTALE-15	*Vigun03g366200.1*	0.00738	0.233834718	0.031561
PvTALE-16	*Vigun03g310400.1*	0.019846	0.231253041	0.08582
PvTALE-15	*Vigun09g054100.1*	0.087595	0.93471784	0.093713
PvTALE-15	*Vigun09g212500.1*	0.188552	2.177159270	0.086605
PvTALE-21	*Vigun03g443600.1*	0.152635	0.667994177	0.228497
PvTALE-20	*Vigun05g246400.1*	0.033072	0.173861496	0.190223
PvTALE-20	*Vigun11g139500.1*	0.225721	0.871024693	0.259145
PvTALE-24	*Vigun05g112800.1*	0.027558	0.10142009	0.271718
PvTALE-25	*Vigun08g174300.1*	0.008444	0.104708949	0.080641
PvTALE-22	*Vigun01g209400.1*	0.186595	0.734191461	0.25415
PvTALE-23	*Vigun01g180200.1*	0.142215	0.906307659	0.156917
PvTALE-23	*Vigun05g231800.1*	0.336943	2.2759461772	0.148045
PvTALE-23	*Vigun07g014500.1*	0.025915	0.190010045	0.136389
PvTALE-22	*Vigun07g139600.1*	0.038201	0.162113161	0.235647
PvTALE-23	*Vigun11g121700.1*	0.339274	1.8493882852	0.183452
PvTALE-31	*Vigun02g079500.1*	0.349067	1.6666065105	0.209448
PvTALE-31	*Vigun03g234200.1*	0.349975	2.45074949177	0.142803
PvTALE-30	*Vigun03g366200.1*	0.188914	2.166366654	0.087203
PvTALE-32	*Vigun03g366200.1*	0.085658	1.0130778823	0.084552
PvTALE-26	*Vigun08g011700.1*	0.391874	1.823666919	0.214883
PvTALE-31	*Vigun09g088300.1*	0.060939	0.111972	0.544236
PvTALE-32	*Vigun09g054100.1*	0.014907	0.20542	0.072566
PvTALE-30	*Vigun09g212500.1*	0.02855	0.233976	0.12202
PvTALE-26	*Vigun09g268200.1*	0.043229	0.137349	0.314741
Gene name	***SlTALE* gene ID**	**Ka**	**Ks**	**Ka/Ks**
PvTALE-33	*Solyc09g011380.3*	0.34493787	NaN	NaN
PvTALE-07	*Solyc02g065490.3*	0.278852429	1.767145134	0.157798
PvTALE-07	*Solyc02g089940.3*	0.172168528	3.116924238148960	0.055237
PvTALE-07	*Solyc04g079830.2*	0.355166718	2.5593793120433200	0.138771
PvTALE-08	*Solyc05g005090.3*	0.276239105	NaN	NaN
PvTALE-11	*Solyc08g081385.1*	0.144028701	2.061220561334100	0.069875
PvTALE-09	*Solyc08g080120.3*	0.101414428	3.2038724113854600	0.031654
PvTALE-06	*Solyc01g100510.3*	0.300225078	NaN	NaN
PvTALE-05	*Solyc01g007070.3*	0.266188344	2.475059297159460	0.107548
PvTALE-03	*Solyc04g077210.3*	0.160789391	2.1409175644423700	0.075103
PvTALE-05	*Solyc06g074120.3*	0.389344356	NaN	NaN
PvTALE-04	*Solyc08g065420.3*	0.461172919	1.655114458462790	0.278635
PvTALE-18	*Solyc06g072480.1*	0.404856074	2.824064648427190	0.143359
PvTALE-16	*Solyc08g080120.3*	0.152738586	NaN	NaN
PvTALE-15	*Solyc12g010410.2*	0.254905882	3.0146682199448100	0.084555
PvTALE-21	*Solyc02g081120.3*	0.177487134	1.6566394614984900	0.107137
PvTALE-24	*Solyc09g011380.3*	0.335463417	1.5513756040971200	0.216236
PvTALE-24	*Solyc10g086640.2*	0.38565558	1.920450079715140	0.200815
PvTALE-22	*Solyc01g100510.3*	0.299314896	3.101640357304650	0.096502
PvTALE-23	*Solyc01g007070.3*	0.299722031	2.397656688441450	0.125006
PvTALE-23	*Solyc06g074120.3*	0.397358277	NaN	NaN
PvTALE-23	*Solyc11g068950.2*	0.39691663	5.130730683970450	0.077361
PvTALE-26	*Solyc01g109980.3*	0.311696891	1.4693797072002800	0.212128

Table S5. (Continued.)

PvTALE-26	*Solyc04g080790.3*	0.386576939	2.051218240516580	0.188462
PvTALE-28	*Solyc04g080780.2*	0.321452115	2.9513860054283900	0.108916
PvTALE-31	*Solyc05g005090.3*	0.407424838	2.3353265718847100	0.174462
PvTALE-32	*Solyc12g010410.2*	0.249913475	NaN	NaN
Gene name	***GmTALE* gene ID**	**Ka**	**Ks**	**Ka/Ks**
PvTALE-33	*Glyma.01G104200.1*	0.036359121	0.256739558	0.14161869
PvTALE-33	*Glyma.03G070900.1*	0.040501555	0.285198526	0.14201179
PvTALE-09	*Glyma.01G214800.1*	0.016954228	0.314541398	0.05390142
PvTALE-08	*Glyma.01G028300.1*	0.035372849	0.330992558	0.10686902
PvTALE-07	*Glyma.02G060000.1*	0.13363698	0.979924256	0.13637481
PvTALE-08	*Glyma.02G036900.1*	0.044253134	0.340244599	0.13006271
PvTALE-11	*Glyma.05G210300.1*	0.05714669	0.336092154	0.1700328
PvTALE-10	*Glyma.05G210300.1*	0.141302671	0.708528135	0.19943128
PvTALE-09	*Glyma.05G050600.1*	0.040164036	0.654107894	0.06140277
PvTALE-11	*Glyma.08G016900.1*	0.060353554	0.305387556	0.19762938
PvTALE-10	*Glyma.08G016900.1*	0.132715393	0.674493898	0.19676293
PvTALE-08	*Glyma.08G284300.1*	0.114712518	0.718237219	0.15971397
PvTALE-07	*Glyma.11G062300.1*	0.048516885	0.2440372	0.19880938
PvTALE-10	*Glyma.11G022200.1*	0.042557187	0.287117519	0.14822219
PvTALE-09	*Glyma.11G027100.1*	0.017059407	0.32710361	0.05215292
PvTALE-11	*Glyma.11G022200.1*	0.130364774	0.841147469	0.15498445
PvTALE-07	*Glyma.16G142800.1*	0.132593481	0.930912933	0.14243382
PvTALE-09	*Glyma.17G132600.1*	0.043083245	0.621945756	0.06927171
PvTALE-08	*Glyma.18G141200.1*	0.121821174	0.751843751	0.16202991
PvTALE-05	*Glyma.03G202300.1*	0.036468739	0.321397337	0.11346933
PvTALE-06	*Glyma.03G231100.1*	0.082374942	0.2985841	0.27588523
PvTALE-03	*Glyma.04G049500.1*	0.123728786	0.710042549	0.17425545
PvTALE-04	*Glyma.04G050800.1*	0.182793194	0.55090703	0.33180407
PvTALE-04	*Glyma.06G051600.1*	0.173940904	0.60131688	0.28926662
PvTALE-06	*Glyma.10G147000.1*	0.168490645	0.541070025	0.31140266
PvTALE-05	*Glyma.13G312900.1*	0.304357854	2.5760802	0.11814766
PvTALE-03	*Glyma.14G091200.1*	0.022354267	0.27821881	0.08034779
PvTALE-05	*Glyma.19G199500.1*	0.036931543	0.294374869	0.12545753
PvTALE-18	*Glyma.07G104800.1*	0.064461322	0.20077468	0.321063
PvTALE-18	*Glyma.09G172800.1*	0.060527789	0.216633141	0.27940226
PvTALE-16	*Glyma.01G214800.1*	0.058235221	0.749092253	0.07774105
PvTALE-12	*Glyma.02G086400.1*	0.136741168	0.393608899	0.34740365
PvTALE-16	*Glyma.05G050600.1*	0.041215341	0.284093308	0.14507677
PvTALE-12	*Glyma.07G172500.1*	0.124088889	0.378392311	0.32793713
PvTALE-16	*Glyma.11G027100.1*	0.054416607	0.773672289	0.07033547
PvTALE-15	*Glyma.13G157300.1*	0.025890737	0.356016054	0.07272351
PvTALE-15	*Glyma.15G202200.1*	0.080826607	0.977934754	0.08265031
PvTALE-16	*Glyma.17G132600.1*	0.037539534	0.238951517	0.15710105
PvTALE-15	*Glyma.17G104800.1*	0.020942307	0.322004957	0.06503722
PvTALE-21	*Glyma.07G263600.1*	0.128275932	0.637946604	0.20107628
PvTALE-21	*Glyma.09G007500.1*	0.033643882	0.245374886	0.13711217
PvTALE-21	*Glyma.15G111900.1*	0.039705133	0.245374886	0.16181417
PvTALE-21	*Glyma.17G010200.1*	0.086371319	0.602132817	0.1434423
PvTALE-20	*Glyma.12G077800.1*	0.239900342	0.881807872	0.27205512

Table S5. (Continued.)

PvTALE-24	*Glyma.01G104200.1*	0.177208978	0.821593284	0.21568942
PvTALE-25	*Glyma.02G269900.1*	0.126140494	0.446960697	0.28221831
PvTALE-24	*Glyma.03G070900.1*	0.173911949	0.778815722	0.22330308
PvTALE-25	*Glyma.14G047000.1*	0.018412349	0.267272818	0.06888972
PvTALE-22	*Glyma.03G231100.1*	0.19272208	0.621882827	0.30990095
PvTALE-23	*Glyma.03G202300.1*	0.150621195	0.785503542	0.19175113
PvTALE-22	*Glyma.10G147000.1*	0.062772825	0.31600129	0.19864737
PvTALE-23	*Glyma.19G199500.1*	0.149573576	0.742044972	0.20156942
PvTALE-31	*Glyma.02G036900.1*	0.352984271	1.86439257	0.18932937
PvTALE-30	*Glyma.04G064100.1*	0.04675505	0.290703591	0.1608341
PvTALE-28	*Glyma.04G029200.1*	0.048107608	0.274537801	0.17523127
PvTALE-31	*Glyma.04G185100.1*	0.117248567	0.271312382	0.43215339
PvTALE-30	*Glyma.06G065200.1*	0.049404017	0.278413767	0.17744818
PvTALE-26	*Glyma.06G009500.1*	0.07910104	0.236608222	0.33431231
PvTALE-28	*Glyma.06G029200.1*	0.052345251	0.273888949	0.19111852
PvTALE-32	*Glyma.09G095700.1*	0.025828378	0.431697345	0.05982983
PvTALE-32	*Glyma.13G157300.1*	0.084375058	0.832386271	0.10136527
PvTALE-32	*Glyma.14G112400.1*	0.10642562	0.655329448	0.16240018
PvTALE-32	*Glyma.15G202200.1*	0.022729012	0.417893658	0.05438946
PvTALE-32	*Glyma.17G104800.1*	0.077932642	0.843337194	0.09240982
PvTALE-30	*Glyma.17G215500.1*	0.122488769	0.72810295	0.16823001
Gene name	***OsTALE* gene ID**	**Ka**	**Ks**	**Ka/Ks**
PvTALE-33	*LOC_Os01g62920.1*	0.423272594	NaN	NaN
PvTALE-33	*LOC_Os05g38120.1*	0.479272538	NaN	NaN
PvTALE-06	*LOC_Os01g19694.1*	0.295878737	NaN	NaN
PvTALE-05	*LOC_Os11g06020.1*	0.440734576	NaN	NaN
PvTALE-03	*LOC_Os03g51690.2*	0.292580866	NaN	NaN
PvTALE-15	*LOC_Os02g08544.1*	0.259651356	3.54162645013789	0.073314157
PvTALE-15	*LOC_Os06g43860.1*	0.237184146	2.55823904102121	0.092713833
PvTALE-15	*LOC_Os08g19650.1*	0.143387314	3.53015119905106	0.040617896
PvTALE-24	*LOC_Os01g62920.1*	0.53332406	NaN	NaN
PvTALE-24	*LOC_Os05g38120.1*	0.48631332	NaN	NaN
PvTALE-22	*LOC_Os01g19694.1*	0.323705565	NaN	NaN
PvTALE-22	*LOC_Os05g03884.1*	0.345422023	NaN	NaN
PvTALE-30	*LOC_Os02g08544.1*	0.199698917	NaN	NaN
PvTALE-32	*LOC_Os06g43860.1*	0.264394693	2.87013713301765	0.092119185

NaN = no result.

Table S6. The features of cis-acting elements in the promoters of *PvTALE* genes.

Gene name	**Start position**	**End position**	**Predicted cis-element**
PvTALE-01	1394	1403	G-box
PvTALE-01	653	658	ABRE
PvTALE-01	443	450	MRE
PvTALE-01	1481	1492	Unnamed__1
PvTALE-01	25	34	GATA-motif
PvTALE-01	461	467	ARE
PvTALE-01	558	564	Box 4
PvTALE-01	713	719	Box 4

Table S6. (Continued.)

PvTALE-01	878	884	Box 4
PvTALE-01	1028	1034	Box 4
PvTALE-01	1488	1494	Box 4
PvTALE-01	652	658	G-Box
PvTALE-01	1108	1117	ATCT-motif
PvTALE-02	366	374	AE-box
PvTALE-02	224	230	G-box
PvTALE-02	725	731	G-box
PvTALE-02	1431	1437	G-box
PvTALE-02	725	731	G-Box
PvTALE-02	1375	1382	GARE-motif
PvTALE-02	1228	1234	TCT-motif
PvTALE-02	1442	1448	TCT-motif
PvTALE-02	171	178	MRE
PvTALE-02	1391	1402	Unnamed__1
PvTALE-02	1224	1233	TC-rich repeats
PvTALE-02	1442	1451	TC-rich repeats
PvTALE-02	729	738	Box II
PvTALE-02	124	130	Box 4
PvTALE-02	182	188	Box 4
PvTALE-02	203	209	Box 4
PvTALE-02	865	871	Box 4
PvTALE-02	1398	1404	Box 4
PvTALE-02	224	229	ABRE
PvTALE-02	725	731	ABRE
PvTALE-02	726	731	ABRE
PvTALE-02	1432	1437	ABRE
PvTALE-02	1340	1346	TGA-element
PvTALE-02	506	512	MBS
PvTALE-02	208	218	3-AF1 binding site
PvTALE-02	1128	1137	AT-rich sequence
PvTALE-02	779	788	I-box
PvTALE-02	150	156	ARE
PvTALE-02	967	973	ARE
PvTALE-02	1368	1374	ARE
PvTALE-02	1487	1493	ARE
PvTALE-03	268	274	MBS
PvTALE-03	196	201	ABRE
PvTALE-03	426	432	CAT-box
PvTALE-03	794	805	Unnamed__1
PvTALE-03	952	959	GT1-motif
PvTALE-03	953	959	GT1-motif
PvTALE-03	1445	1451	GT1-motif
PvTALE-03	168	174	Box 4
PvTALE-03	637	643	Box 4
PvTALE-03	781	787	Box 4
PvTALE-03	793	799	Box 4
PvTALE-03	948	954	Box 4
PvTALE-03	620	633	AT1-motif

Table S6. (Continued.)

PvTALE-03	757	770	AT1-motif
PvTALE-03	187	193	ARE
PvTALE-03	1122	1128	ARE
PvTALE-03	1329	1335	ARE
PvTALE-03	542	549	MRE
PvTALE-03	955	962	MRE
PvTALE-03	195	201	G-Box
PvTALE-03	549	555	LTR
PvTALE-03	431	438	P-box
PvTALE-03	281	287	TGA-element
PvTALE-03	648	654	TGA-element
PvTALE-04	793	799	TCT-motif
PvTALE-04	1327	1333	G-Box
PvTALE-04	1236	1241	ABRE
PvTALE-04	1327	1332	ABRE
PvTALE-04	1310	1317	GCN4_motif
PvTALE-04	1197	1204	GATA-motif
PvTALE-04	1358	1364	ARE
PvTALE-04	71	78	MRE
PvTALE-04	808	821	AT1-motif
PvTALE-04	141	149	AE-box
PvTALE-04	1110	1115	TGACG-motif
PvTALE-04	1238	1243	TGACG-motif
PvTALE-04	1110	1115	CGTCA-motif
PvTALE-04	1238	1243	CGTCA-motif
PvTALE-04	193	199	Box 4
PvTALE-04	237	243	Box 4
PvTALE-04	271	277	Box 4
PvTALE-04	364	370	Box 4
PvTALE-04	374	380	Box 4
PvTALE-04	510	516	Box 4
PvTALE-04	594	600	Box 4
PvTALE-04	604	610	Box 4
PvTALE-04	651	657	Box 4
PvTALE-04	655	661	Box 4
PvTALE-04	741	747	Box 4
PvTALE-04	1236	1242	G-box
PvTALE-04	652	663	Unnamed__1
PvTALE-05	158	164	ARE
PvTALE-05	428	434	ARE
PvTALE-05	1395	1401	TGA-element
PvTALE-05	607	613	CCAAT-box
PvTALE-05	353	361	AE-box
PvTALE-05	382	388	Box 4
PvTALE-05	875	881	Box 4
PvTALE-05	1161	1167	Box 4
PvTALE-05	542	548	TCT-motif
PvTALE-05	1084	1095	MBSI
PvTALE-05	261	266	CGTCA-motif

Table S6. (Continued.)

PvTALE-05	261	266	TGACG-motif
PvTALE-05	847	854	TATC-box
PvTALE-05	57	64	AuxRR-core
PvTALE-05	1202	1211	HD-Zip 3
PvTALE-05	924	931	GARE-motif
PvTALE-05	42	53	Y-box
PvTALE-05	242	251	TCA-element
PvTALE-05	1375	1384	TCA-element
PvTALE-05	247	256	O2-site
PvTALE-05	777	785	O2-site
PvTALE-05	35	41	CAT-box
PvTALE-05	226	232	MBS
PvTALE-05	1223	1229	MBS
PvTALE-05	75	82	TCCC-motif
PvTALE-06	173	184	Unnamed__1
PvTALE-06	486	496	GATA-motif
PvTALE-06	425	431	Box 4
PvTALE-06	1091	1097	Box 4
PvTALE-06	79	92	AT1-motif
PvTALE-06	323	329	TCT-motif
PvTALE-06	1463	1469	G-box
PvTALE-07	1123	1132	I-box
PvTALE-07	1125	1133	I-box
PvTALE-07	888	894	G-Box
PvTALE-07	1232	1237	CGTCA-motif
PvTALE-07	951	957	TCT-motif
PvTALE-07	40	46	CCAAT-box
PvTALE-07	403	409	CCAAT-box
PvTALE-07	164	173	ATC-motif
PvTALE-07	527	536	ATC-motif
PvTALE-07	945	955	3-AF1 binding site
PvTALE-07	1360	1370	3-AF1 binding site
PvTALE-07	888	894	G-box
PvTALE-07	1080	1090	TCA-element
PvTALE-07	856	863	MRE
PvTALE-07	1232	1237	TGACG-motif
PvTALE-07	1284	1292	AE-box
PvTALE-07	1125	1134	GATA-motif
PvTALE-07	1429	1436	GATA-motif
PvTALE-07	886	895	ABRE
PvTALE-07	888	894	ABRE
PvTALE-07	889	894	ABRE
PvTALE-07	121	127	Box 4
PvTALE-07	204	210	Box 4
PvTALE-07	484	490	Box 4
PvTALE-07	567	573	Box 4
PvTALE-07	969	975	Box 4
PvTALE-07	1278	1284	Box 4
PvTALE-07	845	852	TATC-box

Table S6. (Continued.)

PvTALE-07	193	206	AT1-motif
PvTALE-07	556	569	AT1-motif
PvTALE-08	1464	1473	O2-site
PvTALE-08	837	845	chs-CMA1a
PvTALE-08	949	958	TC-rich repeats
PvTALE-08	73	79	TCT-motif
PvTALE-08	503	509	TCT-motif
PvTALE-08	568	574	TCT-motif
PvTALE-08	949	955	TCT-motif
PvTALE-08	936	944	GA-motif
PvTALE-08	1171	1177	ARE
PvTALE-08	260	267	MRE
PvTALE-08	805	811	CAT-box
PvTALE-08	1098	1105	P-box
PvTALE-08	45	51	Box 4
PvTALE-08	1456	1463	GATA-motif
PvTALE-09	338	347	ACE
PvTALE-09	220	225	ABRE
PvTALE-09	515	523	RY-element
PvTALE-09	97	103	Box 4
PvTALE-09	309	315	Box 4
PvTALE-09	464	470	Box 4
PvTALE-09	605	611	Box 4
PvTALE-09	872	878	Box 4
PvTALE-09	882	888	Box 4
PvTALE-09	890	896	Box 4
PvTALE-09	224	230	GT1-motif
PvTALE-09	219	225	G-Box
PvTALE-09	485	494	HD-Zip 3
PvTALE-09	1533	1542	HD-Zip 3
PvTALE-09	1381	1387	ARE
PvTALE-09	1489	1499	I-box
PvTALE-10	187	195	GA-motif
PvTALE-10	111	117	Box 4
PvTALE-10	123	129	Box 4
PvTALE-10	278	284	Box 4
PvTALE-10	902	911	GATA-motif
PvTALE-10	844	850	GT1-motif
PvTALE-10	851	858	TATC-box
PvTALE-10	853	862	I-box
PvTALE-10	372	381	TC-rich repeats
PvTALE-10	353	361	AE-box
PvTALE-10	1016	1022	ARE
PvTALE-10	1267	1273	ARE
PvTALE-10	1021	1028	MRE
PvTALE-10	358	364	TCT-motif
PvTALE-10	955	961	TCT-motif
PvTALE-10	339	346	GCN4_motif
PvTALE-10	1267	1278	AAAC-motif

Table S6. (Continued.)

PvTALE-11	1215	1222	GT1-motif
PvTALE-11	1202	1207	ABRE
PvTALE-11	90	96	Box 4
PvTALE-11	965	971	Box 4
PvTALE-11	1293	1299	Box 4
PvTALE-11	1375	1381	Box 4
PvTALE-11	112	120	chs-CMA1a
PvTALE-11	723	732	GATA-motif
PvTALE-11	1166	1172	TCT-motif
PvTALE-11	1192	1199	AuxRR-core
PvTALE-11	1201	1207	G-box
PvTALE-11	1485	1494	WUN-motif
PvTALE-11	1488	1497	WUN-motif
PvTALE-11	713	722	circadian
PvTALE-11	442	451	TCA-element
PvTALE-12	549	562	AT1-motif
PvTALE-12	34	39	ABRE
PvTALE-12	34	40	G-Box
PvTALE-12	89	99	GATA-motif
PvTALE-12	1229	1237	RY-element
PvTALE-12	1233	1241	RY-element
PvTALE-12	64	70	Box 4
PvTALE-12	106	112	Box 4
PvTALE-12	699	707	O2-site
PvTALE-12	949	958	O2-site
PvTALE-12	1209	1215	GT1-motif
PvTALE-12	1275	1281	GT1-motif
PvTALE-12	1005	1014	TCA-element
PvTALE-12	575	581	TCT-motif
PvTALE-12	1045	1051	TCT-motif
PvTALE-12	892	899	GCN4_motif
PvTALE-12	639	649	AT-rich element
PvTALE-12	1067	1078	Box III
PvTALE-12	1045	1054	TC-rich repeats
PvTALE-12	1246	1255	TC-rich repeats
PvTALE-12	417	424	GARE-motif
PvTALE-12	1290	1297	GARE-motif
PvTALE-13	606	606	Box 4
PvTALE-13	641	641	Box 4
PvTALE-13	655	655	Box 4
PvTALE-13	659	659	Box 4
PvTALE-13	957	957	Box 4
PvTALE-13	1182	1182	Box 4
PvTALE-13	783	783	CGTCA-motif
PvTALE-13	1318	1318	MRE
PvTALE-13	838	838	AT-rich element
PvTALE-13	783	783	TGACG-motif
PvTALE-13	873	873	GATA-motif
PvTALE-13	873	873	I-box

Table S6. (Continued.)

PvTALE-13	875	875	I-box
PvTALE-13	1283	1283	I-box
PvTALE-13	1311	1311	GT1-motif
PvTALE-13	366	366	AE-box
PvTALE-13	1288	1288	TATC-box
PvTALE-13	1080	1080	LAMP-element
PvTALE-13	429	429	AT1-motif
PvTALE-14	116	122	ARE
PvTALE-14	168	174	ARE
PvTALE-14	1275	1281	ARE
PvTALE-14	1172	1180	chs-CMA1a
PvTALE-14	1109	1118	O2-site
PvTALE-14	1473	1482	O2-site
PvTALE-14	1453	1462	I-box
PvTALE-14	146	153	TATC-box
PvTALE-14	939	948	TCA-element
PvTALE-14	335	345	GATA-motif
PvTALE-14	1451	1460	GATA-motif
PvTALE-14	739	745	Box 4
PvTALE-14	1407	1413	Box 4
PvTALE-14	1493	1499	Box 4
PvTALE-14	471	477	CAT-box
PvTALE-15	1178	1186	LAMP-element
PvTALE-15	1062	1068	ARE
PvTALE-15	175	184	TCA-element
PvTALE-15	1469	1478	TCA-element
PvTALE-15	927	934	GARE-motif
PvTALE-15	1083	1089	MBS
PvTALE-15	1359	1365	Box 4
PvTALE-15	4	9	TGACG-motif
PvTALE-15	464	469	TGACG-motif
PvTALE-15	445	452	AuxRR-core
PvTALE-15	4	9	CGTCA-motif
PvTALE-15	464	469	CGTCA-motif
PvTALE-15	318	324	CAT-box
PvTALE-15	509	515	CAT-box
PvTALE-15	573	579	CAT-box
PvTALE-15	2	11	O2-site
PvTALE-15	297	306	O2-site
PvTALE-15	789	796	P-box
PvTALE-15	357	363	GT1-motif
PvTALE-15	1256	1262	GT1-motif
PvTALE-15	5	11	G-box
PvTALE-15	476	482	G-box
PvTALE-15	292	301	HD-Zip 3
PvTALE-15	6	11	ABRE
PvTALE-16	1464	1470	TCT-motif
PvTALE-16	856	867	Unnamed__1
PvTALE-16	1065	1076	Unnamed__1

Table S6. (Continued.)

PvTALE-16	1334	1343	TC-rich repeats
PvTALE-16	1460	1469	TC-rich repeats
PvTALE-16	47	55	LAMP-element
PvTALE-16	1242	1250	chs-CMA1a
PvTALE-16	213	218	ABRE
PvTALE-16	1324	1329	ABRE
PvTALE-16	2	8	Box 4
PvTALE-16	843	849	Box 4
PvTALE-16	855	861	Box 4
PvTALE-16	892	898	Box 4
PvTALE-16	988	994	Box 4
PvTALE-16	1009	1015	Box 4
PvTALE-16	1072	1078	Box 4
PvTALE-16	1084	1090	Box 4
PvTALE-16	1361	1367	Box 4
PvTALE-16	1143	1150	GARE-motif
PvTALE-16	1457	1464	GARE-motif
PvTALE-16	572	578	GT1-motif
PvTALE-16	975	981	GT1-motif
PvTALE-16	213	219	G-box
PvTALE-16	1324	1330	G-box
PvTALE-16	548	556	GA-motif
PvTALE-16	1355	1363	GA-motif
PvTALE-17	1279	1289	MBSI
PvTALE-17	845	851	Box 4
PvTALE-17	764	770	G-box
PvTALE-17	1079	1085	G-box
PvTALE-17	764	770	ABRE
PvTALE-17	765	770	ABRE
PvTALE-17	1002	1011	GATA-motif
PvTALE-17	135	143	GA-motif
PvTALE-17	440	446	TCT-motif
PvTALE-17	587	593	TCT-motif
PvTALE-17	827	833	TCT-motif
PvTALE-17	893	899	TCT-motif
PvTALE-17	163	172	TCA-element
PvTALE-17	958	968	TCA-element
PvTALE-17	1296	1305	TCA-element
PvTALE-17	732	739	MRE
PvTALE-17	355	362	GT1-motif
PvTALE-17	356	362	GT1-motif
PvTALE-17	447	453	GT1-motif
PvTALE-17	1365	1371	GT1-motif
PvTALE-17	764	770	G-Box
PvTALE-17	1000	1009	I-box
PvTALE-17	1002	1010	I-box
PvTALE-17	1383	1393	I-box
PvTALE-17	1033	1039	ARE
PvTALE-17	1115	1121	ARE

Table S6. (Continued.)

PvTALE-17	1438	1447	Box II
PvTALE-18	1404	1410	Box 4
PvTALE-18	954	960	LTR
PvTALE-18	430	439	ATCT-motif
PvTALE-18	1173	1179	G-box
PvTALE-18	107	116	TCA-element
PvTALE-18	408	414	ARE
PvTALE-18	805	811	ARE
PvTALE-18	1388	1394	CAT-box
PvTALE-18	1062	1071	circadian
PvTALE-18	1173	1178	ABRE
PvTALE-18	1249	1258	O2-site
PvTALE-18	1285	1294	O2-site
PvTALE-18	81	88	MRE
PvTALE-18	118	126	AE-box
PvTALE-18	1247	1257	I-box
PvTALE-18	1168	1174	GT1-motif
PvTALE-19	292	300	chs-CMA2a
PvTALE-19	711	720	TCA-element
PvTALE-19	1308	1317	TCA-element
PvTALE-19	1261	1267	Box 4
PvTALE-19	1397	1403	Box 4
PvTALE-19	1287	1295	GA-motif
PvTALE-19	131	137	ARE
PvTALE-19	1094	1101	MRE
PvTALE-19	685	691	MBS
PvTALE-19	530	536	GC-motif
PvTALE-19	73	78	ABRE
PvTALE-19	771	778	GATA-motif
PvTALE-19	1177	1188	GATA-motif
PvTALE-19	758	764	TCT-motif
PvTALE-19	72	78	G-Box
PvTALE-19	707	714	GCN4_motif
PvTALE-19	9	17	I-box
PvTALE-19	984	993	I-box
PvTALE-19	416	424	O2-site
PvTALE-20	1084	1095	Unnamed__1
PvTALE-20	1280	1289	TCA-element
PvTALE-20	627	636	TC-rich repeats
PvTALE-20	1192	1201	TC-rich repeats
PvTALE-20	505	513	AE-box
PvTALE-20	372	379	TCCC-motif
PvTALE-20	885	892	GT1-motif
PvTALE-20	886	892	GT1-motif
PvTALE-20	1196	1202	TCT-motif
PvTALE-20	307	314	GARE-motif
PvTALE-20	349	359	3-AF1 binding site
PvTALE-20	840	846	Box 4
PvTALE-20	1027	1033	Box 4

Table S6. (Continued.)

PvTALE-20	1091	1097	Box 4
PvTALE-20	9	15	ARE
PvTALE-20	255	261	ARE
PvTALE-20	312	318	ARE
PvTALE-20	1395	1401	ARE
PvTALE-21	59	68	Gap-box
PvTALE-21	113	120	MRE
PvTALE-21	162	169	MRE
PvTALE-21	508	514	G-box
PvTALE-21	508	513	ABRE
PvTALE-21	1270	1277	TATC-box
PvTALE-21	186	192	TCT-motif
PvTALE-21	707	713	CAT-box
PvTALE-21	510	515	TGACG-motif
PvTALE-21	336	343	P-box
PvTALE-21	288	297	TCA-element
PvTALE-21	907	916	TCA-element
PvTALE-21	1309	1318	TCA-element
PvTALE-21	510	515	CGTCA-motif
PvTALE-21	1027	1036	circadian
PvTALE-21	24	31	GT1-motif
PvTALE-21	25	31	GT1-motif
PvTALE-21	281	287	Box 4
PvTALE-21	611	617	Box 4
PvTALE-21	638	644	Box 4
PvTALE-22	469	475	TCT-motif
PvTALE-22	873	879	TCT-motif
PvTALE-22	934	940	TCT-motif
PvTALE-22	1418	1423	CGTCA-motif
PvTALE-22	1440	1445	CGTCA-motif
PvTALE-22	307	312	ABRE
PvTALE-22	1399	1408	ABRE
PvTALE-22	1401	1407	ABRE
PvTALE-22	1402	1407	ABRE
PvTALE-22	1416	1421	ABRE
PvTALE-22	992	998	Box 4
PvTALE-22	1418	1423	TGACG-motif
PvTALE-22	1440	1445	TGACG-motif
PvTALE-22	34	43	TC-rich repeats
PvTALE-22	645	654	TC-rich repeats
PvTALE-22	626	639	AT1-motif
PvTALE-22	1124	1133	GT1-motif
PvTALE-22	1097	1106	ATCT-motif
PvTALE-22	918	926	GA-motif
PvTALE-22	1401	1407	G-Box
PvTALE-22	154	161	P-box
PvTALE-22	306	312	G-box
PvTALE-22	1401	1407	G-box
PvTALE-22	1416	1422	G-box

Table S6. (Continued.)

PvTALE-23	296	305	I-box
PvTALE-23	650	658	RY-element
PvTALE-23	47	53	Box 4
PvTALE-23	493	499	Box 4
PvTALE-23	1105	1111	Box 4
PvTALE-23	1109	1115	Box 4
PvTALE-23	1113	1119	Box 4
PvTALE-23	1460	1466	Box 4
PvTALE-23	772	781	Gap-box
PvTALE-23	247	256	TC-rich repeats
PvTALE-23	450	456	TGA-element
PvTALE-23	586	592	TGA-element
PvTALE-23	984	990	TGA-element
PvTALE-23	546	552	MBS
PvTALE-23	1218	1225	TATC-box
PvTALE-23	877	883	ARE
PvTALE-24	953	959	G-Box
PvTALE-24	400	413	AT1-motif
PvTALE-24	953	959	ABRE
PvTALE-24	954	959	ABRE
PvTALE-24	624	630	Box 4
PvTALE-24	1183	1189	Box 4
PvTALE-24	1094	1105	Unnamed__1
PvTALE-24	959	964	TGACG-motif
PvTALE-24	959	964	CGTCA-motif
PvTALE-24	953	959	G-box
PvTALE-24	1126	1135	G-box
PvTALE-24	376	382	ARE
PvTALE-25	1325	1331	TCT-motif
PvTALE-25	1443	1448	ABRE
PvTALE-25	1125	1131	ARE
PvTALE-25	751	758	TCCC-motif
PvTALE-25	77	83	MBS
PvTALE-25	177	183	Box 4
PvTALE-25	202	208	Box 4
PvTALE-25	206	212	Box 4
PvTALE-25	210	216	Box 4
PvTALE-25	214	220	Box 4
PvTALE-25	218	224	Box 4
PvTALE-25	1296	1302	Box 4
PvTALE-25	1442	1448	G-box
PvTALE-25	666	672	A-box
PvTALE-25	1063	1069	A-box
PvTALE-25	765	774	ATCT-motif
PvTALE-26	810	815	TGACG-motif
PvTALE-26	189	195	ARE
PvTALE-26	263	269	ARE
PvTALE-26	510	516	GC-motif
PvTALE-26	810	815	CGTCA-motif

Table S6. (Continued.)

PvTALE-26	1435	1444	TCA-element
PvTALE-26	74	80	MBS
PvTALE-26	834	840	MBS
PvTALE-26	1010	1016	MBS
PvTALE-26	1263	1269	MBS
PvTALE-27	1089	1098	ABRE
PvTALE-27	1091	1097	ABRE
PvTALE-27	1092	1097	ABRE
PvTALE-27	486	492	CAT-box
PvTALE-27	62	69	GT1-motif
PvTALE-27	1056	1062	GT1-motif
PvTALE-27	1058	1064	GT1-motif
PvTALE-27	914	919	TGACG-motif
PvTALE-27	1098	1103	TGACG-motif
PvTALE-27	58	64	LTR
PvTALE-27	541	548	GCN4_motif
PvTALE-27	914	919	CGTCA-motif
PvTALE-27	1098	1103	CGTCA-motif
PvTALE-27	303	309	Box 4
PvTALE-27	1196	1205	ACE
PvTALE-27	1222	1231	ATCT-motif
PvTALE-27	1089	1098	G-box
PvTALE-27	1091	1097	G-box
PvTALE-27	2	8	ARE
PvTALE-27	258	264	ARE
PvTALE-27	1376	1382	ARE
PvTALE-27	934	943	circadian
PvTALE-27	1191	1200	TC-rich repeats
PvTALE-27	1091	1097	G-Box
PvTALE-28	889	895	GT1-motif
PvTALE-28	107	112	ABRE
PvTALE-28	1423	1431	AE-box
PvTALE-28	107	113	G-box
PvTALE-28	329	335	Box 4
PvTALE-28	442	448	Box 4
PvTALE-28	453	459	Box 4
PvTALE-28	1463	1469	Box 4
PvTALE-28	1445	1450	CGTCA-motif
PvTALE-28	423	429	CAT-box
PvTALE-28	1105	1111	ARE
PvTALE-28	81	90	WUN-motif
PvTALE-28	1397	1404	GARE-motif
PvTALE-28	221	228	AuxRR-core
PvTALE-28	1082	1091	TC-rich repeats
PvTALE-28	1445	1450	TGACG-motif
PvTALE-28	466	475	TCA-element
PvTALE-29	744	754	GATA-motif
PvTALE-29	1150	1157	GARE-motif
PvTALE-29	569	578	circadian

Table S6. (Continued.)

PvTALE-29	1392	1401	TC-rich repeats
PvTALE-29	1390	1396	ARE
PvTALE-29	1442	1448	ARE
PvTALE-29	141	147	G-box
PvTALE-29	285	291	G-box
PvTALE-29	12	18	MBS
PvTALE-29	298	304	MBS
PvTALE-29	504	510	Box 4
PvTALE-29	897	903	Box 4
PvTALE-29	95	100	TGACG-motif
PvTALE-29	1380	1385	TGACG-motif
PvTALE-29	141	147	ABRE
PvTALE-29	142	147	ABRE
PvTALE-29	285	290	ABRE
PvTALE-29	95	100	CGTCA-motif
PvTALE-29	1380	1385	CGTCA-motif
PvTALE-29	141	147	G-Box
PvTALE-30	1443	1451	RY-element
PvTALE-30	410	417	GARE-motif
PvTALE-30	601	607	CCAAT-box
PvTALE-30	743	749	GC-motif
PvTALE-30	825	832	TATC-box
PvTALE-30	1142	1148	GT1-motif
PvTALE-30	910	917	P-box
PvTALE-30	247	256	O2-site
PvTALE-30	1311	1317	ARE
PvTALE-30	1472	1478	ARE
PvTALE-31	1049	1056	MRE
PvTALE-31	508	517	G-box
PvTALE-31	1165	1171	Box 4
PvTALE-31	1325	1331	Box 4
PvTALE-31	1407	1413	Box 4
PvTALE-31	47	52	CGTCA-motif
PvTALE-31	165	171	A-box
PvTALE-31	512	518	CAT-box
PvTALE-31	1279	1292	AT1-motif
PvTALE-31	853	859	CCAAT-box
PvTALE-31	198	204	ARE
PvTALE-31	1235	1241	ARE
PvTALE-31	1240	1246	ARE
PvTALE-31	430	439	circadian
PvTALE-31	702	710	chs-CMA1a
PvTALE-31	1113	1119	TCT-motif
PvTALE-31	124	130	TGA-element
PvTALE-31	994	1002	AE-box
PvTALE-31	47	52	TGACG-motif
PvTALE-31	955	968	sbp-CMA1c
PvTALE-31	716	722	GT1-motif
PvTALE-31	1313	1319	GT1-motif

Table S6. (Continued.)

PvTALE-32	1377	1383	G-box
PvTALE-32	475	481	ARE
PvTALE-32	754	760	ARE
PvTALE-32	921	930	TC-rich repeats
PvTALE-32	1015	1024	TC-rich repeats
PvTALE-32	232	241	ATCT-motif
PvTALE-32	927	933	GT1-motif
PvTALE-32	269	275	Box 4
PvTALE-32	743	749	Box 4
PvTALE-32	1461	1467	Box 4
PvTALE-32	1377	1382	ABRE
PvTALE-32	386	393	TCCC-motif
PvTALE-32	1015	1021	TCT-motif
PvTALE-32	1205	1211	TCT-motif
PvTALE-33	854	860	G-Box
PvTALE-33	1176	1182	G-Box
PvTALE-33	157	163	Box 4
PvTALE-33	229	235	Box 4
PvTALE-33	886	892	Box 4
PvTALE-33	1184	1190	Box 4
PvTALE-33	1376	1382	TGA-element
PvTALE-33	1420	1427	TCCC-motif
PvTALE-33	854	859	ABRE
PvTALE-33	1174	1183	ABRE
PvTALE-33	1177	1182	ABRE
PvTALE-33	1175	1184	G-box
PvTALE-33	268	276	LAMP-element
PvTALE-33	1152	1161	ATCT-motif
PvTALE-33	8	18	GATA-motif
PvTALE-33	552	559	GATA-motif
PvTALE-33	628	638	GATA-motif
PvTALE-34	943	949	GT1-motif
PvTALE-34	1090	1096	GT1-motif
PvTALE-34	707	717	AT-rich element
PvTALE-34	958	969	AuxRE
PvTALE-34	1257	1267	TCA-element
PvTALE-34	14	28	AT1-motif
PvTALE-34	1429	1435	G-box
PvTALE-34	1429	1434	ABRE
PvTALE-34	113	119	GC-motif
PvTALE-34	293	299	Box 4
PvTALE-34	640	646	Box 4
PvTALE-34	644	650	Box 4
PvTALE-34	1075	1082	TATC-box
PvTALE-34	1484	1492	HD-Zip 1
PvTALE-34	184	191	GCN4_motif
PvTALE-34	275	286	Unnamed__1
PvTALE-34	1356	1363	MRE
PvTALE-34	661	667	CAT-box

Table S6. (Continued.)

PvTALE-34	568	574	ARE
PvTALE-35	166	172	Box 4
PvTALE-35	1192	1198	Box 4
PvTALE-35	1398	1404	Box 4
PvTALE-35	301	307	TCT-motif
PvTALE-35	1001	1007	TCT-motif
PvTALE-35	297	306	TC-rich repeats
PvTALE-35	634	639	ABRE
PvTALE-35	634	640	G-box

Table S7. The structure and function of miRNAs targeting the *PvTALE* genes.

miRNA_Acc.	Target_Acc.	Expectation	miRNA_aligned_fragment	Alignment	Target_aligned_fragment	Inhibition
Pvu-miR1514	*PvTALE-13*	3.0	UUCAUUUUGAAAAUAGGCAUUG	.::: . ::::::::.:::::	AGAUGGUGAUUUUCAAGAUGAA	Cleavage
Pvu-miRN2594	*PvTALE-29*	4.0	GGCAGGUUAUCUUUGGCUAU	.:: .::.::.:.::.::.	GUACUCAGAGGUGACUUGCU	Cleavage
Pvu-miR169	*PvTALE-18*	4.0	UGCAGCCAAGGAUGACUUGCC	.::: :: :::: ::::	ACUAAGCCAACCUUGCCUGCA	Cleavage
Pvu-miR319	*PvTALE-16*	4.0	CUUGGACUGAAGGGAGCUCCC	: :: :::.:: :::::::	GUGACCUCUCUGAAGUCCAAG	Translation
Pvu-miR396	*PvTALE-21*	4.0	UUCCACAGCUUUCUUGAACUG	:: :::::::: :::.	AUGUGCAAGAAAGCCAUGGAG	Cleavage
Pvu-miR4415	*PvTALE-28*	4.0	UUGAUUCUCAUCACAACAUGU	: :::: ::. :::::::	AAAUGUGGUGGAUAGAAUCAA	Translation
Pvu-miR5770	*PvTALE-24*	4.0	UAGGACUAUGGUUUGGAUGAG	.::::: :::.:::.:: .:.	UUCAUCAAAAUCAUGGUAUUG	Cleavage
Pvu-miRN2590	*PvTALE-3*	4.0	UUUUGGACAAUUCCAUGCACC	::: :: ::.::: :.::::	GGUUCAAGGGAUUCUUCAAAA	Cleavage
Pvu-miRN2599	*PvTALE-34*	4.0	UUCUUUCAAACAGGCCCUGAG	:: : :.:::: .::::	UCCAAGCCUUGUUUUGAAGAA	Cleavage
Pvu-miR159	*PvTALE-19*	5.0	UUUGGAUUGAAGGGAGCUCUA	.: :::::::::: :	GGAGGGUCCCUUCAAUCCCAC	Cleavage
Pvu-miR160	*PvTALE-35*	5.0	UGCCUGGCUCCCUGUAUGCCA	::: ::: :::::::::..	GGGCUUACUCGGAGCCAGGUG	Cleavage
Pvu-miR160	*PvTALE-5*	5.0	UGCCUGGCUCCCUGUAUGCCA	::: ::: :::::::.	GGGCUAACAAGGAGCCAGGUU	Cleavage
Pvu-miR171	*PvTALE-26*	5.0	UGAUUGAGUCGUGCCAAUAUC	:.:.:: ::..::. ::::.:	GGUGUUUGCGUGAUGCAAUUA	Cleavage
Pvu-miR172	*PvTALE-25*	5.0	AGAAUCUUGAUGAUGCUGCAU	::::. ::::: ::::.:	UAGCAGUUUCAUCA-GAUUUU	Cleavage
Pvu-miR2118	*PvTALE-14*	5.0	UUGCCGAUUCCACCCAUUCCUA	:::: :: : : ::::::..:	GUGGAACGGCUAGAAUCGGUGA	Translation
Pvu-miR2118	*PvTALE-25*	5.0	UUGCCGAUUCCACCCAUUCCUA	.:: :::::: ::::	GCUUGAUUGGUGGAACCGGCAU	Cleavage
Pvu-miR319	*PvTALE-19*	5.0	UGGACUGAAGGGGAGCUCCUUC	:::: .:::::: :::	AUCGGAGGGUCCCUUCAAUCCC	Cleavage
Pvu-miR395	*PvTALE-7*	5.0	UGAAGUGUUUGGGGGAACUCU	: :::::.: :: .:::::	CCAAUUCCCUCCAAGGCUUCA	Translation
Pvu-miR395	*PvTALE-12*	5.0	CUGAAGUGUUUGGGGGAACUC	: :.:::::::::: :	CUCCUACUCCAAACACUUCUG	Cleavage
Pvu-miR395	*PvTALE-1*	5.0	UUGAAGUGUUUGGAG--GAACUC	::::: :: ::::::::::	UAGUUCUGCUGCAAACACUUCAU	Cleavage
Pvu-miR395	*PvTALE-6*	5.0	UUGAAGUGUUUGGAGGAACUC	:::: :::.:: :::::	GAGUUG-UCCAGACCCUUCAA	Cleavage
Pvu-miR396	*PvTALE-5*	5.0	UUCCACAGCUUUCUUGAACUG	:::..:: ::::: ::..	UCGUUUGAGCAAGCUGCGGGG	Cleavage
Pvu-miR397	*PvTALE-11*	5.0	UCAUUGAGUGCAGCGUUGAUG	::: :::: ::.:::	AGACAAGGCUGCUCUCGAUGC	Cleavage
Pvu-miR5770	*PvTALE-33*	5.0	UAGGACUAUGGUUUGGAUGAG	.:.::: ::.:::.:: .:.	UUUAUCAAAAUCAUGGUAUUG	Cleavage
Pvu-miRN2587	*PvTALE-17*	5.0	UCUUCUCUCUAUUGUCACCUU	:: :::.: :::::..::	CAGAAGACGAGAGAGAGGGGA	Translation
Pvu-miRN2587	*PvTALE-3*	5.0	UCUUCUCUCUAUUGUCACCUU	:::: :.:: ::::::::	CUUGUGA-AGUAUAGAGAAGA	Cleavage
Pvu-miRN2588	*PvTALE-4*	5.0	CAUUUGUUUCUUUCUCUCCUUA	: :: . ::.:.:::::::.:	UUAGAGCAGGAGGAAACAAGUC	Cleavage
Pvu-miRN2590	*PvTALE-22*	5.0	UUUUGGACAAUUCCAUGCACC	::: ::.:::::::.	CACGCA-GGAGUUGUCCAAGC	Cleavage
Pvu-miRN2591	*PvTALE-27*	5.0	UUGUUUUUCCUAUUCCACCAAU	:::::: ::::::::::	CAACCUGGAAU-GGAAAAACAA	Translation
Pvu-miRN2594	*PvTALE-4*	5.0	GGCAGGUUAUCUUUGGCUAU	.:. .::.:::.: ::::.	GUGCUCAGAGAUGAGCUGCU	Cleavage
Pvu-miRN2600	*PvTALE-32*	5.0	CAAUGAGGGCAUGUUGUAGGC	: :::::::.:: ..::	UGCAACAACAUGUCCGUGUUC	Cleavage

Table S8. qPCR primers used in this study.

Primer name	Primer (5'- 3')	Length	Tm (°C)	GC (%)
PvTALE-06-F	GAGTTGTCCAGACCCTTCAAT	21	58	48
PvTALE-06-R	TTCATCAGAGCGGTTGTTATCT	22	57	41
PvTALE-10-F	AAGCCTTCTCTGACCTCTCT	20	57	50
PvTALE-10-R	GGTAGTGCTTGTACCTTCTGTC	22	60	50
PvTALE-16-F	GTCAAGTGAAGGCGGAGATAG	21	60	52
PvTALE-16-R	AAGAGCGGAGGAGATTGTTG	20	57	50
PvTALE-20-F	CCTTACCCTACAGACACTGATAAA	24	59	42
PvTALE-20-R	ATTGGCTTCCATACTCGTACTC	22	58	45
PvTALE-22-F	CAAAGGAGGCAAGGAGGATAC	21	60	52
PvTALE-22-R	CCAGACCCGTCATTTCTGATAG	22	60	50
PvTALE-27-F	GAGAACACGGTCAAAGGTAAGA	22	58	45
PvTALE-27-R	TATCCAGTTCGGCTAGAGGT	20	57	50
PvTALE-28-F	TGGTGGAAATGTGGTGGATAG	21	58	48
PvTALE-28-R	GCCTTCAACTCTGAGGAAAGT	21	58	48
PvTALE-29-F	CCACCTCAAATCCTCTCATTCA	22	58	45
PvTALE-29-R	CAAGCATCAGCATGGACATTAC	22	58	45
PvTALE-30-F	TCACTCACTCATCTCTCCTCTC	22	60	50
PvTALE-30-R	GTGGTAGTGTTGTCGGTGTAG	21	60	52
PvTALE-33-F	CTTCCTTTGTACGACCCTTCTT	22	58	45
PvTALE-33-R	TCTTCCTTCACACCACAACC	20	57	50

Tm: melting temperature.