

Short Research Article

Bioinformatics analysis and expression profiling of the jasmonic acid-responsive transcription factor *SIMYB83*

ABSTRACT

Aims: In this study, the candidate gene *SIMYB83* was identified through screening in wild-type tomato and the jasmonic acid (JA)-deficient *spr2* mutant. Its protein structural characteristics, putative regulatory functions, and expression patterns in response to exogenous methyl jasmonate (MeJA) were systematically characterized through bioinformatics and expression analyses. These findings provide a theoretical basis for subsequent functional validation and molecular mechanism studies of *SIMYB83*.

Study Design: This study was conducted using a research design that integrated bioinformatics prediction with gene expression analysis. The *SIMYB83* protein was first subjected to prediction and characterization, followed by an examination of its expression differences across various tissues. Additionally, changes in *SIMYB83* expression were compared between the *spr2* mutant and under conditions of exogenous methyl jasmonate (MeJA) application.

Methodology: In this study, bioinformatics approaches were employed to comprehensively analyze the physicochemical properties, signal peptide, transmembrane domains, conserved domains, secondary structure, tertiary structure, protein–protein interactions, subcellular localization, and promoter cis-regulatory

elements of tomato *SIMYB83*. These analyses were integrated with quantitative real-time PCR to determine the tissue-specific expression pattern of *SIMYB83* and to assess its expression changes in the *spr2* mutant and following exogenous MeJA application.

Results: *SIMYB83* was characterized as a non-transmembrane, hydrophilic protein with a slightly acidic nature, localized to the nucleus. It contains a SANT domain (PLN03091), which is implicated in the regulation of diverse biological processes, including growth and development, light response, drought stress, abscisic acid signaling, and anaerobic induction, as well as various stress-responsive pathways. Among the tissues examined, *SIMYB83* transcript levels were highest in leaves and lowest in fruits. These results suggest a negative regulatory relationship between *SIMYB83* and JA in tomato.

Conclusions: In this study, the fundamental characteristics of tomato *SIMYB83* were elucidated, thereby enriching the current knowledge of the MYB protein family. These findings provide a theoretical foundation for further investigation into the role and molecular mechanisms of *SIMYB83* in JA-mediated regulation of plant growth and development.

Keywords: Tomato; *SIMYB83*; Jasmonic acid (JA); Bioinformatics; Signaling pathway

1. INTRODUCTION

Tomato (*Solanum lycopersicum*), a member of the Solanaceae family, is recognized as a major horticultural crop worldwide due to its considerable nutritional value and distinct flavor (Shang et al., 2025). Tomato is a key model plant in molecular research (Li et al., 2026); however, it is susceptible to both biotic and abiotic stresses during growth (Sánchez-Reyna et al., 2025), which severely impair its yield and quality. Therefore, elucidating the signal transduction and transcriptional regulatory networks of tomato under stress conditions is of great significance for understanding the crop's "growth–defense" trade-off mechanism and for providing potential targets for stress-tolerant breeding.

Plant hormones are a class of low-molecular-weight compounds that play critical roles in plant growth, development, and stress responses (Vrobel et al., 2024; Chen et al., 2024), with jasmonic acid (JA) being one of them. Studies have shown that JA plays a critical role in activating the antioxidant system, thereby enabling plants to adapt to abiotic stress (Li et al., 2024). In the presence of JA, JA is converted into its bioactive form, (+)-7-iso-jasmonoyl-L-isoleucine (JA-Ile) (Dougherty et al., 2024). Upon binding of JA-Ile to the COI1-JAZ complex within the SCF^[COI1] E3 ubiquitin ligase

complex, JAZ proteins undergo ubiquitination followed by proteasomal degradation, thereby relieving the repression of the transcription factor MYC2 by JAZ. This subsequently activates the transcription of MYC2-downstream genes and triggers plant defense and repair responses (Wasternack & Hause, 2024; Luo et al., 2023).

Methyl jasmonate (MeJA), a methyl ester derivative of JA, enhances JA-mediated cold tolerance in tomato by upregulating the expression of SIMYB13 (Demiwal et al., 2024; Zhang et al., 2025). MeJA and JA are widely recognized as lipid-derived stress hormones that play critical roles in plant growth and development, as well as in defense against biotic and abiotic stresses (Wang & Zhang, 2021; Sonatan et al., 2026). They form intricate crosstalk networks with other hormones, including salicylic acid (SA), abscisic acid (ABA), and ethylene (Liu & Timko, 2021), to collectively orchestrate plant growth and defense responses. In the tomato *spr2* (*suppressor of prosystemin-mediated responses 2*) mutant, JA biosynthesis is impaired due to a defect in the chloroplastic fatty acid desaturase involved in the octadecanoid pathway, resulting in significantly reduced JA content in leaves and suppressed JA synthesis upon wounding (Zhao et al., 2016). Therefore, the JA mutant *spr2* represents an ideal material for dissecting the function of the JA signaling pathway and provides a valuable genetic platform for elucidating the JA signaling regulatory network.

Transcription factors enhance plant stress tolerance by regulating the expression of downstream genes (Zhang & Xia, 2023). The MYB transcription factor family is one of the largest transcription factor families in plants (Chen et al., 2025). MYB transcription factors are directly involved in the regulation of the JA signaling pathway: COI1 promotes the ubiquitin-dependent degradation of JAZ proteins, thereby releasing Arabidopsis *AtMYB21* and *AtMYB24*, and subsequently activating the expression of JA-mediated genes involved in anther development and filament elongation (Lu et al., 2025). Overexpression of *PnMYB2* in ginseng enhances the expression of JA biosynthesis-related genes and confers resistance to root rot (Wang et al., 2025). However, MYB transcription factors regulated by the JA signaling pathway in tomato remain to be systematically identified, and the response mechanism of *SIMYB83* in the *spr2* mutant background is still unclear.

Based on our previous transcriptome screening of wild-type tomato and the jasmonic acid-deficient *spr2* mutant, we identified *SIMYB83* as a candidate gene. It was speculated that JA and *SIMYB83* may co-regulate stress tolerance and growth and development in tomato. Whether *SIMYB83* is involved in the JA pathway, as well as its structural and functional characteristics, remain to be elucidated through systematic bioinformatics analysis and experimental validation. Here, we conducted a comprehensive bioinformatics analysis of *SIMYB83*, combined with tissue-specific

expression profiling and analysis of expression changes in mutants and in response to hormone treatment, to systematically characterize its structural features and expression patterns, thereby providing a foundation for subsequent functional studies.

2. MATERIALS AND METHODS

2.1 Plant Materials and Growth Conditions

The plant materials used in this experiment included the cultivated tomato variety ‘Jinguan No. 5’ (wild-type, WT) and the jasmonic acid-deficient mutant *spr2*. The tomato seeds were subjected to heat shock treatment for 5 minutes and subsequently incubated in darkness at 25 °C for 2 days to promote germination. The germinated seedlings were transplanted into nutrient soil and cultivated in a controlled environment growth chamber under the following conditions: light intensity of 20,000 lux, a photoperiod of 16 h light/8 h dark, and temperatures of 25 °C (day) and 18 °C (night). Plants were grown until the four-leaf stage with one unfolding leaf, at which point they were used for subsequent experiments.

2.2 Target Sequences

The gene and protein sequences of tomato SIMYB83 were downloaded from the Phytozome website (<https://phytozome-next.jgi.doe.gov/>).

2.3 Bioinformatics Analysis Tools

The *SIMYB83* gene was analyzed using bioinformatics websites and analysis software (Table 1) for predictions and assessments of protein physicochemical properties, hydrophilicity/hydrophobicity, signal peptide, transmembrane domains, subcellular localization, and promoter cis-regulatory elements.

Table 1. Tools used for bioinformatics analysis

Tools	Website	Effect
ProtParam	http://web.expasy.org/protparam/	Prediction of physicochemical properties of protein sequences
ExPASy ProIScale	Expasy - ProtScale	Hydrophilic and hydrophobic analysis of proteins
SignalP-5.0	https://services.healthtech.dtu.dk/service.php?SignalP-5.0	Prediction and analysis of signal peptides
TMHMM-2.0	https://services.healthtech.dtu.dk/service.php?TMHMM-2.0	Transmembrane structure analysis of proteins
NCBI	https://www.ncbi.nlm.nih.gov/Structure/cdd/wrpsb.cg	Conserved domain analysis
SOPMA	http://npsa-pbil.ibcp.fr/cgi-bin/npsa_automat.pl?page=/NPSA/npsa_sopma.html	Protein secondary structure prediction
SWISS-MODEL	https://swissmodel.expasy.org/	Protein 3D structure prediction

Plant-mPLoc	http://www.csbio.sjtu.edu.cn/bioinf/plant-multi/	Prediction of subcellular localization of proteins
PLANTCARE	https://bioinformatics.psb.ugent.be/webtools/plantcare/html/	Promoter prediction
STRING	STRING: functional protein association networks	Protein-protein interaction prediction
Tbtools-II		Analysis of cis-regulatory elements in promoters

2.4 MeJA Treatment and Gene Expression Analysis

To investigate the response of *SIMYB83* to jasmonic acid signaling, tomato seedlings were grown until the two-leaf stage. Seedlings with uniform growth were selected and subjected to exogenous application of 100 μ M methyl jasmonate (MeJA) solution. Samples were collected at 5 days after treatment, immediately flash-frozen in liquid nitrogen, and subsequently stored at -80°C until further use for RNA extraction.

To collect samples from *spr2* mutant seedlings, plants were grown until the two-leaf stage. Seedlings were then harvested, wrapped in aluminum foil, labeled, immediately frozen in liquid nitrogen, and subsequently stored at -80°C until further use for RNA extraction.

The successfully extracted RNA was reverse transcribed into cDNA, and the expression levels of *SIMYB83* in MeJA-treated plants and *spr2* mutants were examined by quantitative real-time PCR (qRT-PCR).

2.5 Statistical Analysis

Data were organized using Microsoft Excel 2021. Multiple comparisons were performed using Tukey's honestly significant difference (HSD) test ($P < 0.05$) in GraphPad Prism 10 software to assess statistically significant differences. Graphs were generated using GraphPad Prism 10, and data are presented as the mean \pm standard deviation (SD).

3. RESULTS AND ANALYSIS

3.1 Physicochemical Properties and Hydrophobicity of the SIMYB83 Protein

As shown in Table 2, the SIMYB83 protein sequence was analyzed using ProtParam. The results indicated that the SIMYB83 protein consists of 353 amino acids, with a predicted molecular weight of 39,459.76 Da and a theoretical isoelectric point (pI) of 6.06, indicating a slightly acidic nature. The instability index was 56.82, classifying it as an unstable protein. ProtScale analysis (Fig. 1) revealed a grand average of hydropathicity (GRAVY) of -0.725 , suggesting a hydrophilic property.

Table 2. Analysis of physicochemical properties of SIMYB83

Gene name	Gene ID	Genome position	ORF length (bp)	Deduced Protein				
				Length (aa)	MW (Da)	pI	GRAVY	Instability index
<i>SIMYB83</i>	LOC101262590	Chr11:56833703-56837291	1062bp	353	39459.76	6.06	-0.725	56.82

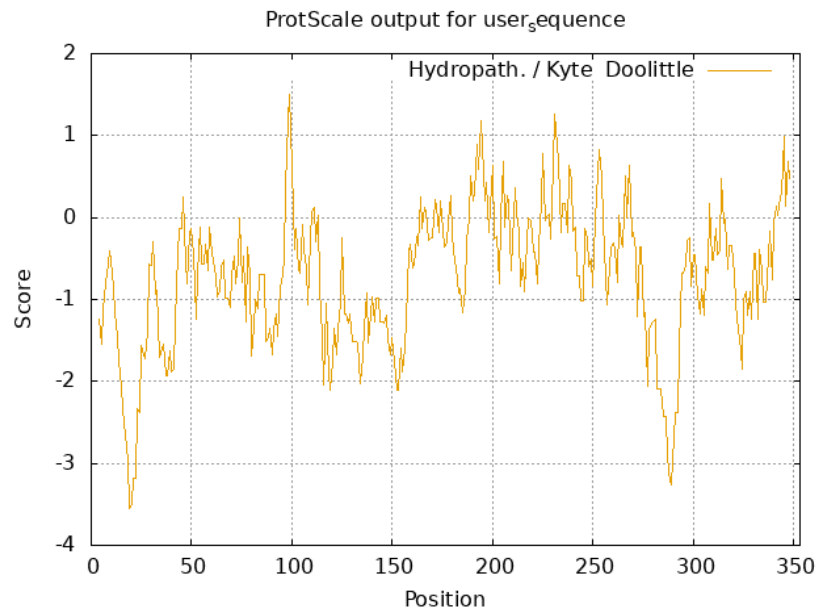


Fig. 1. Prediction of hydrophilicity and hydrophobicity of the SIMYB83 protein

3.2 Prediction of Signal Peptide and Transmembrane Domains in the SIMYB83 Protein

A signal peptide is a short peptide chain, typically 5 to 30 amino acids in length, that directs newly synthesized proteins to the secretory pathway and is usually located at the N-terminus of secreted proteins. Analysis of the SIMYB83 protein using SignalP-5.0 revealed the absence of a signal peptide cleavage site, indicating that SIMYB83 does not contain a signal peptide (Fig. 2A).

The transmembrane domain structure of the SIMYB83 protein was further predicted using TMHMM-2.0 (Fig. 2B). The results showed that SIMYB83 contains no transmembrane domains, indicating that it is a non-secretory, non-transmembrane protein.

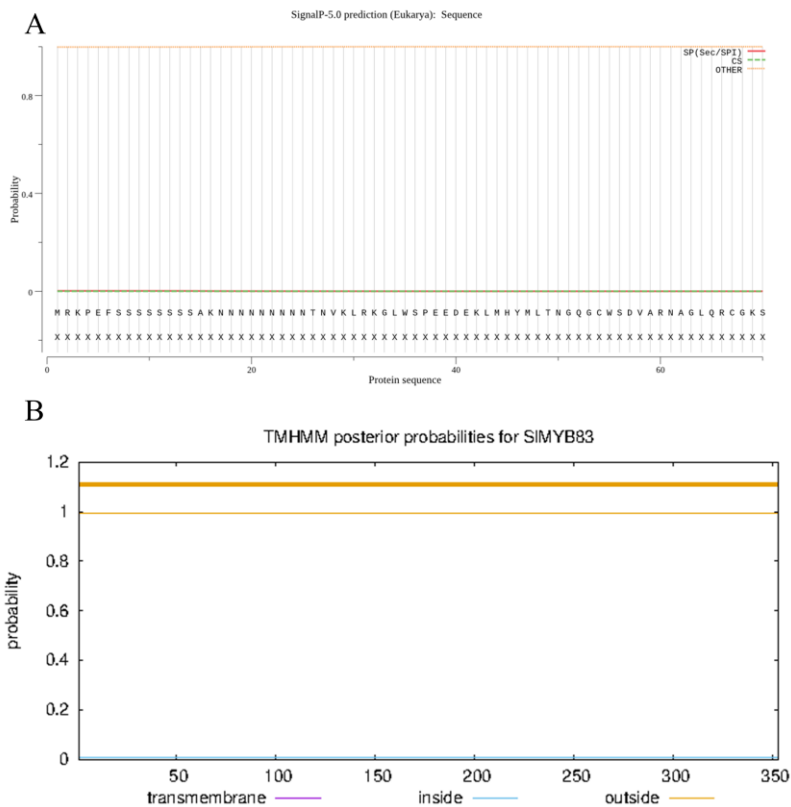


Fig. 2. Prediction of signal peptides and transmembrane domains for SIMYB83 protein

Note: A, Prediction of the signal peptide of the SIMYB83 protein. B, Prediction of transmembrane domains in the SIMYB83 protein.

3.3 Prediction of Secondary and Tertiary Structures of the SIMYB83 Protein

The secondary structure of the SIMYB83 protein was predicted using SOPMA (Fig. 3A). The results showed that the secondary structure of this protein is primarily composed of α -helices, extended strands, β -turns, and random coils. Among these, random coils accounted for the highest proportion at 62.89%, corresponding to 222 amino acids, followed by α -helices at 24.65% (87 amino acids), extended strands at 7.37% (26 amino acids), and β -turns at 5.10% (18 amino acids). This suggests that SIMYB83 may exert its biological functions through random coil regions mediating interactions with target proteins.

The tertiary structure of the protein encoded by SIMYB83 was predicted using homology modeling via the SWISS-MODEL online server (Fig. 3B). The results showed that the protein typically exists as a monomer, with magnesium ions likely serving as its binding ligands. Tertiary structure modeling revealed that SIMYB83 shares 57.14% sequence identity with the Arabidopsis transcription factor WER, suggesting that it may function as an R2R3-MYB transcription factor involved in DNA recognition.

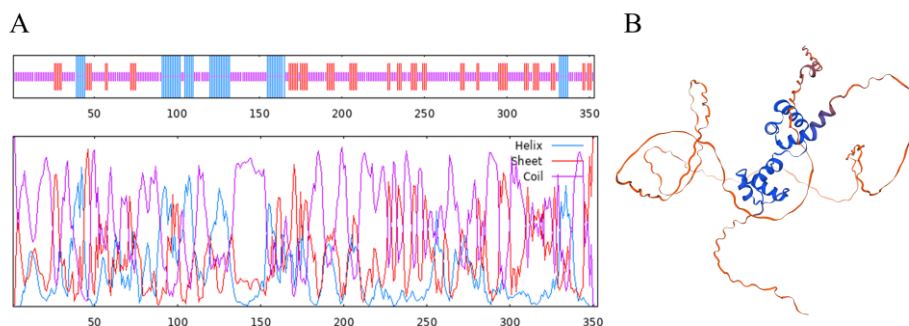


Fig. 3. Prediction of the secondary and tertiary structures of the SIMYB83 protein

Note: A, Prediction of the secondary structure of the SIMYB83 protein. B, Prediction of the tertiary structure of the SIMYB83 protein.

3.4 Prediction of Functional Domains and Subcellular Localization of the SIMYB83 Protein

Analysis of the *SIMYB83* gene sequence was performed using the Phytozome online tool, revealing that the coding sequence (CDS) is 1,062 bp in length. The conserved domain of the gene was analyzed using the NCBI online tool, and the results showed that the SIMYB83 protein contains a SANT domain (PLN03091) and belongs to the cl33633 superfamily (Fig. 4A).

Subcellular localization was predicted using the Plant-mPLOC online tool. The results indicated that the SIMYB83 protein is localized to the nucleus (Fig. 4B), suggesting that this protein primarily functions in the nucleus, which is consistent with the previous finding that it lacks transmembrane domains.



Fig. 4. Prediction of functional domains and subcellular localization of the SIMYB83 protein

Note: A, Prediction of functional domains in the SIMYB83 protein. B, Prediction of subcellular localization of the SIMYB83 protein.

3.5 Cis-Regulatory Element Analysis of the *SIMYB83* Gene Promoter

The promoter sequence of the *SIMYB83* gene was obtained by extracting the 2,000 bp region upstream of the transcription start site from the annotated tomato genome file. This sequence was then submitted to the PlantCARE database for analysis to identify cis-regulatory elements within the *SIMYB83* promoter. The resulting data were subsequently imported into Tbttools II software for visualization and further analysis (Fig. 5). The results revealed the presence of multiple hormone-responsive and stress-responsive elements within the promoter sequence, including those involved in MeJA, abscisic acid (ABA), drought, promoter and enhancer regions, light response, anaerobic induction, and auxin-responsive element. These findings suggest that *SIMYB83* may be involved in light regulation, stress responses, and crosstalk among multiple hormone signaling pathways.

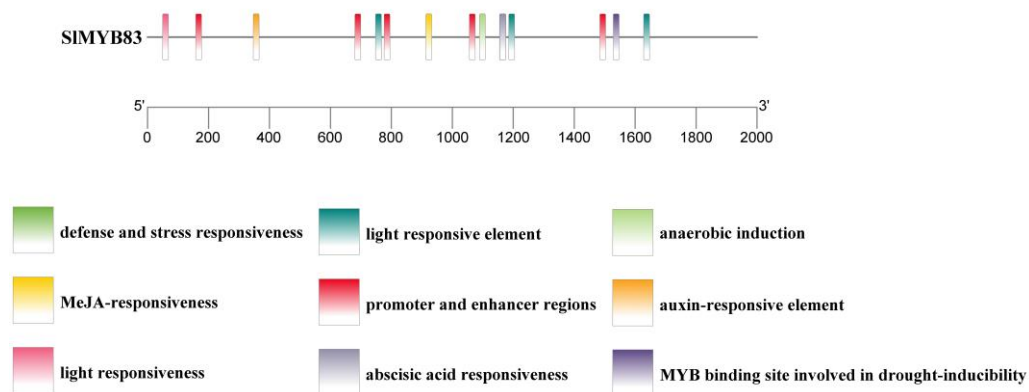


Fig. 5. Analysis of cis-acting elements in the *SIMYB83* gene

3.6 Protein-Protein Interaction Network Analysis of *SIMYB83*

Protein-protein interaction network analysis was performed using STRING to identify proteins potentially interacting with *SIMYB83* (Fig. 6). The results indicated that *SIMYB83* may interact with proteins such as Phy7 and RPS25, suggesting a potential role in mediating temperature stress responses (Table 3).

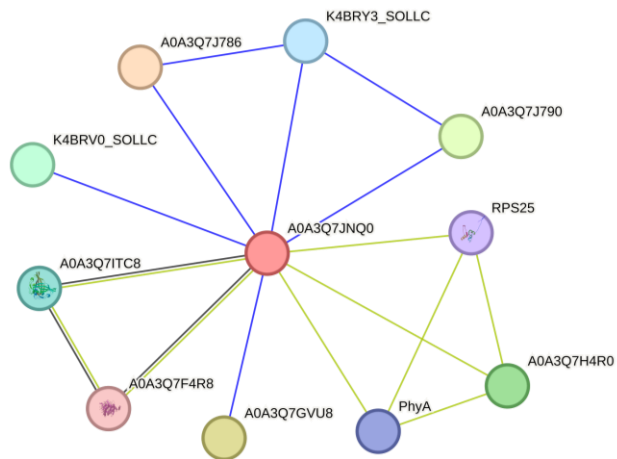


Fig. 6. Analysis of the protein-protein interaction network of the SIMYB83 protein

Table 3. Predicted interacting proteins and their information

Name	Interacting protein name	Information on interacting proteins
SIMYB83	A0A3Q7J786	HTH myb-type domain-containing protein.
	A0A3Q7GVU8	HTH myb-type domain-containing protein.
	A0A3Q7J790	HTH myb-type domain-containing protein.
	A0A3Q7H4R0	Uncharacterized protein.
	K4BRV0_SOLLC	HTH myb-type domain-containing protein.
	A0A3Q7ITC8	NAC domain-containing protein.
	K4BRY3_SOLLC	HTH myb-type domain-containing protein.
	PhyA	Phytochrome; Regulatory photoreceptor which exists in two forms that are reversibly interconvertible by light: the Pr form that absorbs maximally in the red region of the spectrum and the Pfr form that absorbs maximally in the far-red region.
	RPS25	40S ribosomal protein S25; Belongs to the eukaryotic ribosomal protein eS25 family.
	A0A3Q7F4R8	Cellulose synthase; Belongs to the glycosyltransferase 2 family. Plant cellulose synthase subfamily.

3.7 Tissue-Specific Expression Analysis of SIMYB83

To determine the expression levels of *SIMYB83* in different tomato tissues, RNA was extracted from root, stem, leaf, flower, and fruit tissues, followed by qRT-PCR analysis. The results showed that *SIMYB83* transcript levels were highest in leaves, followed by flowers and roots, then stems, and lowest in fruits (Fig. 7).

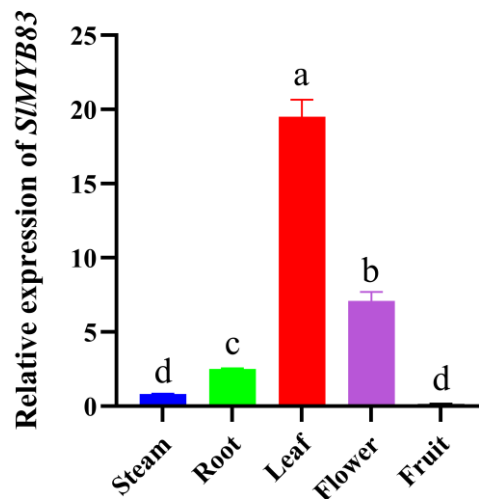
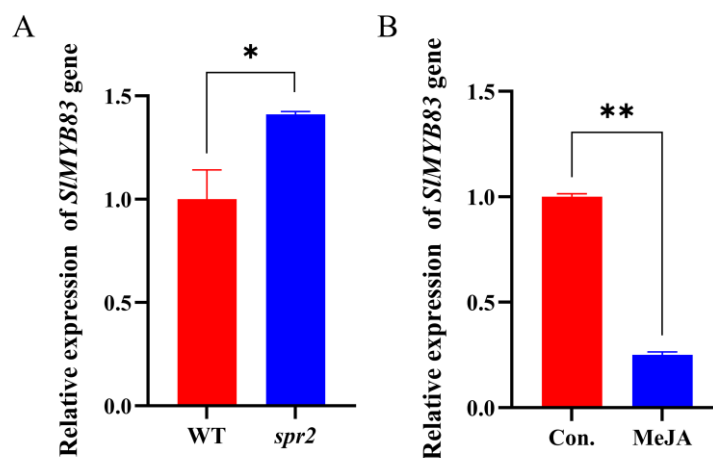


Fig. 7. Tissue specific expression analysis of *SIMYB83*

Note: Each bar represents the RNA content in different parts of the tomato, and the letters (a, b, c, d) indicate the statistical means of the groups based on Tukey's test ($P < 0.05$).

3.8 Regulatory Role of JA in *SIMYB83* Expression

To investigate the regulatory relationship of JA on *SIMYB83* expression, changes in *SIMYB83* transcript levels were examined in the jasmonic acid-deficient mutant *spr2* and in plants subjected to exogenous MeJA application. In the *spr2* mutant, the expression level of *SIMYB83* was significantly higher than that in the WT (Fig. 8A), suggesting that JA may negatively regulate *SIMYB83* expression. To further validate this, we applied exogenous MeJA to WT plants. The results showed a highly significant decrease in *SIMYB83* expression (Fig. 8B), further supporting the negative regulatory role of JA on *SIMYB83*.



Note: A, Comparison of *SIMYB83* expression in WT and *spr2*. B, Comparison of *SIMYB83* expression levels in WT and exogenous sprayed MeJA plants. *indicate significant differences between WT and different plants. (* $P < 0.05$ and ** $P < 0.01$).

4. DISCUSSION

The JA signaling pathway is closely associated with stress defense and resistance in plants (Xu et al., 2024), constituting a central pathway that integrates stress responses with the regulation of growth and development. MYB transcription factors enhance plant tolerance by regulating reactive oxygen species (ROS) metabolism and the JA signaling pathway (Wang et al., 2026). As pivotal downstream regulators of the JA signaling pathway, functional characterization of MYBs has become a major focus in plant molecular biology research. In this study, we performed systematic bioinformatics analysis and expression profiling of *SIMYB83*, which was identified through screening in wild-type tomato and the *spr2* mutant, and characterized its jasmonic acid-responsive features, providing important theoretical value.

The structural features of transcription factors are often closely associated with their functions. *SIMYB83* exhibits a typical MYB conserved domain, lacks signal peptide and transmembrane domains, and is a nuclear-localized hydrophilic protein, consistent with the typical characteristics of plant transcription factors (Cui & An, 2024). The secondary structure is primarily composed of random coils and α -helices, while the tertiary structure forms a conserved helix-turn-helix (HTH) motif, providing a structural basis for DNA binding and transcriptional regulatory function. Cis-regulatory element analysis of the promoter is a crucial approach for inferring gene regulatory mechanisms. The promoter region contains specific MeJA-responsive elements, indicating that *SIMYB83* is regulated by JA signaling. Additionally, the presence of light-responsive, stress-responsive, and various hormone-related elements suggests that *SIMYB83* may not only participate in JA-mediated defense responses but also play a role in orchestrating the balance between growth, development, and stress adaptation. Protein-protein interaction network analysis further indicated that *SIMYB83* may interact with proteins such as Phy7 and RPS25 and participate in temperature stress responses, suggesting that it likely occupies a key node in the hormone regulation and defense response network of tomato. Tissue-specific expression analysis revealed that *SIMYB83* is highly expressed in leaves, which are primary organs for stress perception and JA synthesis in plants. Consistent with this, it has been reported that *MaMYBR30* in mulberry is also expressed in leaves and can be induced by drought stress and salt stress (Liu et al., 2024). It is therefore speculated that *SIMYB83* primarily functions in leaves to participate in JA-mediated stress responses, such as disease resistance and drought tolerance. Its constitutive expression pattern

suggests that *SIMYB83* is not only involved in stress responses but may also regulate the growth and development processes of tomato roots, stems, flowers, and fruits, indicating functional diversity.

Notably, *SIMYB83* was significantly upregulated in the *spr2* mutant but markedly downregulated following exogenous MeJA treatment, suggesting a potential negative correlation between its expression and jasmonic acid signaling. In the *spr2* mutant, which exhibits reduced endogenous JA content, *SIMYB83* expression is elevated, suggesting that JA may exert a repressive effect on *SIMYB83* under normal conditions. Following exogenous application of MeJA, *SIMYB83* expression was significantly downregulated, further validating this negative regulatory relationship. This observation suggests that *SIMYB83* may act as a negative feedback-responsive gene in the JA signaling pathway, or alternatively, that it is derepressed under JA-deficient conditions to participate in compensatory transcriptional regulation.

5. CONCLUSIONS

Bioinformatics analysis of the *SIMYB83* gene revealed that *SIMYB83* lacks a signal peptide and transmembrane domains, with subcellular localization predicted to be in the nucleus. Its conserved domain is limited to the SANT domain PLN03091, and random coils constitute the largest proportion of its secondary structure. Three-dimensional structural modeling predicted 57.14% sequence similarity with the Arabidopsis transcription factor WER, confirming its role as an R2R3-MYB transcription factor involved in DNA recognition. Additionally, multiple stress-responsive elements, including MeJA, drought and ABA, were identified in its promoter sequence. qRT-PCR analysis revealed that *SIMYB83* exhibited the highest expression level in leaves and the lowest in fruits. In the *spr2* mutant, *SIMYB83* expression was significantly higher than that in the WT, whereas upon exogenous application of MeJA to WT plants, *SIMYB83* expression was markedly decreased, suggesting that JA plays a negative regulatory role in suppressing *SIMYB83* expression.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

All authors have declared that no competing interests exist, and that no artificial intelligence (AI) technologies were used in the process of writing this paper.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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