

Comprehensive genome-wide identification and expression profiles of SAUR family genes in *Fagopyrum tataricum*

Yi Yuan

Wuhan Polytechnic University

Huanhuan Qi

Wuhan Polytechnic University

Juan Zou

Wuhan Polytechnic University

Zihao Gui

Wuhan Polytechnic University

Yun Li

Wuhan Polytechnic University

Qihang Zu

Wuhan Polytechnic University

Yingying Gao

Wuhan Polytechnic University

Chenglu Feng

Wuhan Polytechnic University

Guoqing Dong

Wuhan Polytechnic University

Dengxiang Du

`ddx@whpu.edu.cn`

Wuhan Polytechnic University

Research Article

Keywords: Small Auxin Up-regulated RNAs, Gene family, Tartary buckwheat, Expression analysis

Posted Date: November 5th, 2024

DOI: <https://doi.org/10.21203/rs.3.rs-5261034/v1>

License:   This work is licensed under a Creative Commons Attribution 4.0 International License.

[Read Full License](#)

Additional Declarations: No competing interests reported.

Abstract

Background

Plant development and environmental adaptation are intricately regulated by hormones, with auxins playing a pivotal role in these processes. Within the auxin response family, the Small Auxin Up-regulated RNAs (SAURs) family holds a significant position in controlling plant development and responding to abiotic stress. However, investigations into this family in tartary buckwheat (*Fagopyrum tataricum*) are still lacking. The whole genome sequence of buckwheat offers a precious opportunity for comprehensive research on the genome-wide expression profile of the SAURs family.

Results

Based on the SAUR domain, 95 *FtSAURs* genes were identified in buckwheat. Additionally, the structure, evolution, and expression patterns of *FtSAURs* members were also analyzed. The *FtSAURs* genes exhibit diverse expression profiles across various developmental tissues and stages, revealing 27 tissue-specific expression genes and 35 hormone-responsive genes. Furthermore, under drought and salt stress conditions, 19 and 13 genes with current changes were detected, respectively. Notably, genes like *FtSAUR30* have undergone significant changes after treatment, emerging as potential candidates for stress response.

Conclusions

Our research lays a valuable foundation for further analysis of the abiotic stress resistance mechanism in buckwheat.

Introduction

As a pivotal plant phytohormone, auxin assumes a critical role not only in governing plant growth and development but also in orchestrating responses to biotic and abiotic stress [1]. Auxin executes its biological functions primarily through specific biosynthetic, homeostatic, transport, and signal transduction pathways. The entire process encompasses six key gene families in auxin signaling pathways: small auxin upregulated RNA (SAUR), auxin/indoleacetic acid (Aux/IAA), auxin amide synthase gene family (Gretchenhagen-3, GH3), Auxin input carrier (AUX1), transcriptional inhibitor response 1 (TIR1), and auxin response factors (ARF) gene family [2, 3]. Among them, the SAUR gene family is the most widely distributed auxin-responsive gene family in plants, capable of responding to early auxin stimulus [4].

In higher plants, most SAURs genes possess only one exon and lack introns. The promoters of most SAURs genes encompass one or more AuxREs, facilitating rapid transcription and translation of SAURs in response to auxin sensing. The protein encoded by the SAURs gene features a SAUR-specific domain (SSD) comprising approximately 60 amino acids [5], which includes conserved I-IV motifs. The N-terminal

or C-terminal histidine-rich regions of SAURs aid in their binding to metal ions. Additionally, some SAURs possess calcium binding sites for calmodulin interaction. The SAUR gene plays a pivotal role in regulating plant development, encompassing hormone biosynthesis and transport, light response, leaf growth and senescence, as well as root growth and development [6, 7]. Since their identification in soybean hypocotyls through subtractive hybridization in 1987, SAURs have been continuously discovered across various species [8]. Through bioinformatics technology, researchers have identified 81 SAURs in *Arabidopsis thaliana* [9], 99 in tomato [10], 134 in potato [11], 73 in cucumber [12], 91 in maize [13], and 68 in wax gourd [5].

The study of SAUR genes in model plants has revealed some functions and mechanisms of SAUR genes [7]. The SAUR genes are pivotal in regulating plant development, encompassing hormone biosynthesis and transport, light responsiveness, leaf growth and senescence, as well as root growth and development [14, 15, 16]. In *Arabidopsis*, overexpression of SAUR genes, such as *AtSAUR19*, *AtSAUR36*, *AtSAUR53* and *AtSAUR63* can induce cell and hypocotyl elongation [17–20]. Furthermore, SAUR genes exhibit responsiveness to diverse abiotic stresses. *AtSAUR41* has been demonstrated to enhance plant resistance to conditions such as drought and salt stress [21].

Tartary buckwheat (*Fagopyrum tataricum*), a gluten-free crop, serves not only an energy source but also a healthy source of bioactive compounds [22]. Tartary buckwheat boasts abundant flavonoids and polyphenols [23], which possess anti-inflammatory, anti-diabetic, anti-hypertensive, and anti-cancer activities [24], hence it is referred to as a functional food [25, 26]. The SAUR family is widely identified across various species, yet there is limited information on SAUR genes in Tartary buckwheat. Leveraging the published reference genome, we conducted a systematic analysis of SAUR genes. This study comprehensively analyzed tartary buckwheat SAUR genes on a genome-wide scale, performing systematic phylogenetic and evolutionary analyses, and exploring their expression patterns in different tissues and under diverse stress conditions. The findings of this study offer valuable insights for further exploration into the potential functions of SAURs in tartary buckwheat.

Materials and methods

Identification SAUR genes from buckwheat genome

Arabidopsis SAUR cascade proteins from <https://www.arabidopsis.org/> were used as queries to search for SAUR proteins in the tartary buckwheat Molecular Breeding Knowledgebase (MBKbase) (<http://mbkbase.org/Pinku1/>). BLASTp program was used to identify candidate target proteins, with a score value ≥ 100 and e-value $\leq 1e - 10$. We employed two HMMER3.0 searches (default parameter) with a cutoff of 0.01 (<http://plants.ensembl.org/hmmer/index.html>) [27] and SMART (<http://smart.embl-heidelberg.de/>) to determine the presence of the SAUR domain [28]. Sequence length, molecular weight, isoelectric point (pI), and subcellular localization predictions for FtSAUR proteins were acquired using tools available on the ExPASy website (https://web.expasy.org/compute_pi/) [29].

Chromosomal distribution and gene duplication

Based on the physical location information of the genome database, all *FtSAUR* genes were mapped to the buckwheat genome [30]. Multiple collinear scanning toolkits (MCScanX) were used to analyze segmental replication events of *FtSAUR* genes based on default parameters [31]. The *SAUR* gene homology between genes was assessed using the Dual Syntony plotter (<https://github.com/CJ-Chen/TBtools>).

Gene structure analysis of FtSAURs

Based on CDS length and corresponding full-length sequence, exon-intron structure of all *FtSAUR* genes was analyzed using the Gene Structure Display Server (GSDS: <http://gsds.cbi.pku.edu.cn>). To understand the structural differences among *FtSAUR* genes, the conserved motifs in *FtSAUR* proteins were further observed [32]. The MEME program (<http://meme.nbcr.net/meme/intro.html>) was used to analyze protein sequences, with the following parameters: the optimal motif width is 6 ~ 200, and the maximum motif number is 10 [33].

Phylogenetic analysis of FtSAURs

Phylogenetic analysis was conducted using *FtSAUR* proteins from buckwheat, arabidopsis, and rice. Phylogenetic trees were built using the NJ method of MEGA 7.0 via Geneious R11, with the following parameters: Jukes-Cantor model, global alignment with free end gaps, and a bootstrap value of 1000. For phylogenetic analysis of *FtMAPK* proteins, full-length amino acid sequences were utilized [34, 35].

Plant Materials and growth conditions

Buckwheat plants were cultivated in the greenhouse at Wuhan Polytechnic University, potted in a mixture of soil and vermiculite (1:1) and growth a 12 h/26°C day and 12 h/22°C night regime, with a relative humidity of 80%. The stems, roots, leaves, flowers and seeds were individually collected from five plants with good growth and similar growth conditions, rapidly frozen in liquid nitrogen, and stored at -80°C for further use.

Stress response experiments, biochemical index identification, and gene expression analysis were conducted when plants reached the 7-leaf stage [36]. Control materials underwent standard management materials, while treated materials were subjected to drought treatment for 7 and 14 days with watering withheld, respectively. Additionally, the buckwheat seedlings of comparable growth status were treated with 100 mmol·L⁻¹ NaCl. Samples were collected at 7 day and 14 day post-treatment.

Expression analysis of FtSAURs in different tissues

Total RNA from plant tissues was extracted using the total RNA extraction kit (Sangon, Shanghai, China, SK1321) and treated with RNase-free DNase-I to remove genomic DNA [37]. The cDNA synthesis was preformed with a 1 mg RNA sample using RNAprep Pure Polysaccharide Polyphenol Plant Total RNA

Extraction Kit (DP441) and FastKing one-step reverse transcription fluorescence quantitative kit (probe method) (FP314). Real-time quantitative PCR (qRT-PCR) was performed in real-time fluorescent quantitative PCR 7500 (ThermoFisher Scientific). Primer sequences for the selected genes were designed using Primer 5.0 and repeated at least three times (Additional document 1: Table S1). The *FtH3* gene is selected as an internal control, due to its stable expression across various tissues and growth stages [38]. Data analysis was performed using the $2^{-(\Delta\Delta CT)}$ method to determine expression correlation [39]. The qRT-PCR reactions were performed in a total volume of 20 μ L, including 2 μ L diluted cDNA, 1 μ L each forward and reverse primer, 10 μ L SYBR Premix Ex Taq and 6 μ L ddH₂O. The qPCR program consisted of an initial denaturation step at 95°C for 3 min, followed by 30 cycles of 95°C for 15 s, 60°C for 30 s, and 72°C for 20 s.

Data Availability Statement

The experiments were performed with three biological replicates, and statistical significance was determined using a Two-way ANOVA and Tukey's test. Statistical significance was denoted as *** $P \leq 0.001$, ** $P \leq 0.01$, and * $P \leq 0.05$.

Results

Identification and annotation of the SAUR genes in tartary buckwheat

Based on homology alignment and SAUR domain screening, 95 SAUR genes were identified, and the 95 *FtSAUR* genes were renamed based on chromosomal localization. Subsequently, the basic characteristics of the proteins were analyzed, including protein length, molecular weight, isoelectric point (pI), and predicted subcellular localization (Table 1). FtSAUR04 is the smallest protein, consisting of 63 amino acids, while FtSAUR94 is the largest, consisting of 193 amino acids. Most proteins fall within the range of 90 to 150 amino acids, aligning with the characteristics of small molecular proteins. The molecular weight range varies from 7.21 kDa (FtSAUR04) to 21.37 kDa (FtSAUR94), and the pI range spans from 3.7 (FtSAUR40) to 11.63 (FtSAUR02). Subcellular localization prediction indicated that 9 proteins (9.47%) were located in chloroplasts, 6 (6.32%) in the cytoplasm, 6 (6.32%) in the extracellular space, 39 (41.05%) in mitochondria, 33 (34.74%) in the nucleus, and 2 (2.11%) were associated with the plasma membrane. The subcellular localization pattern of FtSAURs aligns with their putative functions, particularly in glycosylation.

Table 1
Information on the SAUR gene family

Gene name	Gene ID	Chromosome	aa	MV(Da)	pI	Subcellular
FtSAUR01	FtPinG0000271800.01	Chr1:4319909–4320367	152	17465.23	11.49	Mitochondrial
FtSAUR02	FtPinG0001397900.01	Chr1:5938488–5938955	96	10620.22	7.73	Mitochondrial
FtSAUR03	FtPinG0001398200.01	Chr1:5945483–5945863	106	11847.64	5.7	Nuclear
FtSAUR04	FtPinG0002465500.01	Chr1:7738125–7738358	101	10895.78	10.23	Chloroplast
FtSAUR05	FtPinG0002434500.01	Chr1:8736825–8737106	172	19400.13	8.93	Nuclear
FtSAUR06	FtPinG0003159200.01	Chr1:10808656–10809039	94	10724.25	7.73	Mitochondrial
FtSAUR07	FtPinG0003159000.01	Chr1:10820442–10820770	90	10209.72	5.8	Nuclear
FtSAUR08	FtPinG0003158800.01	Chr1:10827430–10828206	90	10332.89	8.71	Nuclear
FtSAUR09	FtPinG0001621800.01	Chr1:16691991–16692335	76	8771.18	6.33	Extracellular
FtSAUR10	FtPinG0001621900.01	Chr1:16734931–16735266	126	14221.3	7.94	Mitochondrial
FtSAUR11	FtPinG0001622200.01	Chr1:16747322–16747618	128	14615.97	10.36	Mitochondrial
FtSAUR12	FtPinG0001622600.01	Chr1:16755385–16755735	104	11800.57	8.22	Mitochondrial
FtSAUR13	FtPinG0001623000.01	Chr1:16763697–16764020	102	11496.34	9.86	Mitochondrial
FtSAUR14	FtPinG0001623200.01	Chr1:16788906–16789249	101	11573.37	7.82	Mitochondrial
FtSAUR15	FtPinG0008613300.01	Chr1:21949980–21950420	142	15952.41	8.49	Mitochondrial
FtSAUR16	FtPinG0008443500.01	Chr1:34124964–34125407	112	12978.93	8.35	Nuclear
FtSAUR17	FtPinG0008444800.01	Chr1:34173905–34174276	140	15605.13	7.85	Mitochondrial
MW Molecular weight, pI isoelectric point						

Gene name	Gene ID	Chromosome	aa	MV(Da)	pI	Subcellular
FtSAUR18	FtPinG0002423800.01	Chr1:36549600–36549911	127	14801.85	8.68	Nuclear
FtSAUR19	FtPinG0002423200.01	Chr1:36560726–36561040	105	12179.77	7.87	Nuclear
FtSAUR20	FtPinG0007017300.01	Chr2:12571228–12571650	119	13310.73	6.79	Nuclear
FtSAUR21	FtPinG0000807600.01	Chr2:51809464–51809934	105	11718.68	9.66	Nuclear
FtSAUR22	FtPinG0003901600.01	Chr2:58124801–58125211	106	12260.02	8.22	Mitochondrial
FtSAUR23	FtPinG0003904400.01	Chr2:58200932–58201243	134	14901.26	9.94	Nuclear
FtSAUR24	FtPinG0003215600.01	Chr3:620663–621031	88	9936.42	5.56	Mitochondrial
FtSAUR25	FtPinG0000909000.01	Chr3:28153246–28153856	99	11179.16	10.42	Mitochondrial
FtSAUR26	FtPinG0000908700.01	Chr3:28154646–28155200	103	12033.77	7.52	Nuclear
FtSAUR27	FtPinG0008563700.01	Chr3:31652219–31652776	113	12854.1	9.54	Mitochondrial
FtSAUR28	FtPinG0001945300.01	Chr3:43645564–43645994	142	15835.12	10.71	Mitochondrial
FtSAUR29	FtPinG0001096500.01	Chr3:45011893–45012240	99	11390.1	9.69	Nuclear
FtSAUR30	FtPinG0004378100.01	Chr3:46919436–46919777	111	12427.37	6.67	Cytoplasmic
FtSAUR31	FtPinG0007969000.01	Chr3:49165657–49166013	130	14662.01	9.09	Nuclear
FtSAUR32	FtPinG0007968800.01	Chr3:49167041–49167313	138	15622.96	10.81	Nuclear
FtSAUR33	FtPinG0007968400.01	Chr3:49169386–49169739	114	13116.2	8.17	Nuclear
FtSAUR34	FtPinG0007968100.01	Chr3:49171564–49171896	114	12900.73	7.41	Nuclear
FtSAUR35	FtPinG0007967400.01	Chr3:49198833–49199177	116	13140.26	10.02	Nuclear
MW Molecular weight, pI isoelectric point						

Gene name	Gene ID	Chromosome	aa	MV(Da)	pI	Subcellular
FtSAUR36	FtPinG0007966400.01	Chr3:49220995–49221385	112	13061.83	6	Nuclear
FtSAUR37	FtPinG0007965900.01	Chr3:49238868–49239656	133	14759.95	10.19	Mitochondrial
FtSAUR38	FtPinG0002403100.01	Chr3:52348024–52348578	97	10992.79	9.8	Mitochondrial
FtSAUR39	FtPinG0002363800.01	Chr3:53972403–53972817	122	14061.46	7.73	Extracellular
FtSAUR40	FtPinG0000419200.01	Chr3:54163764–54164180	102	11495.32	9.33	Mitochondrial
FtSAUR41	FtPinG0000418400.01	Chr3:54181401–54181826	130	14844.49	11.63	Mitochondrial
FtSAUR42	FtPinG0000764300.01	Chr3:55771482–55771973	72	8177.47	5.6	Cytoplasmic
FtSAUR43	FtPinG0000765000.01	Chr3:55788844–55789209	127	14570.71	10.25	Nuclear
FtSAUR44	FtPinG0005124800.01	Chr4:42643817–42644170	138	15210.74	9.26	Chloroplast
FtSAUR45	FtPinG0007504400.01	Chr4:47335893–47336285	146	16572.03	8.62	Mitochondrial
FtSAUR46	FtPinG0000507900.01	Chr5:1212298–1212651	63	7205.36	6.78	Cytoplasmic
FtSAUR47	FtPinG0005727300.01	Chr5:6062387–6062896	146	16753.46	10.6	Mitochondrial
FtSAUR48	FtPinG0006725000.01	Chr5:18896388–18896741	148	16383.52	8.08	Extracellular
FtSAUR49	FtPinG0006724800.01	Chr5:18901711–18902070	94	10914.56	9.85	Nuclear
FtSAUR50	FtPinG0006724600.01	Chr5:18905227–18905758	129	15023.36	10.02	Nuclear
FtSAUR51	FtPinG0006719000.01	Chr5:18905528–18905758	97	10983.66	8.9	Mitochondrial
FtSAUR52	FtPinG0006724400.01	Chr5:18905528–18905758	115	12864.71	5.15	Nuclear
FtSAUR53	FtPinG0004309500.01	Chr5:31264496–31264936	98	11260.1	8.64	Nuclear
MW Molecular weight, pI isoelectric point						

Gene name	Gene ID	Chromosome	aa	MV(Da)	pI	Subcellular
FtSAUR54	FtPinG0007044300.01	Chr6:7863392–7863814	76	9019.25	4.2	Nuclear
FtSAUR55	FtPinG0007040100.01	Chr6:8067988–8068328	176	20033.82	9	Nuclear
FtSAUR56	FtPinG0002808700.01	Chr7:2950740–2951099	102	11803.7	8.08	Cytoplasmic
FtSAUR57	FtPinG0002875800.01	Chr7:4404167–4404520	108	12688.52	6.96	Cytoplasmic
FtSAUR58	FtPinG0002594300.01	Chr7:13486491–13486886	79	8941.63	8.74	Extracellular
FtSAUR59	FtPinG0001267800.01	Chr7:23393258–23393731	98	11392.12	10.24	Mitochondrial
FtSAUR60	FtPinG0001657300.01	Chr7:41315533–41315925	133	15165.34	8.63	Mitochondrial
FtSAUR61	FtPinG0006935800.01	Chr7:42053307–42053651	117	12968.52	6.78	Nuclear
FtSAUR62	FtPinG0009163500.01	Chr7:47751991–47752425	105	11790.48	4.13	Plasma Membrane
FtSAUR63	FtPinG0009163900.01	Chr7:47758087–47758491	142	16191.67	8.95	Mitochondrial
FtSAUR64	FtPinG0009164300.01	Chr7:47760275–47760724	145	16587.12	7.46	Nuclear
FtSAUR65	FtPinG0009164500.01	Chr7:47762371–47762823	134	15037.63	8.83	Extracellular
FtSAUR66	FtPinG0009164900.01	Chr7:47767892–47768245	121	13243.24	7.34	Chloroplast
FtSAUR67	FtPinG0009165100.01	Chr7:47770960–47771406	137	15404.82	9.32	Chloroplast
FtSAUR68	FtPinG0009165400.01	Chr7:47771998–47772438	138	15760.51	9.9	Mitochondrial
FtSAUR69	FtPinG0009165500.01	Chr7:47773282–47773737	100	10952.69	4.28	Chloroplast
FtSAUR70	FtPinG0009166400.01	Chr7:47793178–47793570	71	7822.76	3.7	Nuclear
FtSAUR71	FtPinG0009166700.01	Chr7:47802394–47802827	146	16208.79	9.21	Mitochondrial
MW Molecular weight, pI isoelectric point						

Gene name	Gene ID	Chromosome	aa	MV(Da)	pI	Subcellular
FtSAUR72	FtPinG0009166900.01	Chr7:47804059–47804490	142	16038.62	9.04	Mitochondrial
FtSAUR73	FtPinG0009167300.01	Chr7:47811077–47811469	138	15343.88	9.04	Chloroplast
FtSAUR74	FtPinG0009167500.01	Chr7:47812689–47812904	137	15356.79	9.17	Mitochondrial
FtSAUR75	FtPinG0009167700.01	Chr7:47814939–47843960	138	15366.99	9.03	Mitochondrial
FtSAUR76	FtPinG0009167900.01	Chr7:47816675–47817283	143	15709.29	10.23	Mitochondrial
FtSAUR77	FtPinG0009168600.01	Chr7:47833424–47833840	138	15431.24	9.85	Mitochondrial
FtSAUR78	FtPinG0009169000.01	Chr7:47837913–47838329	138	15528.1	9.26	Mitochondrial
FtSAUR79	FtPinG0009169200.01	Chr7:47845140–47845589	142	15999.56	8.74	Mitochondrial
FtSAUR80	FtPinG0009169300.01	Chr7:47847226–47847666	139	15636.27	9.03	Mitochondrial
FtSAUR81	FtPinG0009169600.01	Chr7:47848745–47849199	138	15484.94	9.93	Mitochondrial
FtSAUR82	FtPinG0009169800.01	Chr7:47861547–47862035	144	15973.68	8.96	Chloroplast
FtSAUR83	FtPinG0009170000.01	Chr7:47863229–47863699	142	15926.59	9.01	Mitochondrial
FtSAUR84	FtPinG0009170400.01	Chr7:47870394–47870858	146	16300.96	9.03	Mitochondrial
FtSAUR85	FtPinG0009170500.01	Chr7:47871718–47872161	138	15437.88	9.32	Chloroplast
FtSAUR86	FtPinG0009171400.01	Chr7:47887474–47887926	180	20164.8	5.05	Cytoplasmic
FtSAUR87	FtPinG0009171800.01	Chr7:47892148–47892651	150	16996.31	8.86	Nuclear
FtSAUR88	FtPinG0009172200.01	Chr7:47898974–47924004	95	10676.26	8.23	Mitochondrial
FtSAUR89	FtPinG0009172300.01	Chr7:47900035–47900523	101	11192.85	9.17	Chloroplast
MW Molecular weight, pI isoelectric point						

Gene name	Gene ID	Chromosome	aa	MV(Da)	pI	Subcellular
FtSAUR90	FtPinG0003496600.01	Chr7:49828637–49829877	110	12648.49	7.87	Nuclear
FtSAUR91	FtPinG0009367800.01	Chr8:4752893–4753435	111	12604.5	10.65	Nuclear
FtSAUR92	FtPinG0009367500.01	Chr8:4764471–4764859	193	21368.5	9.35	Nuclear
FtSAUR93	FtPinG0007172700.01	Chr8:5909869–5910312	76	9019.25	4.2	Nuclear
FtSAUR94	FtPinG0008667300.01	Chr8:17082885–17083514	142	16003.44	8.48	Extracellular
FtSAUR95	FtPinG0004797700.01	Chr8:20214959–20215318	138	15284.89	8.48	Plasma Membrane
MW Molecular weight, pI isoelectric point						

Distribution and duplication of FtSAURs

After analyzing the chromosomal localization, we discovered that approximately 76.84% (73 out of 95) of *FtSAURs* were clustered on 8 chromosomes, exhibiting a non-random distribution trend. Furthermore, each type of *FtSAUR* demonstrated an irregular distribution, with most genes on the same chromosome belonging to different types, indicating complementary functions among them. Scattered, tandem, and whole genome duplication (WGD)/segmental duplications were observed in the *FtSAUR* genes of buckwheat, suggesting that tandem duplication events may be concentrated in certain chromosomal regions. Gene duplication is considered a significant driving factor in the evolution of genomes and genetic systems. On the other hand, segmental duplication involves the duplication of multiple genes through polyploid events, often accompanied by chromosomal rearrangements.

To investigate the conservation of *FtSAURs* in buckwheat, we identified collinear gene pairs within *FtSAURs*. As shown in Fig. 1, 17 pairs of collinear *FtSAURs* were identified. Additionally, the first chromosome contains the highest number of genes, which are associated with genes on the second, third, seventh, and eighth chromosomes. Furthermore, many genes on chromosome 7 form collinear pairs with genes on chromosomes 1, 2, 3, 6, and 8, all belonging to the same category. These collinear pairs highlight the close relationship between genes in this family and emphasize strict conservation. Interestingly, no collinear genes were found on chromosome 5, indicating that their origins were independent.

Phylogenetic analysis of FtSAURs

We constructed a phylogenetic tree using the neighbor-joining (NJ) method with a bootstrap value of 1000, based on the amino acid sequences of the 95 identified *FtSAUR*, 79 *AtSAUR* and 56 *OsSAUR* proteins. 230 *SAUR* proteins in the phylogenetic tree were divided into 15 branches (families), each with greater than 40% sequence identity (Fig. 2). Of these families, 11 were shared by buckwheat, Arabidopsis and rice; 3

were shared by buckwheat and Arabidopsis; and one was unique to Arabidopsis. The number of FtSAURs in each family varied, with the largest containing 63 SAUR members and the smallest consisting of only two members. The distribution of genes in the families was as follows: 8, 33, 19, 6, 18, 7, 4, 6, 38, 9 and 15 genes, respectively. The phylogenetic tree with buckwheat, arabidopsis and rice revealed close grouping of some *FtSAURs* with *AtSAURs*, suggesting functional similarities among these proteins.

Gene structure, Conserved motifs and cis-elements analysis of FtUGTs

During the evolution of multigene families, newly evolved copies often acquire new gene functions, leading to diversification of gene structures in the FtSAUR gene family. To further gain insights into the structural diversity of *FtSAUR* genes, their exon and intron structures were further analyzed based on genome location (Fig. 3). With the exception of *FtSAUR15*, *FtSAUR55*, *FtSAUR70*, *FtSAUR71*, *FtSAUR75*, *FtSAUR87* and *FtSAUR90*, all of which possessed only one exon, consistent with the characteristics features of the gene family. These results indicate that most members of the SAUR superfamily in buckwheat possess simple gene structures with less than one intron, likely contributing to their high activity and rapid transcription. Moreover, gene structure and intron composition were generally conserved within the same FtSAUR family.

To elucidate the characteristic amino acid regions of FtSAUR proteins, motifs analysis were conducted using the MEME online software, identifying ten conserved motifs designated as motif 1–10 (Fig. 4). Among these motifs, motif 4 exhibited the highest representation with 81 members, followed by motif 6 (65 members) and motif 2 (57 members). Conversely, motifs 9 and 10 were less prevalent, containing 9 and 10 members, respectively. Motifs 1, 3, 5, 7, and 8 comprised 29, 26, 27, 26, and 12 members, respectively. Notably, certain motifs were consistently positioned in the gene structure, with motifs 3, 4 and 8 predominantly located at the protein's beginning, whereas motifs 6, 7 and 10 were typically found towards the end. Moreover, gene clusters with tandem repeated exhibited similar structural features, as exemplified by *FtSAUR75* - *FtSAUR89*, displaying comparable gene lengths, structures, coding sequence, and amino acid motifs.

Promoter *cis*-elements are tightly related to the gene function. To explore the regulatory mechanisms of *FtSAURs*, the *cis*-elements in the promoter regions (2000 bp) were further investigated (Fig. 5). A total of 120 *cis*-regulatory elements were identified and categorized into eight groups: light-responsive, development-related, site-binding-related, hormone-responsive, promoter-related, environmental stress-related, and other elements. Promoter-related elements such as CAAT-box and TATA-box, light-response elements including TCT motif, GT1 motif, Box 4, G-box and I-box, as well as drought-responsive elements MYB and MYC, were ubiquitously present across *FtSAURs*. Additionally, FtSAURs contained 13 hormone-responsive elements, encompassing various plant hormones such as auxin (AuxRR-core, TGA-element, AuxRE, TGA-box), gibberellin (P-box, GARE-motif, P-box, TATC-box), abscisic acid (ABRE), jasmonic acid (TGACG-motif, CGTCA-motif), and salicylic acid (TCA-element, SARE).

All *FtSAURs* were found to contain hormone-responsive elements. Specifically, 95 *FtSAUR* members contained IAA-responsive elements, 90 contained GA-responsive elements, 93 contained ABA-responsive elements, 93 contained JA-responsive elements, and 89 contained SA-responsive elements. Interestingly, 76 *FtSAUR* members exhibited all five hormone-responsive elements. Moreover, environmental stress-related *cis*-elements were identified, including drought, salt, low-temperature, anaerobic and wound. These findings collectively suggest the involvement of *FtSAURs* in the regulating plant growth and development, light response, hormone signaling and adaption to abiotic stress in buckwheat.

Expression patterns of FtSAURs in various tissues

To further investigate the potential biological functions of *FtSAURs*, we investigated expression patterns in various tissues of buckwheat using qRT-PCR (Fig. 6). Among the 95 genes analysis, 24 were not detected in normal tissues, while 17 showed tissue-specific expression. Specifically, *FtSAUR26*, *FtSAUR38* and *FtSAUR59* were exclusively expressed in roots, five genes (*FtSAUR27*, 0.493; *FtSAUR55*, 0.271; *FtSAUR65*, 0.203; *FtSAUR69*, 0.201; *FtSAUR76*, 0.301) in leaves, six genes (*FtSAUR09*, 0.394; *FtSAUR28*, 0.789; *FtSAUR32*, 0.996; *FtSAUR40*, 0.163; *FtSAUR45*, 0.173; *FtSAUR72*, 0.157) in flowers, and three genes (*FtSAUR11*, 1.777; *FtSAUR29*, 0.217; *FtSAUR54*, 0.356) in seeds. Furthermore, eight genes (*FtSAUR04*, *FtSAUR06*, *FtSAUR33*, *FtSAUR35*, *FtSAUR36*, *FtSAUR42*, *FtSAUR50*, *FtSAUR81*) were expressed in vegetative growth tissues, and two genes (*FtSAUR03*, *FtSAUR15*) in reproductive growth tissues. Additionally, 54 genes exhibited expression in multiple tissues, with varied expression levels (Fig. 6a). 12 genes (*FtSAUR06*, *FtSAUR07*, *FtSAUR24*, *FtSAUR42*, *FtSAUR47*, *FtSAUR50*, *FtSAUR57*, *FtSAUR74*, *FtSAUR81*, *FtSAUR91*, *FtSAUR94*, *FtSAUR94*) had the highest expression levels in roots, six (*FtSAUR04*, *FtSAUR35*, *FtSAUR48*, *FtSAUR61*, *FtSAUR79*, *FtSAUR89*) in stems, 17 (*FtSAUR05*, *FtSAUR12*, *FtSAUR17*, *FtSAUR31*, *FtSAUR33*, *FtSAUR34*, *FtSAUR36*, *FtSAUR41*, *FtSAUR43*, *FtSAUR46*, *FtSAUR63*, *FtSAUR64*, *FtSAUR66*, *FtSAUR75*, *FtSAUR86*, *FtSAUR87*, *FtSAUR88*) in leaves, eleven (*FtSAUR02*, *FtSAUR03*, *FtSAUR13*, *FtSAUR18*, *FtSAUR19*, *FtSAUR37*, *FtSAUR49*, *FtSAUR70*, *FtSAUR82*, *FtSAUR92*, *FtSAUR93*) in flowers, and eight (*FtSAUR01*, *FtSAUR08*, *FtSAUR15*, *FtSAUR16*, *FtSAUR30*, *FtSAUR39*, *FtSAUR53*, *FtSAUR83*) seeds. Although some genes showing low expression levels, the diverse expression patterns across tissues suggest regulatory roles of the *FtSAUR* family in buckwheat growth and development.

Given the significance of the SAUR family in auxin signaling, we examined the expression changes after IAA treatment. Focusing on leaf tissues, where gene expression is abundant, we evaluated the expression profiles 3 days of IAA treatment, as shown in Fig. 6b. After IAA treatment, 29 genes (e.g., *FtSAUR03*, *FtSAUR09*, *FtSAUR11*) remained undetectable in leaves. 15 genes (e.g., *FtSAUR04*, *FtSAUR10*, *FtSAUR13*) exhibited expression exclusively after treatment, with no expression in untreated materials. Conversely, Six genes (*FtSAUR18*, *FtSAUR27*, *FtSAUR35*, *FtSAUR55*, *FtSAUR63*, *FtSAUR69*) lost their expression post-IAA treatment. Notably, five genes (*FtSAUR10*, 0.275; *FtSAUR32*, 0.338; *FtSAUR45*, 0.235; *FtSAUR77*, 0.886; *FtSAUR78*, 0.221) were expressed solely on the first day of treatment, and six genes (*FtSAUR13*, 1.225; *FtSAUR26*, 1.788; *FtSAUR38*, 5.723; *FtSAUR39*, 0.478; *FtSAUR59*, 0.838; *FtSAUR94*, 0.315) were only expressed on the third day. Interestingly, a single gene (*FtSAUR04*) expressed at the last two days, while two genes (*FtSAUR40*, *FtSAUR82*) had expression during the initial two days after treatment. Additionally,

among the remaining 45 genes, diverse expression patterns were observed across various stages, with sustained up-regulation seen in two genes (*FtSAUR30*, *FtSAUR47*) post-treatment.

Expression analysis of FtSAURs after drought and salt treatment in leaves

After drought treatment, we assessed the expression of *FtSAUR* genes in leaves compared to untreated control (Fig. 7). Among 59 detected genes, 11 exhibited expression solely in the control group, with no expression after drought treatment. Three genes were expressed at 7 days of treatment, but not in control and 14 days of treatment. Four genes showed expression only at 14 days of treatment, not in control or after 7 days. Among the remaining 41 genes, nine were continuously up-regulated, while 12 were continuously downregulated. Specifically, *FtSAUR18*, *FtSAUR19*, and *FtSAUR72* exhibited no expression in control, but displayed continuous up-regulation after treatment. *FtSAUR30* was significantly upregulated (greater than 800 fold) after 7 days of treatment, maintaining this trend after 14 days. Similarly, *FtSAUR46*, *FtSAUR61*, *FtSAUR64*, and *FtSAUR91* displayed comparable expression patterns, albeit not statistically significant. Conversely, *FtSAUR03*, *FtSAUR05*, *FtSAUR12*, *FtSAUR16*, *FtSAUR27*, *FtSAUR31*, *FtSAUR41*, *FtSAUR55*, *FtSAUR70*, *FtSAUR79*, and *FtSAUR79* showed significantly down-regulated expression after drought treatment.

In the salt stress experiment, 63 genes were found to be expressed. 14 genes were not expressed in the control, but showed expression in the treatment. Among them, 6 genes were only expressed at 7 days of treatment, 5 at 14 days of treatment, and *FtSAUR13* was continuously upregulated at both stages of treatment. Genes *FtSAUR18* and *FtSAUR19* showed extremely significant upregulation at 7 days of treatment, with a slight decrease in expression at 14 days. Among the 49 genes expressed under normal conditions, four (*FtSAUR02*, *FtSAUR11*, *FtSAUR36*, *FtSAUR37*) were not expressed after treatment, while five lost expression after 7 days but regained it at 14 days, seven were not detected at 14 days of treatment. 12 genes were up-regulated after salt treatment. Genes *FtSAUR08*, *FtSAUR24*, and *FtSAUR47* were continuously up-regulated, with significant levels detected. 10 genes were significantly up-regulated at 7 days of treatment, with varying degrees of downregulation at 14 days. Among these, *FtSAUR05*, *FtSAUR07*, *FtSAUR16*, *FtSAUR30*, *FtSAUR34*, and *FtSAUR93* displayed notable changes. 22 genes showed a decreasing trend in expression after 7 days of salt treatment, with *FtSAUR03*, *FtSAUR17*, *FtSAUR27*, *FtSAUR29*, *FtSAUR41*, and *FtSAUR92* showing a significant downward trend. After 14 days, four genes showed a continuous downward trend, while 18 genes showed varying degrees of expression recovery.

Discussion

Auxins are the earliest discovered plant hormones, playing a crucial role in all aspects of plant growth. They regulate cell elongation, division, vascular tissue differentiation, organ differentiation, and the plant's response to the external environment, and are involved in the entire growth process of plant embryos and post-embryonic development [4, 40, 41]. The expression of early auxin response genes is directly regulated by ARF, which primarily comprises three families: Aux/IAAs, Gretchen hagen3s (GH3s), and small auxin upregulated RNA (SAURs). In recent years, the SAURs family has emerged as a hot scientific topic [42, 43].

Extensive functional studies have been conducted using model plants such as *Arabidopsis thaliana* and rice, significantly enhancing our understanding of this family [7, 44]. Through homologous comparison, we identified 95 members of the SAUR family. Most *FtSAURs* possess four conserved motifs within their SSDs, featuring highly conserved functional domains and comparable sequence arrangements. *FtSAURs* exhibit a straightforward structure, with open reading frames typically ranging from 180 to 540 bp in length. Notably, 88 of these genes lack introns, while 7 contain a single intron. This finding suggests minimal sequence length variation within the family genes and a relatively consistent number of exons, aligning with previous scholars' analyses of conserved motifs in 632 SAUR proteins [14, 16]. For instance, the *OsSAURs* gene in rice lacks introns, whereas only *AtSAUR11* in *Arabidopsis thaliana* and *11 PtrSAURs* in *Populus tomentosa* contain introns [6, 45, 46]. The outcomes from gene structure analysis corroborate those derived from gene sequence analysis, highlighting that the SAUR family possesses highly conserved functional domains and analogous arrangement sequences. This underscores the SAUR family's status as an excellent candidate for comparative genomics in evolutionary research. Additionally, promoter cis- and trans-element analyses uncovered numerous hormone response structures, effectively facilitating the swift transcription and translation of SAURs in plants upon sensing auxin. The identification and analysis of 95 SAUR genes within the buckwheat genome establish a valuable genetic foundation for future investigations into their respective functions.

As early as 1978, McClure and Guilfoyle established a link between SAUR and hypocotyl elongation triggered by cell enlargement [47]. Subsequent research in plants has revealed that SAUR family genes are extensively involved in development and tissue differentiation. In this study, we utilized qRT-PCR to analyze the expression levels of 95 *FtSAURs* across various organs of tartary buckwheat. Buckwheat exhibits a wealth of expression variations, notably with 17 genes displaying tissue specificity, including 3, 5, 6, and 3 genes with tissue specificity in roots, leaves, flowers, and seeds, respectively. Furthermore, 8 genes were detected in vegetative tissues, whereas 2 genes were exclusively present in reproductive organs. The diverse expression patterns of these genes suggest that *FtSAURs* may play a significant role in the growth and tissue differentiation of tartary buckwheat. Similarly, in *Arabidopsis thaliana*, genes such as *AtSAUR63*, *AtSAUR61-68*, *AtSAUR75* and *AtSAUR53* have been identified as playing a regulatory role in hypocotyl elongation [19, 20, 48]. Specifically, genes *AtSAUR63* and *AtSAUR76* exhibit an active regulatory function in petals and stamens [44, 49]. Meanwhile, genes *AtSAUR49*, *AtSAUR30*, *AtSAUR39* and *AtSAUR72* regulate the leaf senescence process [16, 50]. Additionally, *AtSAUR9* positively regulates the growth of hypocotyls in etiolated seedlings by promoting cell enlargement. Based on the existing results and the tissue-specific gene expression identified in this study, it can be inferred that SAUR family genes are extensively involved in the development and tissue differentiation of tartary buckwheat [51–53]. Identifying these tissue-specific expressed genes is advantageous for discovering potential key regulatory genes.

Research has found that some SAURs are also involved in regulating the synthesis and transport of auxin [51]. Auxins are crucial hormones for plant growth and development. In this study, we conducted an expression analysis of SAUR family genes in buckwheat following IAA treatment. Our analysis revealed 15 genes that were induced to express after IAA treatment, as well as 6 genes that were expressed under IAA

stress, indicating a significant correlation between these genes and auxin regulation. Furthermore, the diversity of other gene expression changes underscores the widespread involvement of this family in responding to auxin. Similar studies have demonstrated that in rice overexpressing *OsSAUR39* and *OsSAUR45*, the expression levels of auxin synthesis-related genes *OsYUCs* and *OsTRPs*, as well as the content of free IAA, significantly decreased [54]. Kathare et al. examined the growth status of the main roots of *Arabidopsis thaliana* plants overexpressing *AtSAUR53* under treatment with IAA and the synthetic auxins 2,4-D and NAA. Their findings suggest that *AtSAUR53* may play a negative regulatory role in auxin transport [19, 53]. Based on this speculation, SAURs, in addition to influencing the expression of auxin transport-related genes, may also regulate the polar transport of auxin by affecting calcium signaling.

In addition to development, plants also regulate cell enlargement to adapt to various changes in the external environment. For instance, when plants respond to environmental signals such as light, temperature, and gravity, they regulate cell expansion to influence the growth of seedling hypocotyls [14, 17, 52, 55]. However, research on the function of SAUR under drought and other stress conditions remains insufficient. Understanding the mechanism of SAUR genes under drought conditions holds significant importance for enhancing the drought resistance of crops. This study identified 19 genes exhibiting significant drought responses in leaves following drought treatment, among which four genes, *FtSAUR18*, *FtSAUR19*, *FtSAUR30* and *FtSAUR72*, stand out as potential candidates for subsequent analysis. Under salt stress conditions, plants encounter severe growth challenges, particularly in terms of root growth and distribution. The regulation of SAUR genes on root growth primarily encompasses two aspects: changes in root distribution and adjustments in root structure [56, 57]. The upregulation of SAUR genes under salt stress can steer root growth towards low-salt areas. For instance, in *Arabidopsis thaliana*, SAUR genes such as *SAUR19* and *SAUR30* exhibit significant upregulation, leading to a notable increase in root length. This root growth towards low-salt areas is accompanied by an increase in root hair density and a corresponding extension in root hair length, thereby enhancing the absorption capacity of the root system [17]. Further research will facilitate the functional analysis of SAUR genes and promote the application of improving buckwheat's tolerance to abiotic stress. Following salt stress treatment, significant changes in the expression of 36 genes were detected, highlighting their potential application value.

Conclusions

In this study, we identified 95 SAUR family members in tartary buckwheat, all possessing typical SAUR domain regions. We conducted a comprehensive analysis of FtSAUR members, examining physical characteristics, evolutionary relationships, gene structure, conserved motifs, and gene duplication events. Utilizing quantitative reverse transcription polymerase chain reaction (qRT-PCR), we systematically identified the expression patterns of SAUR genes across various tissues, revealing 24 genes that were not expressed. Among the remaining genes, 17 exhibited tissue-specific expression, while 10 were significantly expressed in vegetative or reproductive tissues, indicating their widespread involvement in the growth and development of buckwheat. Given the extensive involvement of the SAUR family in hormone responses, we further examined the expression of the FtSAUR gene under IAA treatment. Our research results indicate that 35 genes exhibited altered expression after treatment, with 5 genes showing

significant changes in expression. Similarly, we observed significant changes in the expression of 19 genes after drought treatment and 13 genes after salt treatment. These findings provide a theoretical basis for further exploring the functional characteristics of SAUR family members and reveal potential metabolic patterns related to growth, development, and stress-resistant metabolism in tartary buckwheat.

Abbreviations

FtSAURs	Small Auxin Up-regulated RNAs in <i>Fagopyrum tataricum</i>
MW	Molecular weight
pI	isoelectric point
qRT-PCR	Real-time quantitative PCR
SAUR	Small Auxin Up-regulated RNAs

Declarations

Authors'contributions

Conceptualization: D.X.D., G.Q.D.; Formal analysis: H.H.Q., Z.H.G.; Funding acquisition:G.Q.D.; Methodology: Y.Y., J.Z., Y.L., Q.H. Zu., Y.Y.G., C.L.F.; Writing – original draft preparation: D.X.D.; Writing- reviewing and editing: D.X.D., G.Q.D. All authors read and approved the final manuscript.

Data Availability

Not applicable

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Funding

The research content of this manuscript has not been funded by any specific Funding.

Competing interests

The authors declare that they have no competing interests.

References

1. Li Y, Han S, Qi Y. Advances in structure and function of auxin response factor in plants. *J Integr Plant Biol.* 2023;65(3):617–32.
2. Caumon H, Vernoux T. A matter of time: auxin signaling dynamics and the regulation of auxin responses during plant development. *J Exp Bot.* 2023;erad132.
3. Gomes GLB, Scortecci KC. Auxin and its role in plant development: structure, signalling, regulation and response mechanisms. *Plant Biol.* 2021;23(6):894–904.
4. Yu Z, Zhang F, Friml J, Ding Z. Auxin signaling: Research advances over the past 30 years. *J Integr Plant Biol.* 2022;64(2):371–92.
5. Luo C, Yan J, He C, Liu W, Xie D, Jiang B. Genome-Wide Identification of the SAUR Gene Family in Wax Gourd (*Benincasa hispida*) and Functional Characterization of *BhSAUR60* during Fruit Development. *Int J Mol Sci.* 2022;23(22):14021.
6. Hu W, Yan H, Luo S, Pan F, Wang Y, Xiang Y. Genome-wide analysis of poplar SAUR gene family and expression profiles under cold, polyethylene glycol and indole-3-acetic acid treatments. *Plant Physiol Biochem.* 2018;128:50–65.
7. Stortenbeker N, Bemer M. The SAUR gene family: the plant's toolbox for adaptation of growth and development. *J Exp Bot.* 2019;70(1):17–27.
8. Ann T, Kono N, Kosemura S, Yamahura S, Hasegawa K. Isolation and characterization of an auxin-inducible SAUR gene from radish seedlings. *DNA Seq.* 1998;9(5–6):329–33.
9. Markakis MN, Boron AK, Van Loock B, Saini K, Cirera S, Verbelen JP, Vissenberg K. Characterization of a small auxin-up RNA (SAUR)-like gene involved in *Arabidopsis thaliana* development. *PLoS ONE.* 2013;8(11):e82596.
10. Shin JH, Mila I, Liu M, Rodrigues MA, Vernoux T, Pirrello J, Bouzayen M. The RIN-regulated Small Auxin-Up RNA SAUR 69 is involved in the unripe-to-ripe phase transition of tomato fruit via enhancement of the sensitivity to ethylene. *New Phytol.* 2019;222(2):820–36.
11. Mathura SR, Sutton F, Bowrin V. Genome-wide identification, characterization, and expression analysis of the sweet potato (*Ipomoea batatas* [L.] Lam.) ARF, Aux/IAA, GH3, and SAUR gene families. *BMC Plant Biol.* 2023;23(1):622.
12. Luan J, Xin M, Qin Z. Genome-wide identification and functional analysis of the roles of SAUR gene family members in the promotion of cucumber root expansion. *Int J Mol Sci.* 2023;24(6):5940.
13. Chen Y, Hao X, Cao J. Small auxin upregulated RNA (SAUR) gene family in maize: identification, evolution, and its phylogenetic comparison with *Arabidopsis*, rice, and sorghum. *J Integr Plant Biol.* 2014;56(2):133–50.
14. Ren H, Gray WM. SAUR proteins as effectors of hormonal and environmental signals in plant growth. *Mol Plant.* 2015;8(8):1153–64.
15. Liu Y, Xiao L, Chi J, Li R, Han Y, Cui F, Li G. Genome-wide identification and expression of SAUR gene family in peanut (*Arachis hypogaea* L.) and functional identification of AhSAUR3 in drought tolerance. *BMC Plant Biol.* 2022;22(1):1–13.

16. Zhang H, Yu Z, Yao X, Chen J, Chen X, Zhou H, Xi Y. Genome-wide identification and characterization of small auxin-up RNA (SAUR) gene family in plants: evolution and expression profiles during normal growth and stress response. *BMC Plant Biol.* 2021;21:1–14.
17. Wang X, Yu R, Wang J, Lin Z, Han X, Deng Z, Chen H. The asymmetric expression of SAUR genes mediated by ARF7/19 promotes the gravitropism and phototropism of plant hypocotyls. *Cell Rep.* 2020;31(3).
18. Hou K, Wu W, Gan SS. SAUR36, a small auxin up RNA gene, is involved in the promotion of leaf senescence in *Arabidopsis*. *Plant Physiol.* 2013;161(2):1002–9.
19. Kathare PK, Dharmasiri S, Dharmasiri N. SAUR53 regulates organ elongation and apical hook development in *Arabidopsis*. *Plant Signal Behav.* 2018;13(10):e1514896.
20. Chae K, Isaacs CG, Reeves PH, Maloney GS, Muday GK, Nagpal P, Reed JW. *Arabidopsis* SMALL AUXIN UP RNA63 promotes hypocotyl and stamen filament elongation. *Plant J.* 2012;71(4):684–97.
21. Qiu T, Qi M, Ding X, Zheng Y, Zhou T, Chen Y, Wang J. The *SAUR41* subfamily of SMALL AUXIN UP RNA genes is abscisic acid inducible to modulate cell expansion and salt tolerance in *Arabidopsis thaliana* seedlings. *Ann Botany.* 2020;125(5):805–19.
22. Martínez-Villaluenga C, Peñas E, Hernández-Ledesma B. Pseudocereal grains: Nutritional value, health benefits and current applications for the development of gluten-free foods. *Food Chem Toxicol.* 2020;137:111178.
23. Appiani M, Rabitti NS, Proserpio C, Pagliarini E, Laureati M. Tartary Buckwheat: A New Plant-Based Ingredient to Enrich Corn-Based Gluten-Free Formulations. *Foods.* 2021;10(11):2613.
24. Kreft M. Buckwheat phenolic metabolites in health and disease. *Nutr Res Rev.* 2016;29(1):30–9.
25. Gao J, Kreft I, Chao G, et al. Tartary buckwheat (*Fagopyrum tataricum* Gaertn.) starch, a side product in functional food production, as a potential source of retrograded starch. *Food Chem.* 2016;190:552–8.
26. Zielińska D, Turemko M, Kwiatkowski J, Zieliński H. Evaluation of flavonoid contents and antioxidant capacity of the aerial parts of common and tartary buckwheat plants. *Molecules.* 2012;17(8):9668–82.
27. Finn RD, Clements J, Eddy SR. HMMER web server: interactive sequence similarity searching. *Nucleic Acids Res.* 2011;39:W29–37. (Web Server issue).
28. Bateman A, Birney E, Durbin R, Eddy SR, Howe KL, Sonnhammer EL. The Pfam protein families database. *Nucleic Acids Res.* 2000;1(1):263–6.
29. Yao Y, Zhao H, Sun L, Wu W, Li C, Wu Q. Genome-wide identification of MAPK gene family members in *Fagopyrum tataricum* and their expression during development and stress responses. *BMC Genomics.* 2022;23(1):1–18.
30. Krzywinski M, Schein J, Birol I, Connors J, Gascoyne R, Horsman D, Jones SJ, Marra MA. Circos: an information aesthetic for comparative genomics. *Genome Res.* 2009;19(9):1639–45.
31. Wang Y, Tang H, Debarry JD, Tan X, Li J, Wang X, Lee TH, Jin H, Marler B, Guo H, Kissinger JC, Paterson AH. MCScanX: a toolkit for detection and evolutionary analysis of gene synteny and

- collinearity. *Nucleic Acids Res.* 2012, 40 (7), e49.
32. Bailey TL, Boden M, Buske FA, Frith M, Grant CE, Clementi L, Ren J, Li WW, Noble WS. MEME SUITE: tools for motif discovery and searching. *Nucleic Acids Res.* 2009;37:W202–8. (Web Server issue).
 33. Liu M, Sun W, Ma Z, Zheng T, Huang L, Wu Q, Chen H. Genome-wide investigation of the AP2/ERF gene family in tartary buckwheat (*Fagopyrum Tataricum*). *BMC Plant Biol.* 2019;19(1):84.
 34. Xie T, Chen C, Li C, Liu J, Liu C, He Y. Genome-wide investigation of WRKY gene family in pineapple: evolution and expression profiles during development and stress. *BMC Genomics.* 2018;25(1):490.
 35. Yonekura-Sakakibara K, Hanada K. An evolutionary view of functional diversity in family 1 glycosyltransferases. *Plant J.* 2011;66(1):182–93.
 36. Dong G, Xiong H, Zeng W, Li J, Du D. Ectopic Expression of the Rice Grain-Size-Affecting Gene GS5 in Maize Affects Kernel Size by Regulating Endosperm Starch Synthesis. *Genes.* 2022;13(9):1542.
 37. Du D, Jin R, Guo J, Zhang F. Infection of embryonic callus with *Agrobacterium* enables high-speed transformation of maize. *Int J Mol Sci.* 2019;20(2):279.
 38. Liu M, Ma Z, Sun W, Huang L, Wu Q, Tang Z, Bu T, Li C, Chen H. Genome- wide analysis of the NAC transcription factor family in Tartary buckwheat (*Fagopyrum tataricum*). *BMC Genomics.* 2019;6(1):113.
 39. Sun W, Jin X, Ma Z, Chen H, Liu M. Basic helix-loop-helix (bHLH) gene family in Tartary buckwheat (*Fagopyrum tataricum*): Genome-wide identification, phylogeny, evolutionary expansion and expression analyses. *Int J Mol Sci.* 2020;155:1478–90.
 40. Enders TA, Strader LC. Auxin activity: Past, present, and future. *Am J Bot.* 2015;102(2):180–96.
 41. Kou X, Zhao X, Wu B, Wang C, Wu C, Yang S, Xue Z. Auxin response factors are ubiquitous in plant growth and development, and involved in crosstalk between plant hormones: a review. *Appl Sci.* 2022;12(3):1360.
 42. Romanov GA, Perception. Transduction and Crosstalk of Auxin and Cytokinin Signals. *Int J Mol Sci.* 2022;23(21):13150.
 43. Zhang M, Gao C, Xu L, Niu H, Liu Q, Huang Y, Li M. Melatonin and indole-3-acetic acid synergistically regulate plant growth and stress resistance. *Cells.* 2022;11(20):3250.
 44. Zhang Y, Ye T, She Z, Huang S, Wang L, Aslam M, Niu X. Small Auxin Up RNA (SAUR) gene family identification and functional genes exploration during the floral organ and fruit developmental stages in pineapple (*Ananas comosus* L.) and its response to salinity and drought stresses. *Int J Biol Macromol.* 2023;237:124061.
 45. Hagen G, Guilfoyle T. Auxin-responsive gene expression: genes, promoters and regulatory factors. *Plant Mol Biol.* 2002;49:373–85.
 46. Jain M, Tyagi AK, Khurana JP. Genome-wide analysis, evolutionary expansion, and expression of early auxin-responsive SAUR gene family in rice (*Oryza sativa*). *Genomics.* 2006;88(3):360–71.
 47. McClure BA, Guilfoyle T. Rapid Redistribution of Auxin-Rregulated RNAs during Gravitropism. *Science.* 1989;243(4887):91–3.

48. Minami A, Takahashi K, Inoue SI, Tada Y, Kinoshita T. (2019). Brassinosteroid induces phosphorylation of the plasma membrane H⁺-ATPase during hypocotyl elongation in *Arabidopsis thaliana*. *Plant and Cell Physiology*. 2019;60(5):935–944.
49. Zanin FC, Freitas NC, Pinto RT, Máximo WPF, Diniz LEC, Paiva LV. The SAUR gene family in coffee: Genome-wide identification and gene expression analysis during somatic embryogenesis. *Mol Biol Rep*. 2022;1–12.
50. Wen Z, Mei Y, Zhou J, Cui Y, Wang D, Wang NN. SAUR49 can positively regulate leaf senescence by suppressing SSPP in *Arabidopsis*. *Plant Cell Physiol*. 2020;61(3):644–58.
51. Xu K, Lou Q, Wang D, Li T, Chen S, Li T, ... Chen L. (2023). Overexpression of a novel small auxin-up RNA gene, *OsSAUR11*, enhances rice deep rootedness. *BMC Plant Biology*. 2023;23(1):1–14.
52. Spartz AK, Lor VS, Ren H, Olszewski NE, Miller ND, Wu G, ... Gray WM. (2017). Constitutive expression of *Arabidopsis* SMALL AUXIN UP RNA19 (SAUR19) in tomato confers auxin-independent hypocotyl elongation. *Plant Physiology*. 2017;173(2):1453–1462.
53. Tian Z, Han J, Che G, Hasi A. Genome-wide characterization and expression analysis of SAUR gene family in Melon (*Cucumis melo* L). *Planta*. 2022;255(6):123.
54. Xu X, Xiao YX, Liu MZ, Fu Y, He JL, Jiang Y. The small auxin-up RNA *OsSAUR45* affects auxin synthesis and transport in rice. *Plant Mol Biol*. 2017;94:97–107.
55. Depuydt S, Hardtke CS. Hormone signalling crosstalk in plant growth regulation. *Curr Biol*. 2011;21(9):R365–73.
56. Sharma N, Chaudhary C, Khurana P. Role of myo-inositol during skotomorphogenesis in *Arabidopsis*. *Sci Rep*. 2020;10(1):17329.
57. Guo G, Yu T, Zhang H, Chen M, Dong W, Zhang S, ... Jia B. Evidence That *PbrSAUR72* Contributes to Iron Deficiency Tolerance in Pears by Facilitating Iron Absorption. *Plants*. 2023;12(11):2173.

Figures

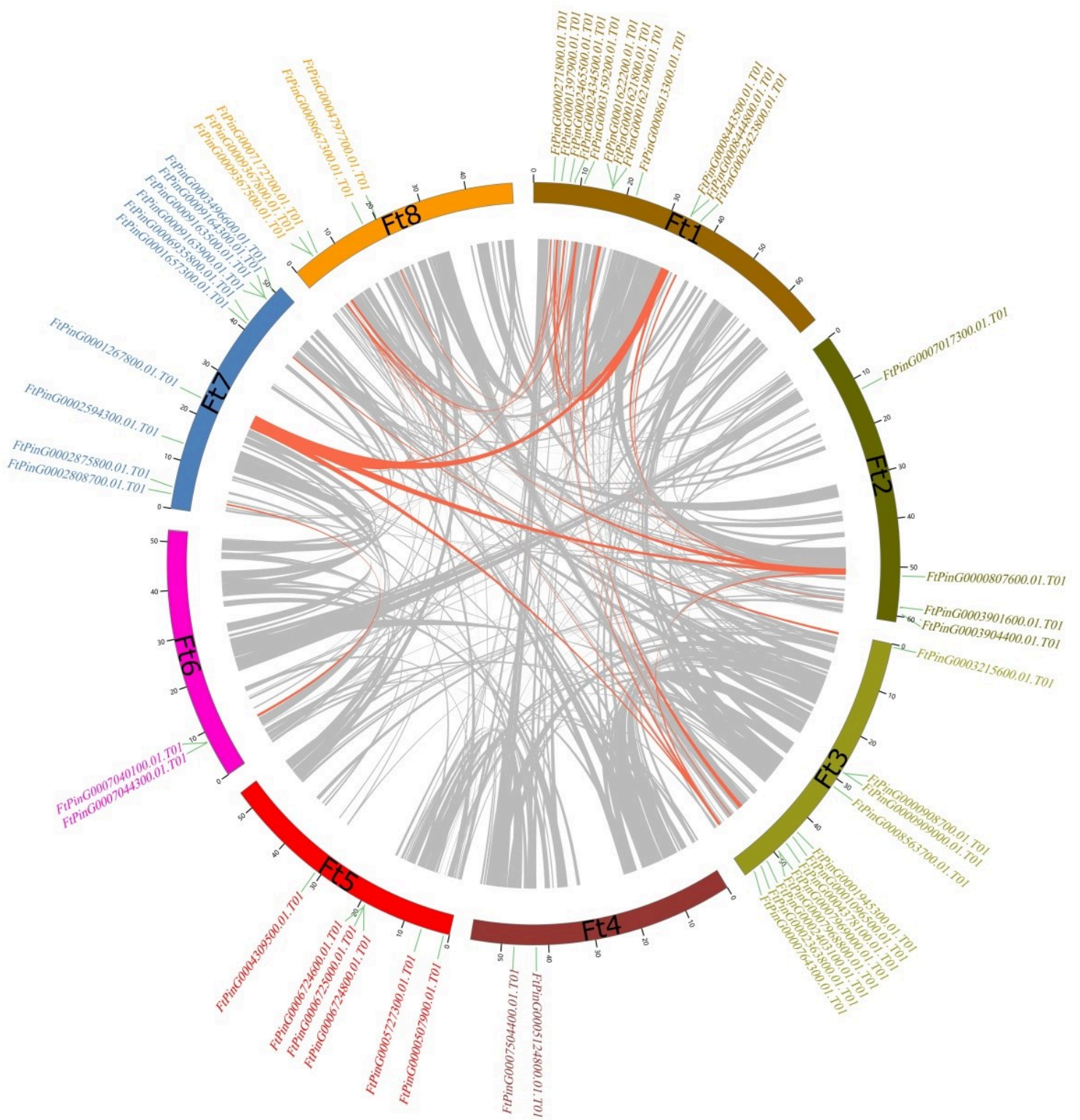


Figure 1

Chromosome localization of duplicated *FtSAURs* in buckwheat. The red lines represented the segmentally duplicated genes and the colored bands represented the collinear block in the genome of buckwheat.

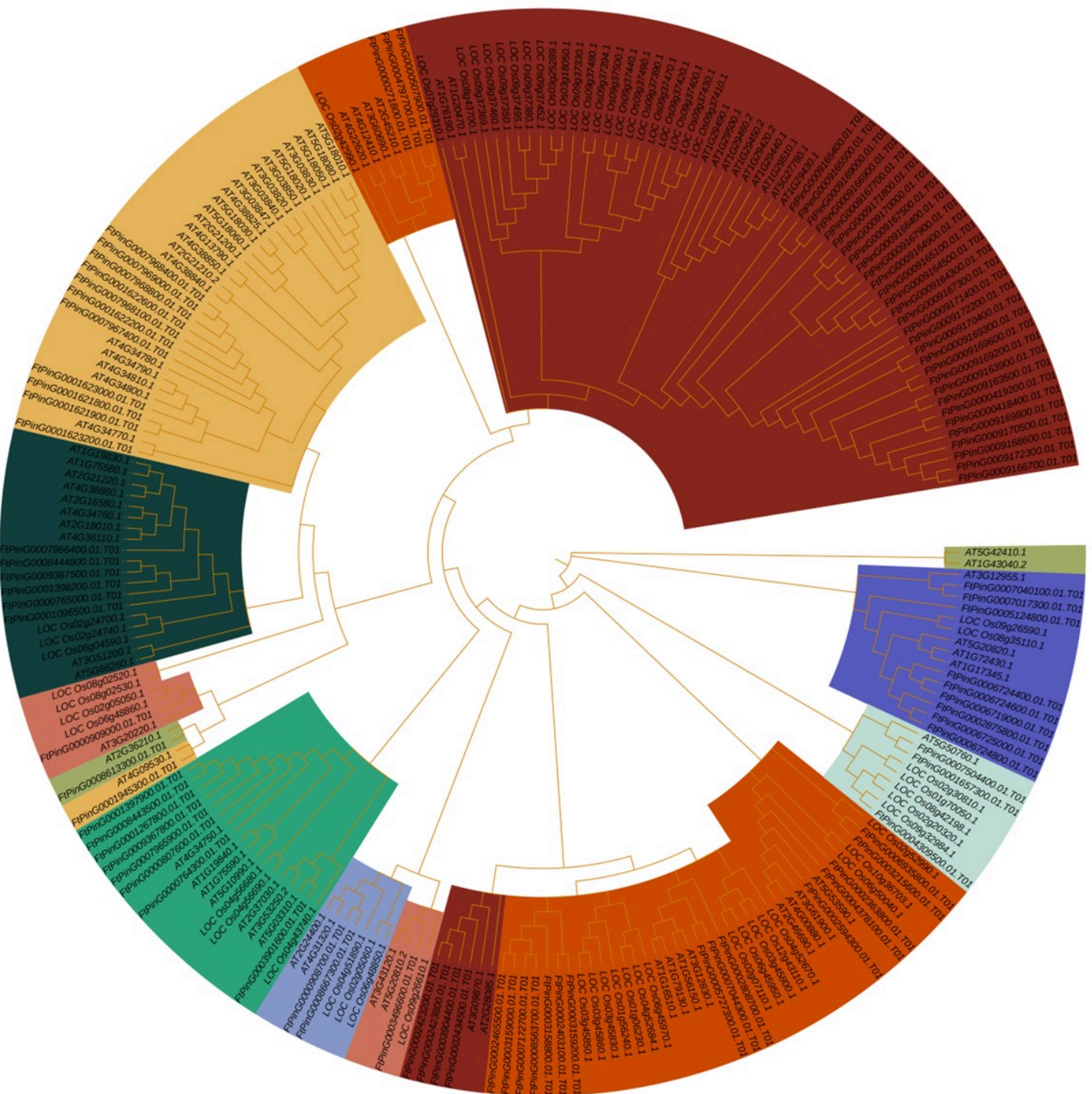


Figure 2

Unrooted phylogenetic tree representing relationships among SAUR domains of buckwheat, Arabidopsis and rice were constructed using the NJ method in MEGA7.0. SAURproteins from Arabidopsis are denoted by the prefix 'At'.



Figure 3

Gene structure of *FtSAURs*. Exons and introns were indicated by yellow rectangles and gray lines, respectively.

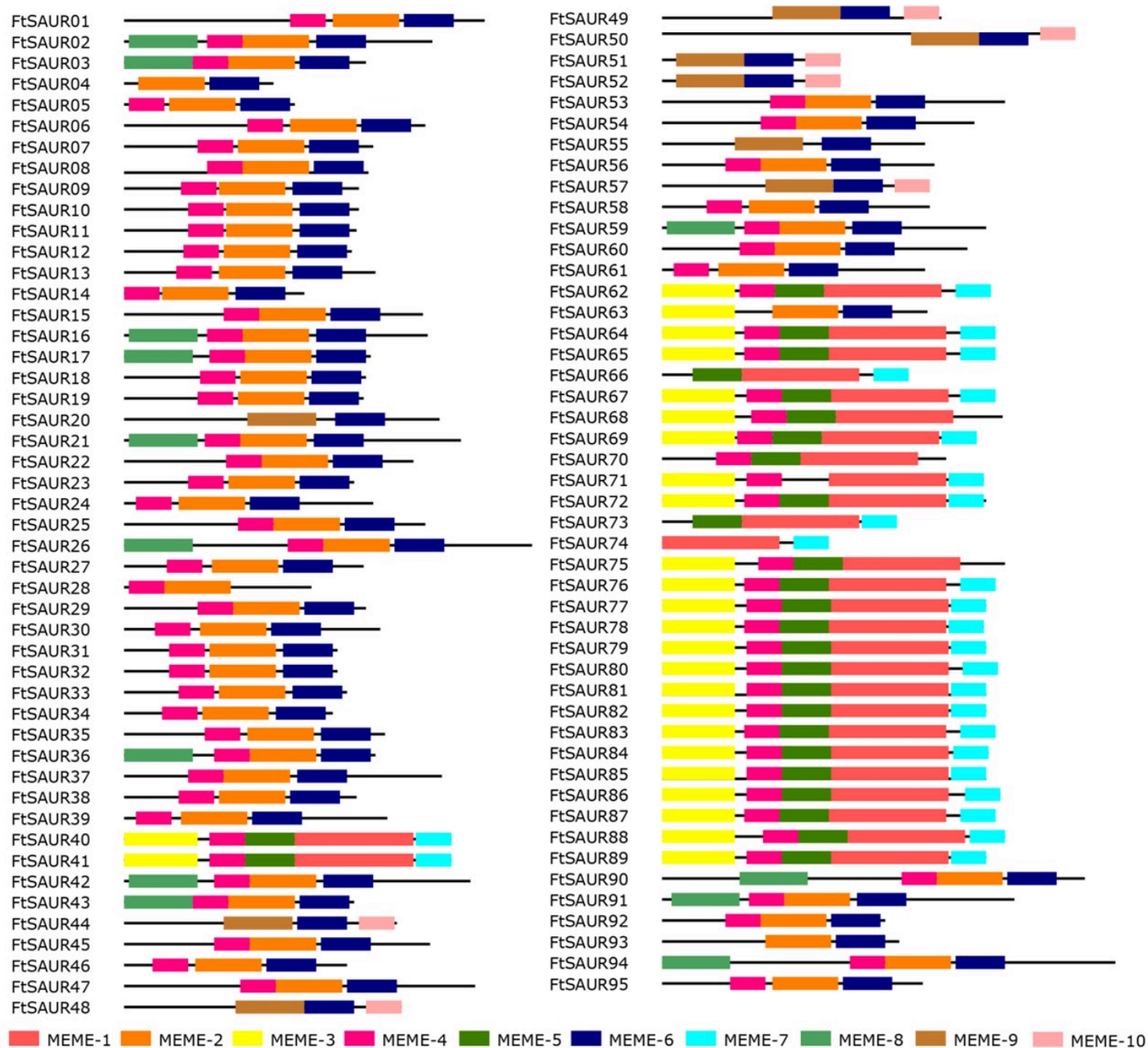


Figure 4

Amino acid motifs in the FtSAUR proteins were represented by colored boxes.

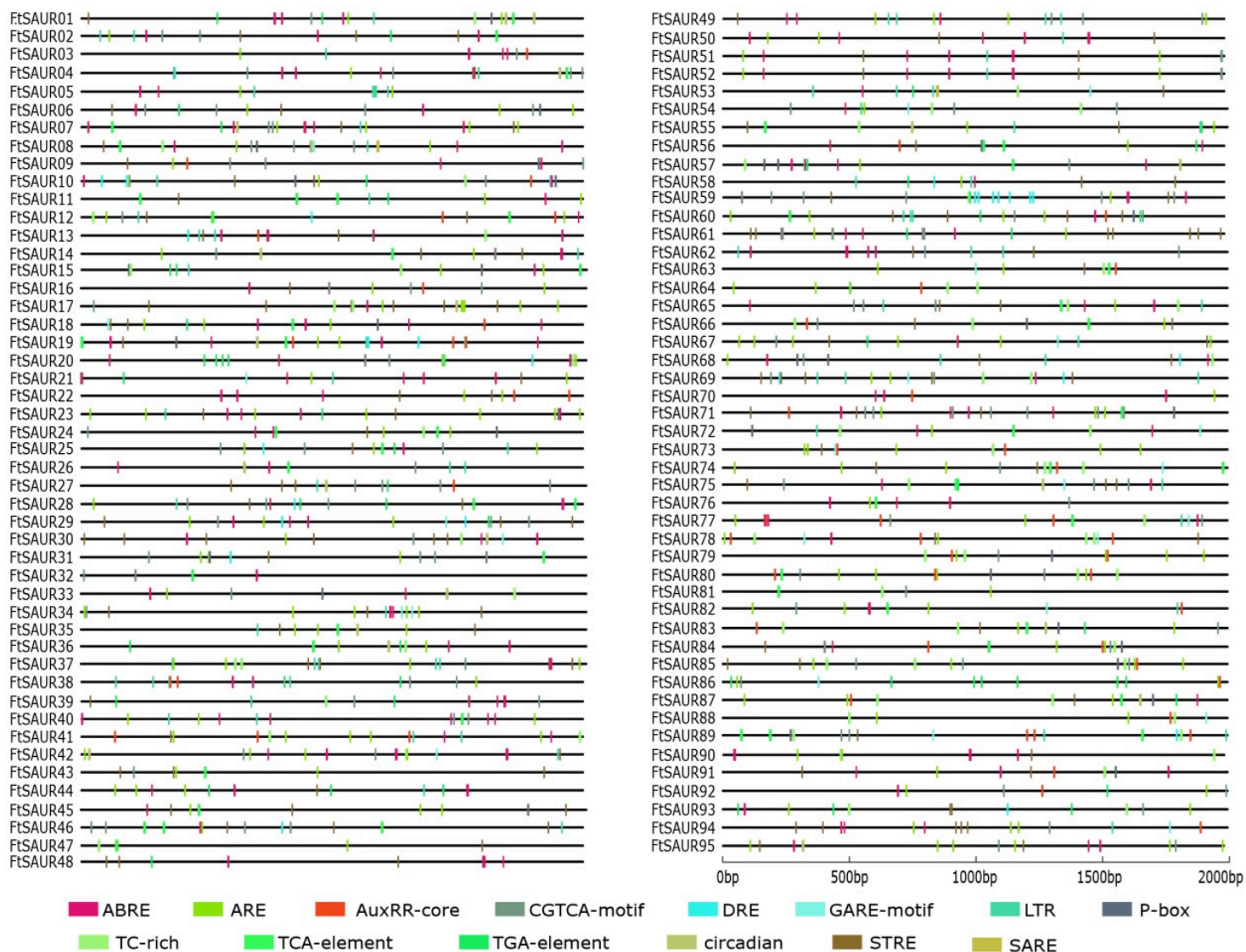


Figure 5

Amino acid motifs in the FtSAUR proteins were represented by colored boxes.

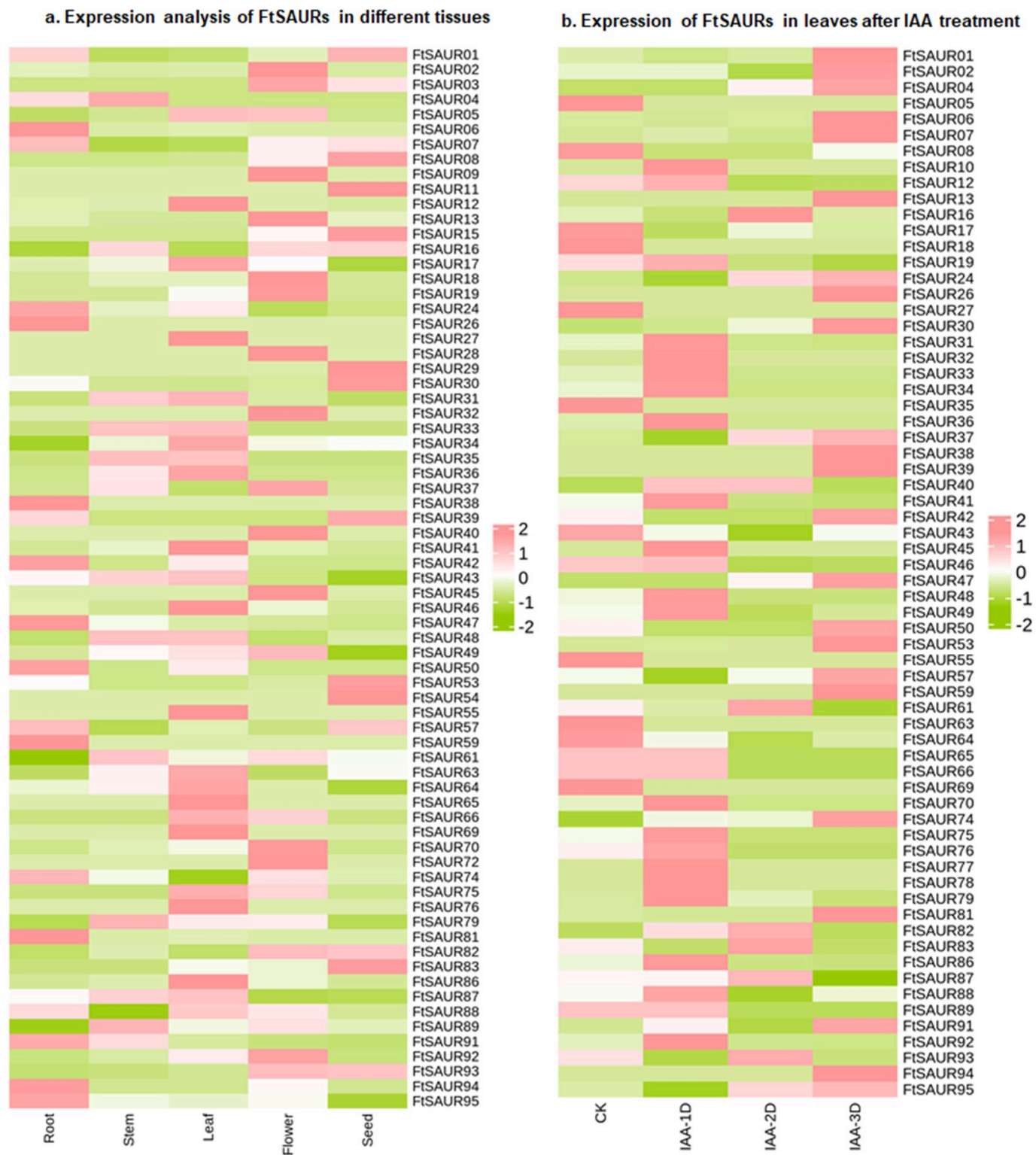


Figure 6

Expression analysis of *FtSAURs*. **a**, Expression patterns of *FtSAURs* across different tissues. Heatmap illustrates gene expression levels, with redder colors indicating higher expression and greener colors indicating lower expression. **b**, Expression patterns of *FtSAURs* in leaves after IAA treatment. Heatmap depicts gene expression levels at different stages of IAA treatment, with redder colors indicating higher expression and greener colors indicating lower expression.



Figure 7

Expression profiling of *FtSAURs* in leaves after drought and salt. Heatmap illustrates gene expression levels, with redder colors indicating higher expression and greener colors indicating lower expression.