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EDITED BY
Wei Zhao,
Institute of Geographic Sciences and Natural Resources Research (CAS),
China

REVIEWED BY
Wei Song,
Peking Union Medical College Hospital (CAMS),
China
Wei Zhou,
Zhejiang Chinese Medical University,
China

*CORRESPONDENCE
Chunsheng Liu
✉ max_liucs@263.net
Dan Jiang
✉ jiangdan1027@163.com

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Root-specific flavones and critical enzyme genes involved in their synthesis changes due to drought stress on *Scutellaria baicalensis*

Ping Li, Guangxi Ren, Fei Wu, Jiaxin Chen, Dan Jiang* and Chunsheng Liu*

School of Chinese Materia Medica, Beijing University of Chinese Medicine, Beijing, China

Introduction: *Scutellaria baicalensis* is rich in bioactive flavonoid, which are widely used in clinical therapy. Many environmental factors, such as water and temperature, affect gene expression and secondary metabolites accumulation in plants.

Methods: In this study, to explore the effect of drought stress on the accumulation of flavonoids and gene expression in *S. baicalensis* seedlings, 4-week-old *Scutellaria baicalensis* seedlings were treated with different concentrations of PEG6000 to simulate drought stress. The contents of four root-specific flavones (baicalein, wogonin, baicalin, and wogonoside) in samples under different treatments were quantitatively analyzed by high performance liquid chromatography (HPLC). The expression levels of flavonoid biosynthesis-related genes (P*AL1*, *PAL2*, *CHS,* and *UBGAT*) were determined by real-time quantitative PCR (qRT-PCR). Also, a correlation analysis between flavonoid contents and gene expression levels was made.

Results: The HPLC results revealed that 5 and 10% PEG6000 treatments significantly increased the content of four flavonoids, with 5% PEG 6000 treatment being the most beneficial to the flavonoids accumulation. The qRT-PCR results showed that *PAL2* and *CHS* gene expressions differed significantly in different organs, while *PAL1* and *UBGAT* had poor organ-specific. For genes in roots, the expression of *PAL1* and *UBGAT* was the highest in 5% PEG6000 treatment, and *PAL2* and *CHS* were the highest in 10% PEG6000 treatment. Compared with other concentrations of PEG6000, 5 and 10% PEG6000 were more advantageous for gene expression. Collectively, PEG6000 at a low concentration promoted the accumulation of flavonoids and the expression of related genes. Additionally, the correlation results demonstrated that *PAL1*, *PAL2*, *CHS*, and *UBGAT* genes in roots stimulated the formation and accumulation of the four flavonoids to varying degrees, while the exception of *PAL2* gene expression in roots was negatively correlated with wogonin content.

Discussion: This study for the first time investigated the effect of drought stress on the downstream gene *UBGAT* in *S.baicalensis* seedlings as well as the correlation between gene expression and flavonoid content in *S. baicalensis* seedlings under drought stress, providing a new sight for studying the effects of drought stress on flavonoid accumulation and related gene expression in *S. baicalensis*.

KEYWORDS

***Scutellaria baicalensis*, drought stress, gene expression, qRT-PCR, HPLC, flavonoids**

1. Introduction

Scutellaria baicalensis (Baikal skullcap), known locally as Huangqin, is a perennial herb native to East Asia (Vrabec et al., 2022). Its dried roots have been an important herb in traditional Chinese medicine for a long time (Wu et al., 2022; Zhou et al., 2022). It has significant therapeutic effects on a wide range of illnesses, including various cancers, inflammation, diabetic nephropathy, oxidative stress (Men et al., 2021; Zhou et al., 2021). Baicalein, wogonin, baicalin, and wogonoside are called as root-specific flavones of *S. baicalensis* (Zhao et al., 2018). There is a lack of 4′-hydroxy (4′-deoxyflavone) in ring B, which is mainly responsible for the pharmacological activities of *S. baicalensis* (Zhao et al., 2016). Baicalein has been shown to suppress lipopolysaccharide-induced acute lung injury, ameliorate osteoporosis, and exert an antitumor effect on cervical cancer (Cai et al., 2021; Luo et al., 2021; Jiang et al., 2022). It has been reported that wogonin inhibits cardiac hypertrophy and alleviates kidney tubular epithelial injury (Lei et al., 2021; Shi et al., 2021). Baicalin has been proven to have remarkable effects on various cancer treatments and cardiovascular, hepatic, and renal protection (Singh et al., 2021; Yang et al., 2021; Zhao et al., 2021). Wogonoside can also treat cancer and inflammation (Gu et al., 2021; Huang et al., 2021). Thus, the accumulation of root-specific flavones is an essential aspect of studying *S. baicalensis*.

Flavonoid synthesis and accumulation depend on the crucial enzymes in their synthesis pathway (Zhao et al., 2018; Wang S. et al., 2019). Studying crucial enzyme genes of the biosynthetic pathway can provide a theoretical basis for the molecular mechanism of active flavonoid accumulation. Flavonoids are usually synthesized through the phenylpropanoid pathway (Ahmed et al., 2021). Their biosynthetic routes are shown in Figure 1. Phenylalanine ammonia-lyase (PAL) catalyzes the first and rate-limiting step of the phenylpropanoid pathway, which provides precursors for a diversity of secondary metabolites, including flavonoids (Liu et al., 2006). It converts primary metabolism into secondary metabolism (Olsen et al., 2008). Chalcone synthase (CHS) is a crucial enzyme that catalyzes the first rate-limiting step of the flavonoid biosynthesis pathway (Wang et al., 2018). UDP-glucuronate (baicalein 7-O-glucuronosyltransferase, UBGAT) is responsible for catalyzing the formation of flavonoid glycosides. Nagashima analyzed the properties of UBGAT purified from cultured cells of *S. baicalensis* and catalyzed the reaction with various glycosides. The outcomes demonstrated that it only reacted with UDP-glucuronide, baicalein, and wogonin as substrates (Nagashima et al., 2000). Studies show that the flavonoid content of the planted *S. baicalensis* is lower than that of wild resources (Li et al., 2018; Tian et al., 2018). Therefore, exploring the method to improve the flavonoid content of cultivated medicinal materials is conducive to protecting the wild medicinal resources and preventing the wild resources from being excessively excavated. It has been proven crucial gene expression correlates with flavonoid accumulation (Ma et al., 2014). Accordingly, it is of practical significance for protecting medicinal plant resources and improving the quality of herbs to explore active compound accumulation and crucial enzyme gene expression.

Abiotic factors are fundamental components of the environment that determine plant distribution and productivity (Zhang H. et al., 2022). In nature, plants are constantly challenged by adverse abiotic environmental conditions such as drought, heat, and excess salt levels

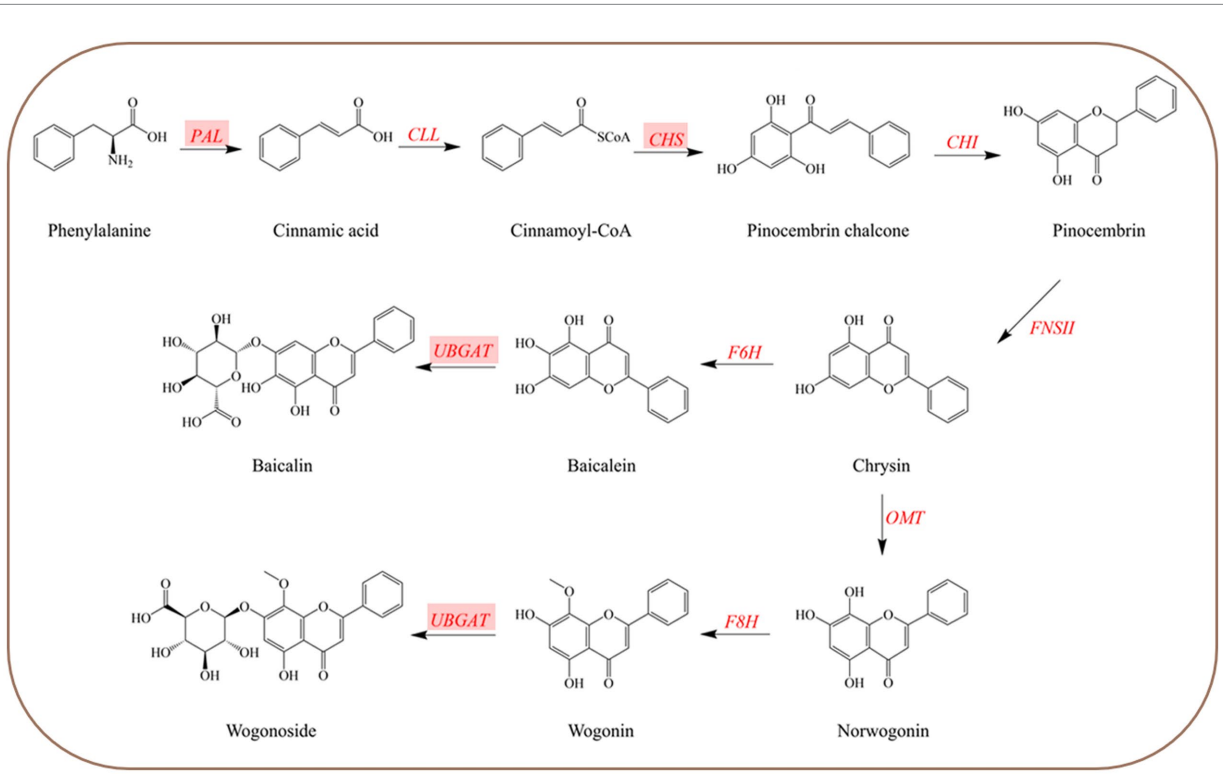

FIGURE 1
Biosynthetic pathways of four root-specific flavones (baicalin, baicalein, wogonin, and wogonoside) in *Scutellaria baicalensis*. Highlighted genes are the studied genes in this paper.

in the soil, which negatively affect crop productivity (Xie et al., 2021; Mahecha et al., 2022; Paparella et al., 2022). The gene expression and bioactive component accumulation are affected by environmental stress (Xu et al., 2020; Jiang et al., 2021; Xu et al., 2021). Accumulating evidence has proven that appropriate drought stress has the potential to promote the production of bioactive metabolites like flavonoids (Jia et al., 2018; Jia and Wang, 2019; Yang et al., 2020; Zhang et al., 2020). Thus, comprehending how plants respond to abiotic stresses and adverse environmental conditions is critical for protecting the ecology and evolution of living systems.

Polyethylene glycol (PEG) is composed of a large molecular size and is nontoxic (Chen et al., 2020). It can reduce the water potential of the surrounding environment of the cells, causing and cause drought (Wang et al., 2020). PEG6000 is often used to simulate drought stress treatments (Jian et al., 2022; Tang et al., 2022). Thus, this study proposed the hypothesis that appropriate drought treatment can promote the accumulation of flavonoids and gene expression in *S. baicalensis* seedlings. PEG6000 was used to treat seedlings to simulate drought stress. The accumulation of four bioactive root-specific flavones in roots during drought stress was determined by high performance liquid chromatography (HPLC). Simultaneously, the expression of four crucial genes related to flavonoid synthesis in the roots, stems, and leaves of *S. baicalensis* was determined by real-time quantitative PCR (qRT-PCR). And the effect of drought on the expression of related genes was comprehensively analyzed. This study would provide meaningful information on the accumulation of compounds in *S. baicalensis* and insights into the study of quality improvement under stressed conditions.

2. Materials and methods

2.1. Plant material and chemicals

The seeds of *S. baicalensis* were collected from Inner Mongolia, China. Healthy seeds were selected for the pot experiment. They were sown in a 16.8 cm × 15.5 cm (height × upper diameter) pot and cultured in an artificial illumination incubator (25°C, 16 h of light, and 8 h of dark light).

Compounds wogonoside (CAS: 51059–44-0), baicalin (CAS: 21967–41-9), wogonin (CAS: 632–85-9), and baicalein (CAS: 491–67-8) were procured from Yuanye (Shanghai, China) and had purity >98%. Acetonitrile (CAS: 75–05-8, HPLC) and formic acid (CAS:6449-79-2, LC/MS) were acquired from Fischer Scientific (Hampton, NJ, United States). The remaining reagents were bought locally and were of analytical grade.

2.2. Drought stress treatment

Four-week-old healthy seedlings were selected to be subjected to the follow-up treatment. There were six groups in each of the three replicates (three pots per replicate, 30 seedlings per pot). The six experimental groups were treated with 0, 5, 10, 15, 20, and 25% PEG 6000 under drought stress (Meher and Shivakrishna, 2018; Wang Z.Y. et al., 2019; Batool et al., 2022). The drought stress experiment lasted 5 days, and the solution was poured every other day for three times. Immediately after the experiment, the seedlings were frozen in liquid nitrogen and stored at −80°C.

2.3. Content determination

Thirty seedlings of similar growth were selected per replicate for each treatment for content determination. Dried the roots of *S. baicalensis* at 55–60°C and rushed them (Zhang L.W. et al., 2022). The powder was precisely weighed to approximately 0.1 g and placed in a triangular flask. Then, flavonoids from *S. baicalensis* were extracted by the extraction method optimized by our research group. 1 mL of 50% ethanol was added. The sample was accurately weighed and extracted with an ultrasonic step for 23 min. The sample was then filtered through a 0.22 μm filter.

The content of flavonoids in *S. baicalensis* was determined by HPLC established by our research group. The mobile phase was acetonitrile (A) −0.1% formic acid aqueous solution (B). The gradient elution was used under the following conditions: 0.01–4 min, 10–20% A; 4–12 min, 20–22% A; 12–22 min, 22–24% A; 22–49 min, 24–28% A; 49–52 min, 28–35% A; 52–60 min, 35–45% A; 60–64 min, 45% ~ A; 64–70 min, 55–10% A; 70–85 min, 10% A. The volume flow rate was 0.8 mL/min. The detection wavelength was 274 nm. The injection volume was 10 μL. The column temperature was 30°C.

2.4. Total RNA extraction, cDNA synthesis, and qRT-PCR

Total RNA was extracted from the roots, stems, and leaves of *S. baicalensis* according to TRNzol Universal Total RNA Extraction Reagent Operation Guide (Tiangen, Beijing, China). RNase-free water was used to dissolve the purified RNA. Using the extracted RNA as a template, cDNA was obtained by reverse transcription based on the instructions of the Evo M-MLV Mix Kit with gDNA Clean for qPCR kit (AGBio, Hunan, China) and carried out qRT-PCR following the instructions of the ChamQ Universal SYBR qPCR Master Mix (Vazyme, Nanjing, China) using an SYBR Green-based PCR assay. The information on primers used in this research is shown in Table 1. The expression levels of *PAL1*, *PAL2*, *CHS*, and *UBGAT* were determined using CT values and calculated using the $2^{-\triangle\triangle Ct}$ method (Liu et al., 2019). Each qRT-PCR reaction technique was three replicates. Each experiment has three biological repetitions.

2.5. Statistical analysis

The HPLC results were monitored and analyzed through Lab Solutions 5.92 (Shimadzu Scientific Instruments INC.). GraphPad Prism 8.3.0 (GraphPad Software INC.) and Adobe Illustrator 2020 (Adobe INC.) were applied to mapping analyses. OriginPro 2021 (OriginLab Corporation.) and IBM SPSS® Statistics 25 (International Business Machines Corporation) were used in this research work for data evaluation. All the biological samples were three replicates.

3. Results

3.1. Accumulation of root-specific flavones in roots *Scutellaria baicalensis* under drought stress

The significance of the Levene variance homogeneity test was greater than the significance level of 0.05 (Table 2), so it can

TABLE 1 The information on primers used in this research.

	Forward primer	Reverse primer	Accession number
Actin	TTGATCTTGCTGGTCGTGATCTCA	TGTTTCTAGCTCTTGCTCGTAGTCG	HQ847728
CHS	GAGCTAGGGTTCTCGTCGTC	AAGAGGGAGTTCCAGTCGG	AB008748
PAL1	GGCAAACCCTGTGACGAA	GCCTGGCATAGAGCGACTA	OP018668
PAL2	TCTACGGCACTTGGAGGAG	CATCAACGGATAGTTCACGCT	OP018669
UBGAT	CCCGACATCACGGACAA	AAATGAGGGGCAAACCAG	OP018670

TABLE 2 Results of statistical analysis of four root-specific flavones content.

Flavonoids	Variance homogeneity test results of mean		Results of one-way ANOVA between groups			
	Levene statistic	Sig.	Sum of squares	Mean square	*F*	Sig.
Baicalein	2.829	0.065	206.223	41.245	7889.677	0.000
Wogonin	2.069	0.140	56.172	11.234	2116.112	0.000
Baicalin	2.572	0.084	415.735	83.147	1530.721	0.000
Wogonoside	2.311	0.109	20.578	4.116	14569.957	0.000

be considered that the variance between sample data is homogeneous. The result indicated that one-way ANOVA could be used. The significance of the *F* value for all four compounds was 0.000, which was lower than the significance level of 0.05 (Table 2). It demonstrated that drought stress significantly affected the accumulation of four flavonoids.

In the LSD test, the mean difference results of each group showed that different PEG6000 concentrations treatments could lead to significant differences in flavonoid content. There was no significant difference in baicalein between 15 and 25% PEG6000 groups, but there were significant differences among other groups. For baicalin, there was no significant difference between 15, 20, and 25% PEG6000, but there were significant differences between other groups. As to wogonin and wogonoside, different treatments resulted in significant differences among all groups.

The contents of baicalein, wogonin, baicalin, and wogonoside were shown in Figure 2. The concentrations of baicalein under drought stress of 10% PEG6000 and 15% PEG6000 were 8.124 and 7.502 mg/g, respectively, which increased by 20.44 and 11.22% compared with the control group (0% PEG6000). Compared with 0% PEG6000, baicalin in the treated group of 10% PEG6000 and 15% PEG6000 was 68.72 and 35.86% higher. Correspondingly, the content of wogonoside increased by 82.75 and 56.59%, respectively, under the two treatments. Compared with the control group, the 5% PEG6000 group increased the content of wogonin by 45.81%.

3.2. Expression of crucial genes in roots, stems, and leaves of *Scutellaria baicalensis* under drought stress

Under drought stress, the RNA of the samples was extracted and the RNA quality is shown in Supplementary Figure S1 and Supplementary Table S1. Then the expression of four crucial enzyme genes was detected in different organs of *S. baicalensis* (Figure 3). The effects of different organs of *S. baicalensis* and the concentration of PEG6000 on gene expression were analyzed by multivariate analysis of variance (Table 3). Results of tests of between-subjects effects revealed that the effects of different concentrations of PEG6000 on *CHS, PAL1, PAL2,* and *UBGAT* were significant, and the expression levels of *CHS*, *PAL1*, and *PAL2* were significantly different in various organs of *S. baicalensis*.

The LSD results suggested that the relative expression of *CHS* was significantly different from other treatments when treated with 10% PEG6000. Compared with other treatment groups (except 10%), the relative expression of *PAL1* in the 5% PEG6000 treatment group was significantly different. Between the 5% PEG6000 treatment group and the other treatment concentration groups, the relative expression of *UBGAT* was considerably different. In the case of *PAL2*, the 10% PEG6000 treatment group was significantly different from the control group, as were the 15% PEG6000 and 25% PEG6000 treatment groups. In addition, the gene expression levels of genes in roots treated with different concentrations of PEG6000 were compared to explore the best treatment of four genes that affect the production of root-specific flavones. For *PAL1* and *UBGAT*, gene expression was highest when 5% treatment was used.

Meanwhile, the highest expression of *PAL1* and *UBGAT* genes was observed in the 5% treatment, and the highest expression of *PAL2* and *CHS* genes was observed in the 10% treatment, suggesting that low concentration (5 and 10%) of PEG6000 treatment was preferable for the expression of the four crucial enzyme genes. To further explore the gene function, these two concentrations of PEG6000 can be selected to treat samples to increase gene expression.

3.3. Correlation analysis

Pearson correlation method was used to analyze the association between gene expression and flavonoid accumulation by OriginPro 2021 (Figure 4). The findings demonstrated that the amounts of the four flavonoid components were highly positively connected, which indicated that the accumulation patterns of the four flavonoid components were similar under the treatment of PEG6000.

The expression of the *CHS* in roots was positively correlated with the content of baicalein, which indicated that the *CHS* could promote

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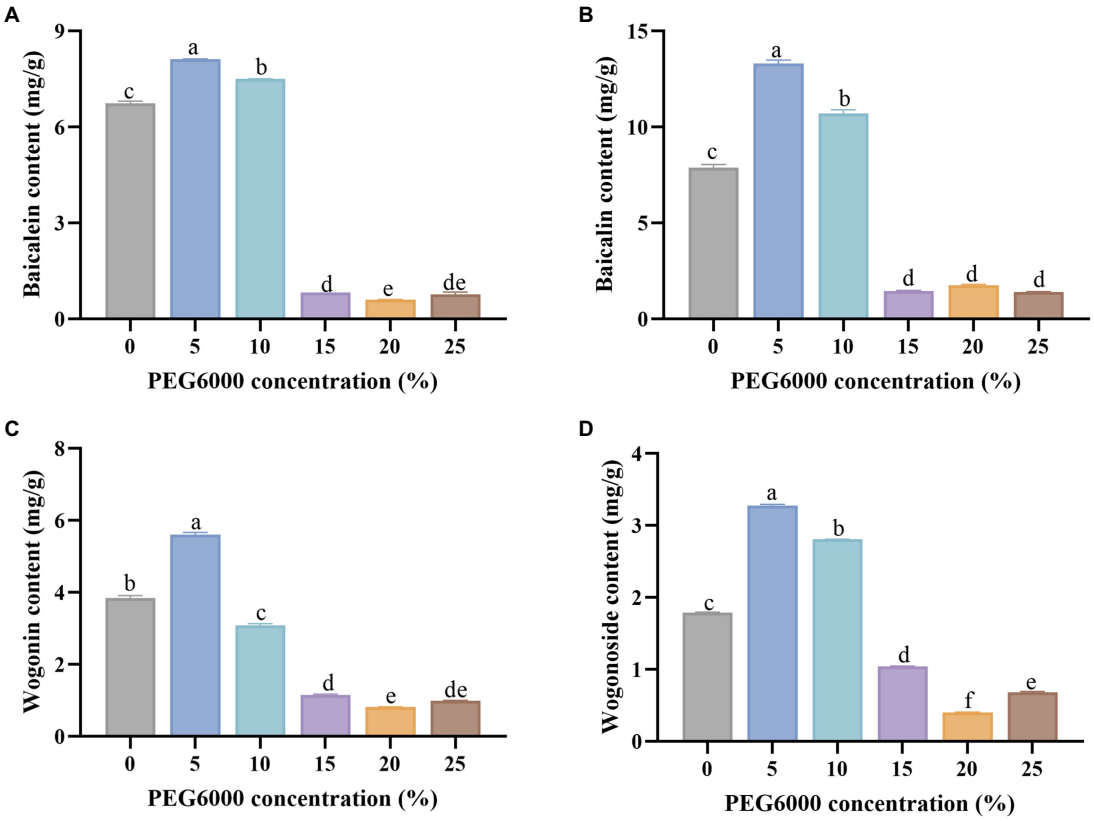

FIGURE 2
The contents of baicalein **(A)**, baicalin **(B)**, wogonin **(C)**, and wogonoside **(D)** under different concentrations of PEG6000 solution treatment. Different letters indicate significant differences by t-test between different treatment (p<0.05).

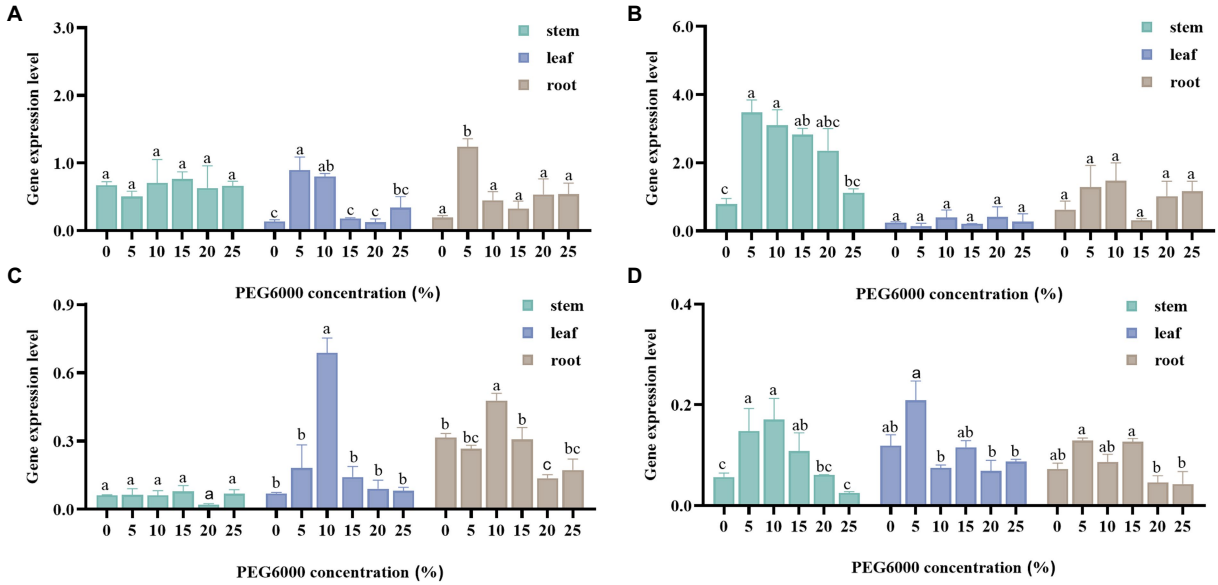

FIGURE 3
The results of the expression levels of *PAL1* **(A)**, *PAL2* **(B)**, *CHS* **(C)**, and *UBGAT* **(D)** in stems, leaves, and roots under drought stress. Different letters indicate significant differences by t-test between different treatment (p<0.05).

the accumulation of baicalein. The expression of *CHS* and *PAL1* in roots was positively correlated with baicalin content, which indicated that these genes played a critical role in regulating the accumulation of baicalin content. Correlation analysis showed that *PAL1* and *UBGAT* in roots were also essential regulators of wogonin accumulation. Moreover, *PAL1*, *CHS*, and *UBGAT* in roots had

TABLE 3 Significance results for tests of between-subjects effects.

	PEG6000				Organs			
	CHS	*PAL1*	*PAL2*	*UBGAT*	*CHS*	*PAL1*	*PAL2*	*UBGAT*
sig	0	0.002	0.005	0	0	0.038	0	0.1

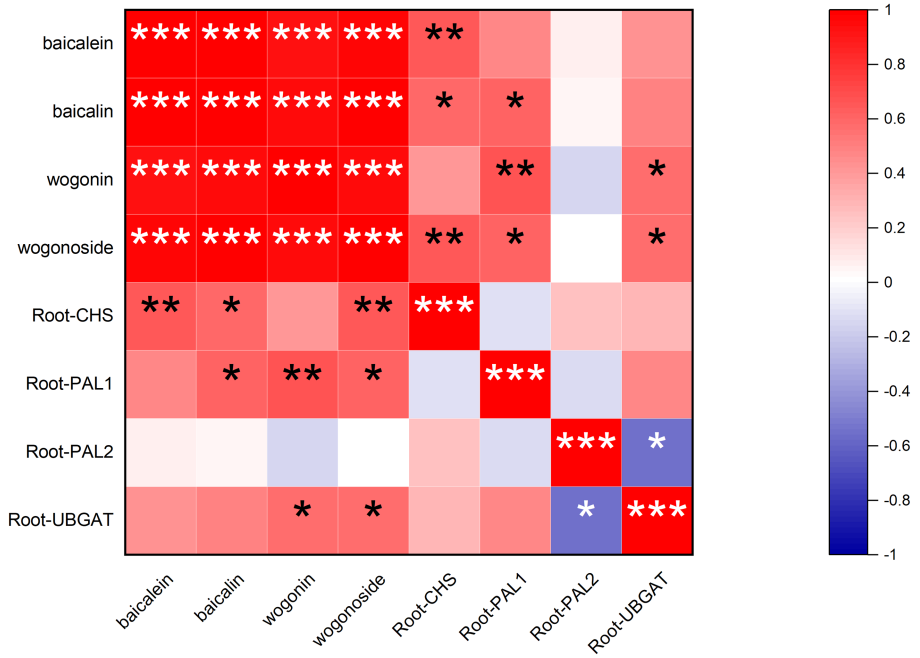

FIGURE 4
The results of correlation analysis between the indicators in this study. The correlation type is Pearson. *p<0.05, **p<0.01, and ***p<0.001.

significant effects on the accumulation of wogonoside. The most crucial enzyme gene expression positively impacted the amount of flavonoid. Only *PAL2* and wogonin have a negative correlation. It was speculated that *PAL2* might inhibit the accumulation of wogonin.

4. Discussion

Plants respond physiologically to environmental changes to maintain their individual or reproductive fitness. Flavonoid is a very significant class of secondary metabolites. Their synthesis and accumulation are related to the growing environment of plants, and they are involved in the process of plant resistance (Rodriguez-Calzada et al., 2019). They are essential protective substances for plants against the adverse external environment. Secondary metabolism is crucial for coping with environmental changes and maintaining plant homeostasis. Compared with primary metabolism, it is more susceptible to environmental changes (Yang et al., 2020). Previous studies have confirmed drought stress influenced flavonoid accumulation in *S. baicalensis*. The effects of PEG6000 stress on the accumulation of baicalin and baicalein in the suspension system of *S. baicalensis* were studied (Yang et al., 2011). Cheng et al., showed the accumulation of baicalin in the root of *S. baicalensis* under drought stress through controlled irrigation (Cheng et al., 2018). However, these studies only focused on the content of baicalin and baicalein. Wang et al., studied the PEG effect on the contents of baicalin, wogonoside, baicalein, and wogonin, while the object was suspension cells (Wang et al., 2020). Therefore, the response of four root-specific flavones in seedlings to drought stress was studied in this study. The results showed that proper drought stress could promote the accumulation of flavonoids in *S. baicalensis*. However, when the concentration of PEG6000 was increased, the content of flavonoids in *S. baicalensis* decreased significantly (Figure 2). This could be due to extreme drought reducing the plant's metabolic capacity. The best concentration for accumulating root-specific flavones out of all those tested was 5%.

Many interconnected metabolic branches constitute the biosynthesis pathway of plant flavonoids, which involves multiple enzyme genes whose expression is highly dependent on tissue type and responses to internal or external signals (Sun et al., 2022). The synthesis and accumulation of flavonoids depend on crucial enzymes in its synthesis pathway (Ahmad et al., 2019). These enzymes are usually located at the bifurcation of the metabolic branch in the plant secondary metabolites synthesis pathway and can further determine the metabolic flow direction (Zhao et al., 2018; Wang S. et al., 2019). In this study, PEG6000 was used to treat *S. baicalensis* seedlings to explore how drought stress affected the related gene expression. The data showed that crucial enzyme genes in the flavonoid biosynthesis pathway responded to abiotic stress differently. The effects of progressive drought stress on the expression patterns of key enzymes

upstream of baicalin biosynthesis were investigated during the vegetative period in 2-year-old *S. baicalensis*. And it was found that appropriate drought treatment resulted in increased gene expression, and over-treatment caused decreased expression (Cheng et al., 2018). This result is similar to our results, but it does not study the downstream key enzyme genes. Yang et al., studied the effects of PEG6000 stress on the expression of *PAL*, *CHS,* and *UBGAT* genes in the *S. baicalensis* suspension system. It was found that 10% PEG treatment of suspension cells significantly increased the expression of *PAL* and 20% PEG treatment significantly increased the expression of *UBGAT*, while there was no significant change in the expression of *CHS*, which was different from our results (Yang et al., 2011). In this paper, seedlings of *S. baicalensis* were used for the study. The newly discovered genes *PAL1*, *PAL2*, and *UBGAT* of *S. baicalensis* were reported. And the effect of drought stress on the downstream gene *UBGAT* in *S.baicalensis* seedlings was studied for the first time. The highest expression levels of four genes in roots were found at 5 and 10%, respectively, which indicated that these concentrations were more favorable for gene expression (Figure 3). These findings shed light on variations in the secondary metabolism of medicinal plants.

The biosynthesis of plant secondary metabolites is regulated by the spatial and temporal expression of biosynthesis genes (Lan et al., 2016). LSD results revealed significant differences in the relative expression levels of the *PAL2* and *CHS* genes among organs. Expression results displayed that the expression level of *PAL2* showed the highest gene expression level in stems, followed by the root and, finally, the leaf (Figures 3B,C). It may be because *PAL* is closely related not only to flavonoid synthesis but also to lignin formation. And the lignification of stems and roots was higher than that of leaves (Li et al., 2019). The expression of *CHS* in roots was the highest (Figure 3C). Generally, flavonoid biosynthesis genes were more strongly expressed in roots with higher flavonoid concentrations. As the medicinal organs, the taproots of *Polygonum minus* had a higher expression level of *CHS* (Roslan et al., 2012). The only organs in purple-fleshed sweet potato for anthocyanin biosynthesis are the purple tuberous roots, where *CHS* expression levels are significantly higher than in any other organ (Mano et al., 2007). However, the relative expression of the *UBGAT* gene differed significantly only between leaf and root, and the relative expression of the *PAL1* gene only differed significantly between stem and leaf (Figures 3A,D). It means that the expression of these two genes was not organ-specific. PAL is the first rate-limiting enzyme in the metabolism of phenylpropanes. CHS is the first rate-limiting enzyme in flavonoid synthesis. Both catalyze the rate-limiting step and affect the speed and direction of the metabolic pathway. And UBGAT is responsible for synthesizing aglycone into glycoside. Therefore, this study selected these four genes as representatives to study the effects of drought stress. Moreover, there are other key enzymes (e.g. CLL, CHI, F8H, OMT, and FNSII) in the flavonoid biosynthesis pathway (Lu et al., 2022). Among them, *CHI* and *OMT* have been proven to be able to respond to drought stress in other species (Castellarin et al., 2007; Gharibi et al., 2019). Subsequent experiments may be able to carry out more extensive research on the genes related to flavonoid biosynthesis in *S. baicalensis*.

There is some previous information on the effect of drought on the flavonoids or genes of *S. baicalensis*. However, up to now, there is no study on the correlation between the four root-specific flavones and crucial gene expression in *S. baicalensis* seedlings under drought stress. Therefore, we studied the correlation for the first time and found that there was a positive correlation between gene expression and flavonoid content, except for the negative correlation between *PAL2* and wogonin. And the correlation between the expression of *CHS*, *PAL1*, and *UBGAT* and flavonoid content was more significant (Figure 4). Therefore, it is suggested that *CHS*, *PAL1*, and *UBGAT* made a greater contribution to the drought-promoted flavonoid biosynthesis in *S. baicalensis* seedlings. These results showed that the increase in flavonoid content is related to the expression of the enzymes. Proving the relationship between flavonoids and gene expression may provide a theoretical basis for the gene function research. Given that 5% is the most suitable concentration for flavonoid accumulation and gene expression, subsequent experiments can use this as the optimal concentration to treat samples, and in-depth screen and verify gene function by multiple methods including comparing transcriptome and genetic engineering.

Altogether, this study found that drought stress had a significant effect on the accumulation of four flavonoids as well as the expression of four related genes in the medicinal and edible plant *S. baicalensis*. These findings deepen the understanding of the drought stress regulating function in *S. baicalensis,* which regulates the accumulation of flavonoids and gene expression. Overall, this research establishes a theoretical foundation for further exploration into the molecular mechanism of *S. baicalensis* in response to abiotic stresses, and it provides new insights into the metabolic regulation mechanism of *S. baicalensis* flavonoids.

Data availability statement

The datasets presented in this study can be found in online repositories. The names of the repository/repositories and accession number(s) can be found at: https://www.ncbi.nlm.nih.gov/genbank/, HQ847728, AB008748, OP018668, OP018669, and OP018670.

Author contributions

PL contributed to the methodology and writing of the original draft. DJ and CL contributed to the project administration, supervision, and draft review. GR, FW, and JC were responsible for resource collection. All authors contributed to the article and approved the submitted version.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Supplementary material

The Supplementary material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fevo.2023.1113823/full#supplementary-material

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