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The sweet sorghum *SbWRKY50* is negatively involved in salt response by regulating ion homeostasis

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Abstract

The WRKY transcription factor family is involved in responding to biotic and abiotic stresses. Its members contain a typical WRKY domain and can regulate plant physiological responses by binding to W-boxes in the promoter regions of downstream target genes. We identified the sweet sorghum *SbWRKY50* (Sb09g005700) gene, which encodes a typical class II of the WRKY family protein that localizes to the nucleus and has transcriptional activation activity. The expression of *SbWRKY50* in sweet sorghum was reduced by salt stress, and its ectopic expression reduced the salt tolerance of *Arabidopsis thaliana* plants. Compared with the wild type, the germination rate, root length, biomass and potassium ion content of *SbWRKY50* over-expression plants decreased significantly under salt-stress conditions, while the hydrogen peroxide, superoxide anion and sodium ion contents increased. Real-time PCR results showed that the expression levels of *AtSOS1*, *AtHKT1* and genes related to osmotic and oxidative stresses in over-expression strains decreased under salt-stress conditions. Luciferase complementation imaging and yeast one-hybrid assays confirmed that *SbWRKY50* could directly bind to the upstream promoter of the *SOS1* gene in *A. thaliana*. However, in sweet sorghum, *SbWRKY50* could directly bind to the upstream promoters of *SOS1* and *HKT1*. These results suggest that the new WRKY transcription factor *SbWRKY50* participates in plant salt response by controlling ion homeostasis. However, the regulatory mechanisms are different in sweet sorghum and *Arabidopsis*, which may explain their different salt tolerance levels. The data provide information that can be applied to genetically modifying salt tolerance in different crop varieties.

Key message

(1) Sweet sorghum *SbWRKY50* is negatively involved in salt response.
(2) Over-expression of *SbWRKY50* in *A. thaliana* affects plant growth, ROS and the ioncontents.
(3) *SbWRKY50* could directly bind to the upstream promoter of the *SOS1* gene in *A. thaliana* and the promoter of *SOS1* and *HKT1* in sweet sorghum.

Keywords Sweet sorghum · *SbWRKY50* · Salt stress · SOS1 · Ion homeostasis

Introduction

Salt stress is an important factor that seriously affects crop growth and yield (Asano et al. 2012; Liu et al. 2017a, b). Salt stress can inhibit the growth and development of plants, disrupt the permeability of cell membranes and biologically functional molecules in cells, and seriously affect the physiological and metabolic processes of plants (Syvertsen and Garcia-Sanchez 2014; Reginato et al. 2014; Wu et al. 2014; Han et al. 2014; Liu et al. 2015; Sui et al. 2017, 2018). Soil salinization is a worldwide problem, especially in arid and semi-arid areas. China

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has severe soil salinization, affecting tens of millions of hectares (Li et al. 2014a, b). Moreover, owing to the improper development and utilization of irrigation measures, a large area of secondary salinized soil has been produced. Most crops do not grow normally on saline–alkali soil (Li et al. 2012). Therefore, studying the molecular mechanisms of salt resistance is necessary for increasing salt tolerance and utilizing salinized soil.

To maintain the normal physiological metabolism of cells under salt-stress conditions, plants undergo a series of physiological and biochemical reactions, including ion transport and absorption (Shao et al. 2014), the synthesis of osmotic regulators (Li et al. 2014; Wei et al. 2017; You et al. 2019) and the enhancement of antioxidant enzyme levels (Guo et al. 2017). Additionally, plants induce the expression levels of genes that have different functions in response to salt stress. Regulating the expression levels of functional genes through transcription factors is a key mechanism of plant responses to stress. At present, there are many transcription factor families responding to stress, such as bZIP, WRKY, AP2/EREBP and MYB and so on.

The WRKY transcription factor is a supergene family named for a highly conserved WRKY domain consisting of 60 amino acids that are found in the family members (Rinerson et al. 2015). However, there are exceptions. In some WRKY transcription factors, WRKY residues in the WRKY domain are replaced by WRRY, WSKY, WKRY, WVKY or WKKY motifs. The N-terminal of the domain has a conservative WRKYGQK core sequence, and the C-terminal has a zinc-finger structure, C_2–H_2 or C_2–HC. WRKY domain proteins bind specifically to W-boxes. The conserved sequence of the W-box is TTGACC or TTG ACT, and the four middle bases TGAC are the core W-box sequence. The selection of a W-box by a WRKY transcription factor protein depends on the two flanking bases. At present, WRKY transcription factors are divided into three protein subfamilies, classes I, II and III.

Since the first WRKY gene was cloned in sweet potatoes (Ishiguro and Nakamura 1994), it has been found that WRKY is a ubiquitous class of transcription factors in plants especially in some important economic crops such as soybean (Zhou 2008; Wang et al. 2015), potato (Liu et al. 2017a, b), cotton (Si et al. 2017), tomato (Wang et al. 2017), maize (Zhang et al. 2017), rice (Lee et al. 2018) and wheat (Gao et al. 2018). The WRKY protein is involved in abiotic stress responses (Ali et al. 2018; López-Galiano et al. 2018), plant disease resistance (Lan et al. 2013), plant development and material metabolism (Yang et al. 2016; Sun et al. 2003), seed dormancy (Ding et al. 2014), plant senescence (Potschin et al. 2014), hormone signaling (Birkenbihl et al. 2015; Tian et al. 2017) and other processes (Chen et al. 2012). WRKY transcription factor is a new participant in signal transduction network. The interaction between downstream target genes and upstream regulators constitutes a complex WRKY transcription factor regulatory network, which is a new research field. At present, the regulatory network of WRKY transcription factors has been studied in some plant species (Zou et al. 2004; Kim et al. 2008; Jiang et al. 2016), but the regulatory network in sweet sorghum is the first introduction.

Sweet sorghum [*Sorghum bicolor* (L.) Moench], known as the "second generation sugarcane", is a kind of crop with high sugar content and high yield (Sui et al. 2015; Schnippenkoetter et al. 2017). It is an important food, feed and energy crop in the world. Sweet sorghum not only has a very high biomass but also has salt and alkali tolerant. Many unique genes were found to be related to salt tolerance of sweet sorghum (Zheng et al. 2011; Yang et al. 2018). Therefore, research on these unique genes and the related characteristics of sweet sorghum can be applied to the genetic modification of other crops, allowing us to obtain stress-tolerant crops, and manage the growth and development of plants in saline environments.

In this study, we isolated *SbWRKY50* from our former transcriptomic data, because the gene was down regulated in M81-E and was not induced in Roma by salt stress (Yang et al. 2017, 2018). We found that *SbWRKY50* down-regulated under salt treatment. After over-expressing *SbWRKY50* in *Arabidopsis thaliana*, the related physiological indexes were determined, and yeast one-hybrid and luciferase complementation imaging assays were performed. *SbWRKY50* can negatively regulate salt responses by altering ion homeostasis in both sweet sorghum and *A. thaliana*. But the regulating mechanism is different in sweet sorghum and *A. thaliana*.

Results

SbWRKY50 is a typical WRKY transcription factor, containing the WRKYGQ(K)K motif

The coding sequence (CDS) of *SbWRKY50* contains 621 bases and encodes 206 amino acids. There is a

typical WRKY domain between amino acids 109 and 168 in SbWRKY50, and a zinc-finger structure, C_2H_2, at the C-terminus (Fig. S1A, B). Using the BLAST function of NCBI, 14 sequences with high homology to SbWRKY50 were identified (Fig. S1C). WRKYGQ(K)K and C_2–H_2 are highly conserved in the WRKY family. The phylogenetic tree showed that the SbWRKY50 protein had high homology with *Panicum hallii* WRKY51 (Fig. S1D).

SbWRKY50 is a negative regulatory transcription factor expressed in the nucleus

To test if SbWRKY50 is located in the nucleus as a transcription factor, we performed the transient transformation in tobacco with *SbWRKY50*: GFP. GFP was expressed in cell membrane and nucleus, but GFP fused with SbWRKY50 was located in nucleus (Fig. 1A). This indicates that SbWRKY50 is a nuclear localization protein.

To elucidate the function of *SbWRKY50* under salt condition, we performed salt treatment experiments in sweet sorghum. As shown in Fig. 1B, during the 50 mM NaCl treatment, the expression of *SbWRKY50* increased in shoot, but decreased sharply during the 100 mM NaCl treatment, and reached its lowest level during the 200 mM NaCl treatment. In roots, its expression level significantly decreased with the increase of salt concentration, reaching its lowest level during the 200 mM NaCl treatment (Fig. 1B). Therefore, these results support that *SbWRKY50* can be induced by salt and might be a negative regulatory transcription factor.

SbWRKY50 is a transcription factor with transcriptional activity

We analyzed the transcriptional activity of *SbWRKY50* using a yeast system to investigate the potential role of *SbWRKY50*. On tryptophan deficient medium (SD/−Trp), both the experimental and control groups grew normally. On tryptophan, adenine and histidine (SD/−Trp − Ade − His) and the same medium supplemented with 5-bromo-4-chloro-3-indole-a-D-galactoside (X-a-gal; SD/−Trp − Ade − His + X-a-gal), the control group did not grow normally, while the experimental group grew normally and colonies turned blue (Fig. 1C). These results indicate that *SbWRKY50* has a transactivational capacity.

Over-expression of *SbWRKY50* enhances plant sensitivity to salt stress

To further demonstrate that *SbWRKY50* acts as a negatively regulated transcription factor in salt tolerance, *SbWRKY50* was derived by CaMV35s promoter and was transformed into *A. thaliana*. We generated two transgenic lines W50-12 and W50-13. The expression level of *SbWRKY50* was 7.84 (W50-12) and 9.67 (W50-13) times higher than wild type (WT) *Arabidopsis* (Fig. 2A), which proved that *SbWRKY50* had been successfully transformed

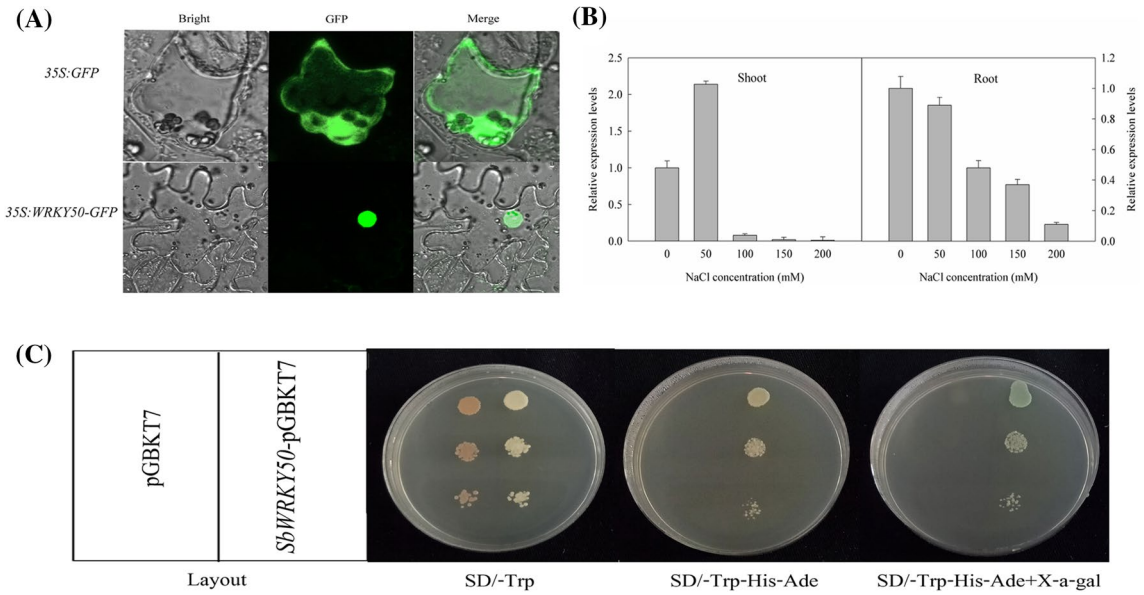

Fig. 1 Subcellular localization, expression pattern and transcriptional activity analyses of *SbWRKY50*. **A** Localization of SbWRKY50 in tobacco cells (35S: GFP as control group, 35S:WRKY50-GFP as experimental group). **B** The expression levels of *SbWRKY50* in roots and leaves under 0-, 50-, 100-, 150- and 200 mM salt conditions. **C** Analysis of transcriptional activity of *SbWRKY50* using a yeast system

into *Arabidopsis*. Furthermore, we measured the biomass of over-expression and WT plants in the treatment of 0 and 100 mM NaCl. The dry weight and fresh weight of each *Arabidopsis* line decreased significantly, but the decline degree of over-expressed lines was significantly higher than that of WT (Fig. 2B, C). This suggests that over-expression of *SbWRKY50* decreased plant biomass.

We also measured seed germination and root length. There were no differences in the seed germination rates and root lengths of the lines not subjected to NaCl treatment. As the salt concentration increased, the seed germination rates and root length of each strain decreased significantly (Fig. 2D). When treated with 50 and 100 mM NaCl, the germination rates and root lengths of the over-expression lines were significantly lower than those of the WT. When germinated on 150 mM NaCl, reduction in seed germination and root length was observed both in WT and over-expression plants, however reduction was significantly higher in overexpression lines as compared to WT plants (Fig. 2E, F). This indicates that the over-expression of *SbWRKY50* affected seed germination and root length.

Under salt-stress conditions, sodium ions (Na^+) are the main ions that cause plant salt damage, while potassium ions (K^+) are an essential element and important osmotic adjustment substance for plant growth. The level of Na^+ has a significant inhibitory effect on the absorption of K^+. To detect ion metabolism in plants, we determined the contents of Na^+ and K^+ in over-expression and WT lines. Under 0 mM NaCl conditions, there were no significant differences in the Na^+ and K^+ contents in the leaves and roots of transgenic *Arabidopsis* compared with WT plants. However, under 100 mM NaCl conditions, compared with WT plants, the Na^+ content in *SbWRKY50* over-expression plants increased significantly, while the K^+ content decreased significantly. As a result, Na^+/K^+ increased significantly in the over-expression plants (Fig. 3A–C). This suggests that the over-expression of *SbWRKY50* reduces the ability of *Arabidopsis* to resist salinity by increasing the Na^+ content and decreasing the K^+ content.

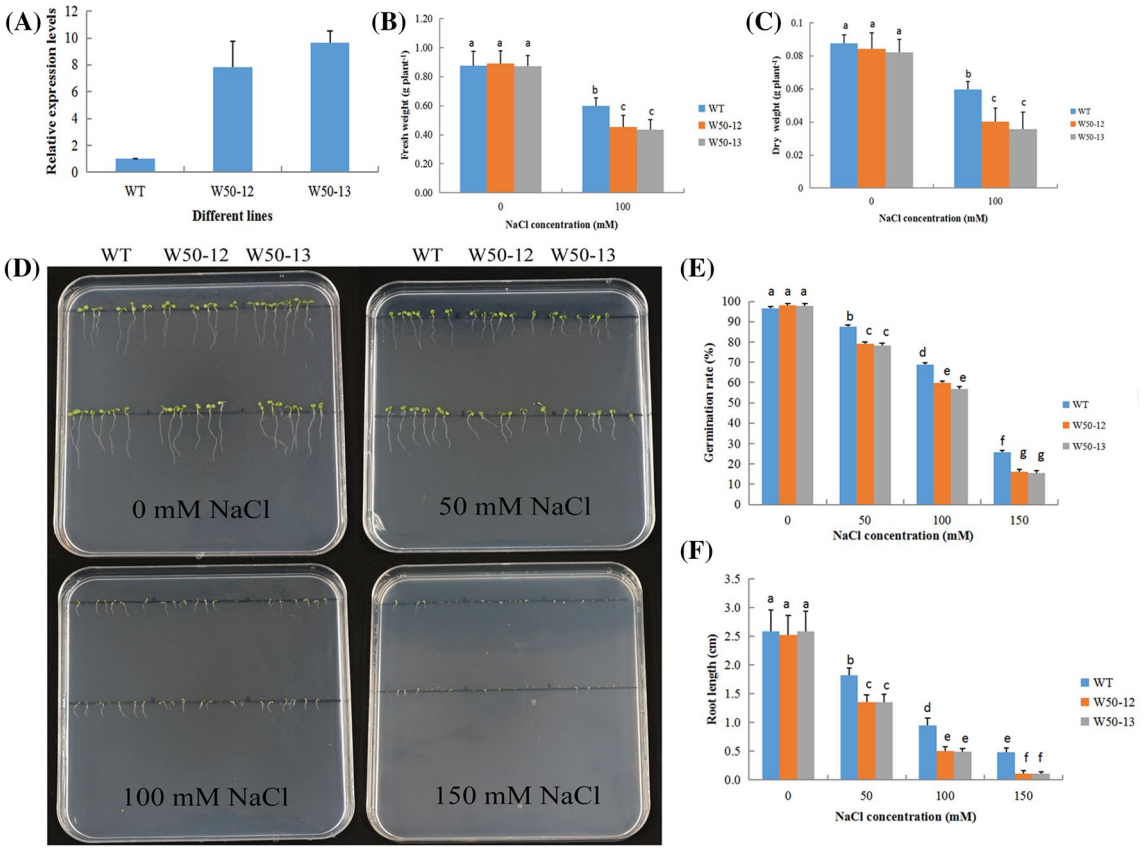

Fig. 2 The determination of physiological parameters in *Arabidopsis* wild-type (WT) and *SbWRKY50* over-expression lines. **A** Expression of *SbWRKY50* in over-expression plants. **B** and **C** Fresh weights (**B**) and dry weights (**C**) of WT and over-expression plants under different salt conditions. **D** Under salt treatment, the phenotypes of WT and over-expressed *A. thaliana* (W50-12, W50-13) at seedling stage, with six seedlings in each group. **E** and **F** Germination rates (**E**) and root lengths (**F**) of WT and over-expression plants under different salt conditions. Each column represents the means ± SDs of five measurements. Means identified by different letters are significantly different at $P < 0.05$

Under salt-stress conditions, plants can produce hydrogen peroxide (H_2O_2), superoxide anions ($O_2^{-\cdot}$) and other substances owing to oxidation. DAB solution can react with plant endogenous H_2O_2 to form reddish brown spots, which reflect the H_2O_2 contents in plants. Similarly, nitro blue tetrazolium (NBT) solution can be reduced to a water-insoluble blue substance under the action of $O_2^{-\cdot}$, which reflects the $O_2^{-\cdot}$ contents in plants. Under 0 mM NaCl conditions, the degrees of DAB and NBT staining of each strain were not significantly different. However, under 100 mM NaCl conditions, the degrees of both DAB and NBT staining in over-expression strains were deeper than in WT (Fig. 3D, E), which indicated that under saline conditions, the over-expression of *SbWRKY50* made *Arabidopsis* plants more sensitive to oxidative stress than WT plants.

SbWRKY50 regulates the expression of stress-related genes

To identify the target genes of *SbWRKY50*, we assessed the transcriptional changes of some genes in response to osmotic (*SOD* and *APX*), oxidative (*RD29B* and *P5CS1*) and ion stresses (*CLC-C*, *SOS1* and *HKT1*) using qRT-PCR. Under salt-stress conditions, the expression levels of these genes were induced, but the levels of induction were less in the *SbWRKY50* over-expression strains than in WT strains (Fig. 4A). Thus, *SbWRKY50* may play an important role in regulating the expression levels of stress-responsive genes. WRKY transcription factors regulate downstream genes by binding with W-boxes. To further screen the target genes of *SbWRKY50*, we analyzed the promoters of these genes (Table S1), and only the promoters of *A. thaliana* salt overly sensitive 1 (*AtSOS1*, Na^+/H^+ reverse transporter), sweet sorghum *SbSOS1* and high-affinity potassium transporter (*SbHKT1*) contained W-boxes. Therefore, we speculate that the *SOS1* and *HKT1* genes might be targets of *SbWKY50*.

SbWRKY50 directly binds to the *AtSOS1*, *SbSOS1* and *SbHKT1* promoters

To determine whether SbWRKY50 binds with the promoters of *AtSOS1*, *SbSOS1* and *SbHKT1*, we performed

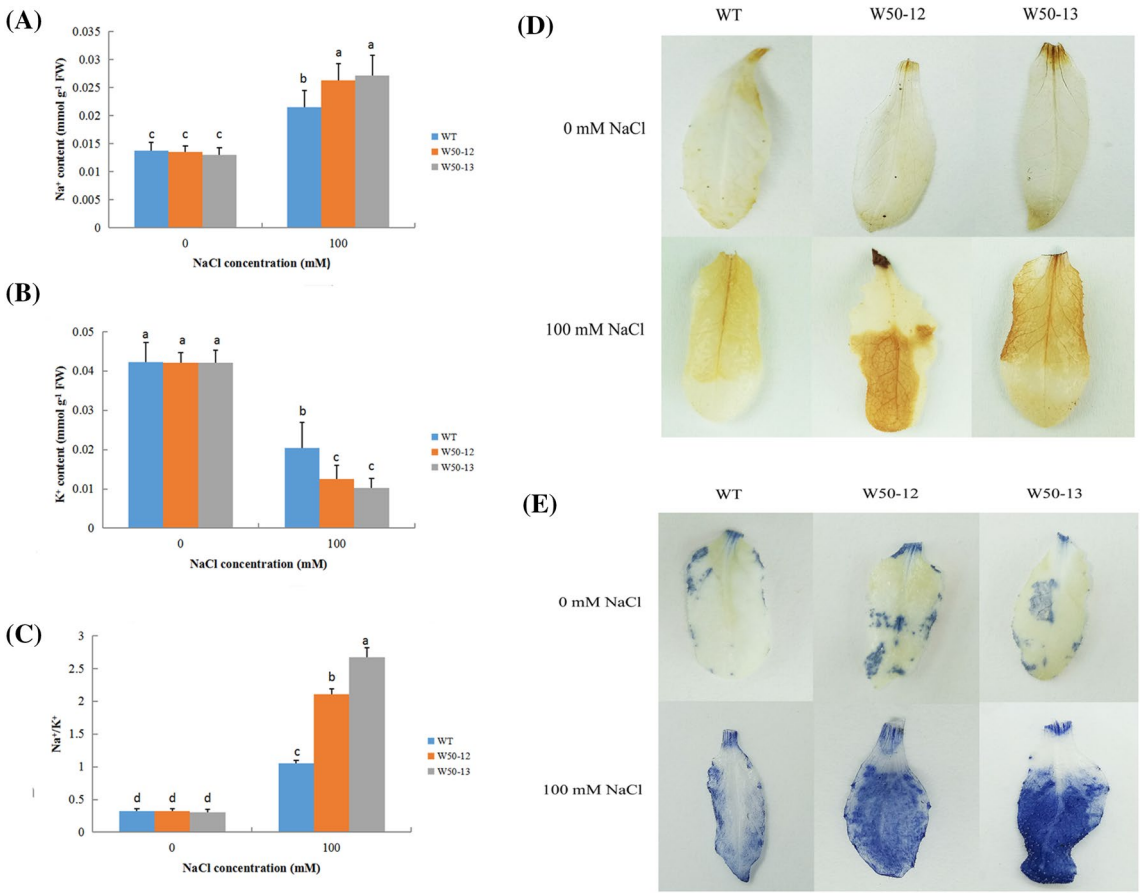

Fig. 3 Changes in ions and reactive oxygen species. **A**–**C** The Na^+ and K^+ contents and Na^+/K^+ ratio in roots of WT and over-expression plants during different salt treatments. Each column represents the means ± SDs of five measurements. Means identified by different letters are significantly different at $P < 0.05$. **D** DAB staining. **E** NBT staining. Each staining experiment was repeated five times

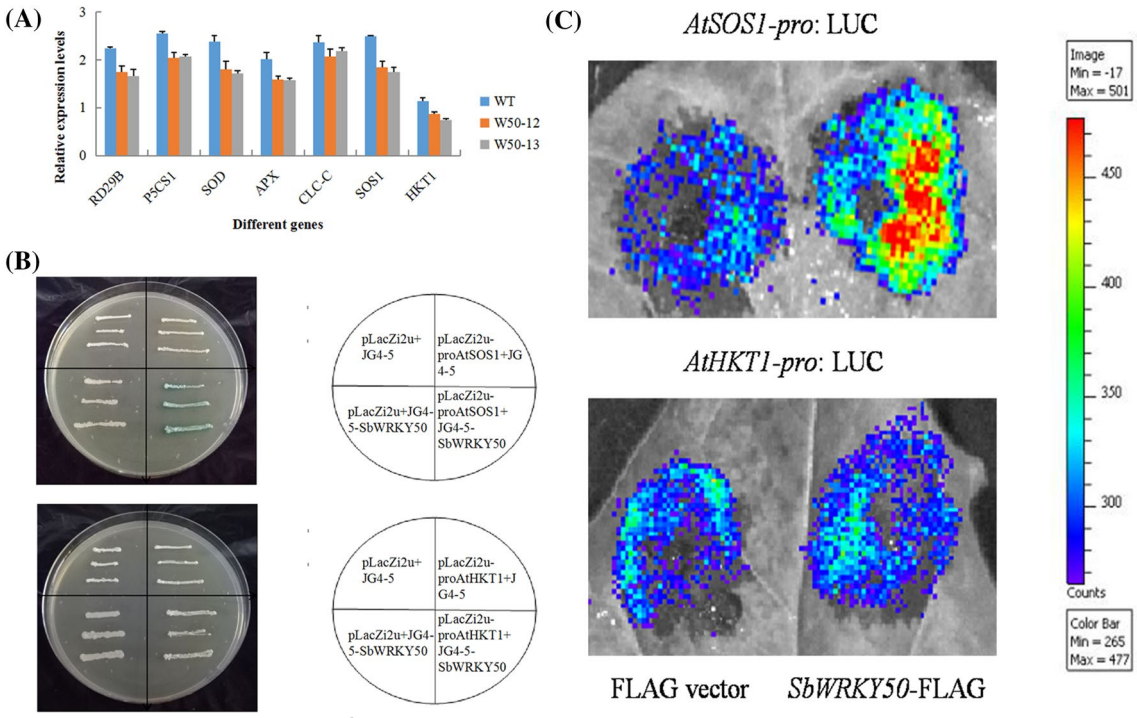

Fig. 4 Expression levels of stress-related genes and interactions between *SbWRKY50* and its downstream target genes. **A** The expression levels of genes responding to osmotic, stress and ionic stresses in *SbWRKY50* over-expression strains compared with wild-type strains under salt-stress conditions. Each column represents the means ± SDs of five measurements. Means identified by different letters are significantly different at $P < 0.05$. **B** and **C** The interactions of *SbWRKY50* with proAtSOS1 and proAtHKT1 were validated using yeast one-hybrid and luciferase complementation imaging assays

yeast one-hybrid and luciferase complementation imaging assays. The yeast one-hybrid assays showed that only when proAtSOS1-pLacZi2u, proSbSOS1-pLacZi2u and proSbHKT1-pLacZi2u were independently co-transformed with GAD-SbWRKY50, LacZ expression was activated and colonies appeared blue on SD/−Trp − Ura + X-a-gal (Figs. 4B, 5A). Similarly, when proAtSOS1-pGreenII-0800, proSbSOS1-pGreenII-0800 and proSbHKT1-pGreenII-0800 were independently co-transformed with SbWRKY50-pMWB110, strong in vivo fluorescence signals were observed using the imaging system (Figs. 4C, 5B). No fluorescence signals were observed in the co-transformation of SbWRKY50-pMWB110 and proAtHKT1-pGreenII-0800. This indicates that SbWRKY50 can regulate plant salt tolerance by interacting directly with *AtSOS1*. However, in sweet sorghum, SbWRKY50 can regulate salt tolerance by directly binding the promoters of *SbSOS1* and *SbHKT1*.

Discussion

SbWRKY50 protein belongs to class II of the WRKY family with the N-terminus containing a conserved WRKY domain, and the C-terminus containing a C_2–H_2 zinc-finger structure (Fig. S1A–C). The transient transformation of tobacco indicated that WRKY50 was expressed in the nucleus, and the transcriptional activity of WRKY50 was verified using a yeast system (Fig. 1A, C). Under salt-stress conditions, the transcriptional level of *WRKY50* in sweet sorghum decreased with the increase of salt concentration (Fig. 1B). These results suggest that the *SbWRKY50* gene is constitutively expressed, and its expression is inhibited by salt stress, which indicate that *SbWRKY50* might be a negatively regulation factor.

Seed germination is the first stage of plant growth, and roots are the main organ involved in plants perceiving environmental changes (Almansouri et al. 2001; Guo et al. 2018; Li et al. 2018). When plants are subjected to adverse conditions, germination rate and root length are the first characteristics to be affected. Jafarzadeh and Aliasgharzad (2007) found that with an increasing salt content, the yield of sugar beet decreased. The salt level had a significant effect on

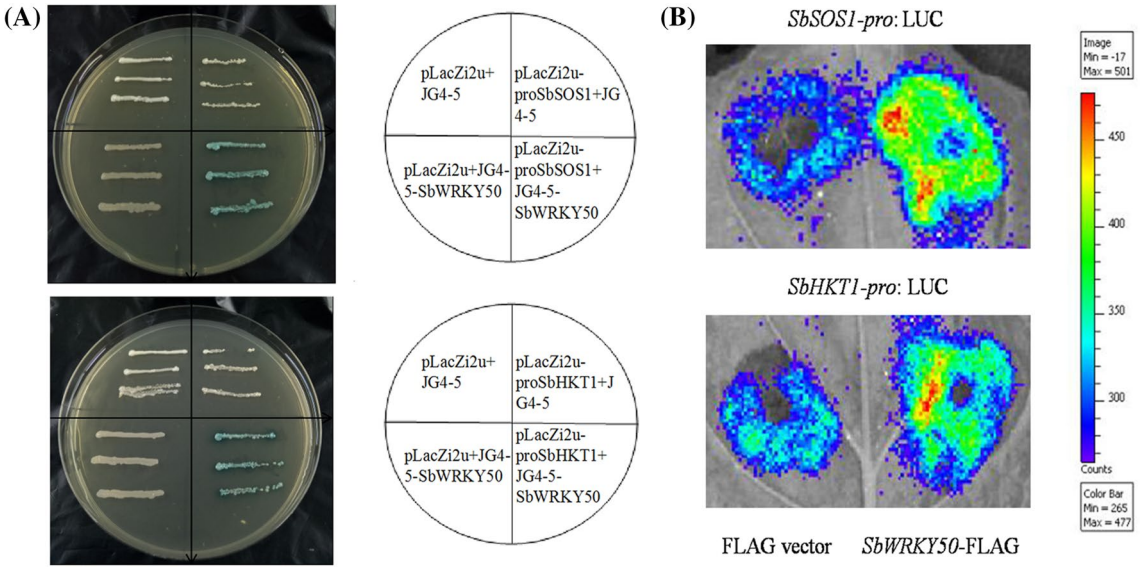

Fig. 5 The interactions of SbWRKY50 with proSbSOS1 and proSbHKT1 were validated using yeast one-hybrid and luciferase complementation imaging assays. **A** Yeast one-hybrid. **B** Luciferase complementation imaging assays

the seed germination rates and root lengths of four sugar beet varieties (pp22, ic2, pP36 and 7233). Cai et al. (2014) showed that the over-expression of *WRKY58* in rice results in delayed germination and inhibited post-germination development. Xu et al. (2018a, b) found that the over-expression of *Glycine max WRKY49* significantly increased the germination rates, survival rates, root lengths, rosette diameters, relative electrolyte leakage levels and proline contents in composite seedlings and transgenic *Arabidopsis* than in WT. In this study, the germination rate, root length and the biomass were significantly inhibited in over-expression *SbWRKY50* plants than in WT.

Under salt stress, Na^+ enters cells in large quantities, resulting in the reduction in K^+ uptake by cells and the destruction of the ion balance in plant cells (Wei et al. 2015). High salt concentrations also destroy plant defense systems, and the accumulation of reactive oxygen species (H_2O_2 and $O_2^{-\cdot}$) leads to membrane lipid peroxidation, which results in a series of physiological metabolic disorders (Li et al. 2000; Song et al. 2019). In this study, the K^+ content of *SbWRKY50* over-expression plants were significantly decreased under high-salt conditions compared with WT, while the Na^+ content and reactive oxygen species accumulation increased, affecting the ion homeostasis. These results indicate that the over-expression of *SbWRKY50* in *A. thaliana* can inhibit plant growth, and dry matter and reactive oxygen species accumulation, leading to oxidative stress, and reduce the abilities of plants to maintain their ion balance. Thus, plant salt tolerance was reduced.

Salt stress also can induce some stress related genes expression. In this study, we determined the expression levels of some stress-related genes, *RD29B*, *P5CS1*, *SOD*, *APX*, *CLC-C*, *SOS1* and *HKT1*. Previous research shows that the osmotic stress-related gene *RD29B* is induced by salt and drought and activates the *Arabidopsis* MAKK or MAPK protein (Hu et al. 2006). Deletions of the *P5CS1* gene in *A. thaliana* result in accumulations of reactive oxygen species (Szekely et al. 2008). The over-expression of *P5CS1* results in a higher biomass and a higher root growth rate during early development (Ibragimova et al. 2015). Oxidative stress-related genes *SOD*, *APX* are induced by salt and play key roles in preventing excessive ROS accumulation and protecting cells against ROS caused by salt stress (Ma 2002; Wang et al. 2004; Pedro et al. 2013). *OsCLC1* plays an important role in regulating Cl^- homeostasis in rice under salt stress (Diedhiou and Golldack 2006). *SOS1* can regulate the excretion of Na^+ in root cells (Ding and Zhu 1997; Shi et al. 2002, 2003). *HKT1* plays an important role in ion redistribution. Our results also show that the over-expression of *SbWRKY50* can reduce the expression levels of *RD29B*, *P5CS1*, *SOD*, *APX*, *CLC-C*, *SOS1* and *HKT1* under salt stress conditions.

The WRKY transcription factor usually combines with the W-box in the promoters of target genes (Bakshi and Oelmüller 2014). In this study, we identified W-boxes only in the promoters of *Arabidopsis SOS1* and sweet sorghum *SOS1* and *HKT1*, while there were no binding sites in the promoters of *RD29B*, *P5CS1*, *SOD*, *APX* and *CLC-C*. Furthermore, luciferase complementation and yeast one-hybrid assays showed that *SbWRKY50* directly binds to the promoters of *AtSOS1*, *SbSOS1* and *SbHKT1*. *SOS1* is mainly responsible for the efflux of Na^+ from roots (Wu et al. 1996).

The over-expression of *SOS1* results in an increase in the salt tolerance of *A. thaliana* (Yang et al. 2009) and decreases the Na^+ accumulation and increases K^+ in tobacco (Yue et al. 2012). Soybean *SOS1* improves salt tolerance by limiting the accumulation of Na^+ and increasing the activity of antioxidant enzymes (Zhao et al. 2017). Similarly, *HKT1* is a high affinity K^+ transporter responsible for transporting aboveground Na^+ back to the root. The over-expression of the soybean *HKT1* gene in tobacco can increase the K^+ content in transgenic plants and decrease the Na^+ content under salt-stress conditions. Thus, it plays an important role in regulating the Na^+/K^+ ratio in roots (Chen et al. 2014). After silencing the *HKT1* in tomato, the Na^+/K^+ ratio in the leaves changed, and the plants were susceptible to salt-related damage (Jaime-Pérez et al. 2017). In this study, the expression of *SOS1* and *HKT1* decreased more in transgenic plants. Corresponding with this result, the content of Na^+ increased and the K^+ content deceased in over-expression *SbWRKY50* plants relative to WT under salt stress. This suggests that over-expression of *SbWRKY50* can change ion homeostasis by directly regulating the expression of *SOS1* and *HKT1*.

In conclusion, *SbWRKY50* can regulate plant salt tolerance mainly by adjusting the Na^+ and K^+ content in roots. In *A. thaliana*, *SbWRKY50* can bind to the promoter of *AtSOS1*, while in sweet sorghum, *SbWRKY50* can directly bind to the promoters of *SbSOS1* and *SbHKT1*, which might explain why sweet sorghum is more tolerant to salt stress than *A. thaliana*. In addition, we speculate that *SbWRKY50* regulates the expression of osmotic stress- and oxidative stress-related genes by some indirectly pathways.

Materials and methods

Plant materials

Seeds of sweet sorghum M-81E were rinsed with running water for 2 h and placed in a wet dish for germination. After germination, the seedlings were transferred to a hydroponic pot and irrigated with 1/2 Hoagland's nutrient solution. When sorghum had grown to the three-leaf stage, they were treated with 0, 50, 100, 150 mM NaCl in Hoagland's nutrient solution for 48 h, and then the root tissues was removed and stored at − 80℃.

Full-length CDS cloning and bioinformatics analysis of the *SbWRKY50* gene

The primers SbW50 (Table S2) were designed using the CDS of *SbWRKY50* found on NCBI. The total RNA of the sweet sorghum root was extracted and reverse transcribed. The obtained cDNA was subjected to PCR amplification to obtain the desired target fragment. The ligation reaction was performed using the p*EASY*-Blunt Simple Cloning Kit Vector, and then positive clones were screened for sequencing. Bioinformatics analyses of *SbWRKY50* were performed using NCBI databases, DNAMAN and MEGA5.0.

Expression pattern of the *SbWRKY50* gene

The primers WRKY50 (Table S2) were designed according to the sequence of *SbWRKY50*. The shoot and root RNA of sweet sorghum were used independently as templates for real-time PCR.

The subcellular localization of *SbWRKY50*

The target fragment and 1300-GFP vector were double-digested with KpnI and BamHI and ligated. The ligation was verified to be correct, and the resulting plasmid was transformed into *Agrobacterium tumefaciens* EHA105. Then, the *Agrobacterium* transformation with WRKY50-1300-GFP was used to infect tobacco. An *Agrobacterium* transformation with only 1300-GFP was used as the control. After 2 days under normal culture conditions, the transformants were observed and photographed using two-photon confocal microscopy.

Transcriptional activation analysis in yeast cells

The target fragment and the pGBKT7 vector were digested with EcoRI and BamHI, then ligated and transformed into *Escherichia coli*. The AH109 yeast strain was transformed after correct digestion. The yeast strain transformed into pGBKT7 was used as the control. The transformed yeast solution was diluted by 1×, 10 × and 100×. Then, 10 μL of solution was dropped on SD/−Trp, SD/−Trp − Ade − His and SD/−Trp − Ade − His + X-a-gal, respectively. The samples were cultured at 28℃ for 48–96 h for observation and imaging.

Expression of *SbWRKY50* in transgenic *Arabidopsis*

The target fragment and pROKII vector were double digestion with KpnI and BamHI, ligated, and then transformed into *E. coli*. After confirming the correct construction, *A. tumefaciens* strain GV3101 was transformed. Homozygous *Arabidopsis* strains over-expressing *SbWRKY50* were obtained by infecting the inflorescence of *Arabidopsis* with *Agrobacterium* GV3101 transformed with WRKY50-pROKII. RNA was extracted from the roots of homozygous plants for reverse transcription, and the cDNA was used as the template for quantitative fluorescence PCR. The *actin2* gene in *Arabidopsis* was used as an internal reference. The primer sequences are shown in Table S2.

Determination of the seed germination rates and main root length

The seeds of WT and over-expression plants were washed three times with 75% alcohol for 3 to 4 min each time. They were sown on medium containing 1/2 MS, 1/2 MS + 50 mM NaCl, 1/2 MS + 100 mM NaCl and 1/2 MS + 150 mM NaCl. After vernalization at a low temperature for 3 days, seeds were cultured in a tissue culture room. After 24 h, the germination rate was calculated using the following formula:

$$\text{Germination rate} = (\text{number of seeds germinated}/\text{total number of seeds}) \times 100\%.$$

Then, the lengths of the main roots were photographed and measured.

Biomass determination

The seedlings of WT, W50-12 and W50-13 were independently treated with 0 and 100 mM NaCl in Hoagland's nutrient solution for 7 days. Then, the seedlings were removed from the soil, and washed with clean water and deionized water. The weights of the seedlings were recorded as fresh weights. The seedlings were then placed in an oven at 70℃ until their weight did not change. These weights were recorded as the dry weights. There were five replicates per treatment.

Determination of sodium ions (Na^+) and potassium ions (K^+) Contents

The seedlings of WT and over-expression plants were treated with 0 and 100 mM NaCl in Hoagland's nutrient solution. After 7 days, 0.1 g leaves and 0.1 g roots were sampled and placed into test tubes. Then, 5 mL of ddH_2O was added, and tubes were placed in boiling water for 3 h. Afterward, samples were filtered and brought to 10 mL. Then, the Na^+ and K^+ contents in leaves and roots were determined using a flame spectrophotometer. Each strain had six replicates per treatment.

DAB and NBT staining

The rosette leaves of WT and *SbWRKY50* over-expression plants of similar size were cut into EP tubes, and 2 mL DAB or NBT staining solution was added to the tubes to soak the leaves. Samples were placed in the dark for more than 12 h. Diaminobenzidine (DAB) or NBT dyes were replaced with bleaching solution (3:1:1 ethanol:acetic acid:glycerol). Bathed in boiling water for 10–15 min, then the bleaching solution was replaced and samples were incubated for 30 min. The color changes of leaves were observed and images taken. The experimental group receiving DAB staining was treated with 2 mL 10 mM Na_2HPO_4 as the control group, and the experimental group receiving NBT staining was treated with 2 mL 10 mM phosphate buffer as the control group. There were three repetitions in each group.

Quantitative real-time PCR of stress-related genes

To assess the effects of *SbWRKY50* expression, we investigated the expression profiles of several stress-response genes in WT and over-expression lines after 48 h of salt stress. Real-time PCR analyses were performed using cDNAs of WT and over-expression plants as templates. We studied the expression levels of the stress-related genes *AtRD29B*, *AtP5CS1*, *AtSOD*, *AtAPX*, *AtCLC-C*, *AtSOS1* and *AtHKT1*. Primers are shown in Table S2.

Yeast one-hybrid assays

In the pLacZi2u vector, EcoRI and XhoI sites were used for the insertion of promoter sequences. Full-length *SbWRKY50* was ligated into the EcoRI and XhoI sites of the GAD vector. The co-transformation of the two plasmids into yeast strain EGY48 were cultured on a SD/−Trp − Ura. The positive clones were cultured on SD/−Trp − Ura + X-a-gal. The results were observed and photographed after 3 to 5 days.

Luciferase complementation imaging assays

The target promoter and pGreen II-0800 vector were double-digested with KpnI and XhoI, and transformed into *E. coli*. The correct construct was transformed into *A. tumefaciens* GV3103 (psoup). The *Agrobacterium* solution containing pGreenII-0800 vector with a promoter and pMWB110 vector harboring *SbWRKY50* were mixed one to one and injected into the back of tobacco leaves. *Agrobacterium* containing pGreenII-0800 with a promoter and pMWB110 were mixed as the control. After 2 days of culturing under normal conditions, transformants were observed and live imaged by Lumina II (Xenogen, America).

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Compliance with ethical standards

Conflict of interest The authors declare no conflict of interest.

References

Ali MA, Azeem F, Nawaz MA, Acet T, Abbas A, Imran QM, Shah KH (2018) Transcription factors WRKY11 and WRKY17 are involved in abiotic stress responses in, *Arabidopsis* J Plant Physiol 226:12–21

Almansouri M, Kinet JM, Lutts S (2001) Effect of salt and osmotic stresses on germination in durum wheat (*Triticum durum* Desf.). Plant Soil 231:243–254

Asano T, Hayashi N, Kobayashi M, Aoki N, Miyao A, Mitsuhara I, Ichikawa H, Komatsu S, Hirochika H, Kikuchi S, Ohsugi R (2012) A rice calcium-dependent protein kinase OsCPK12 oppositely modulates salt-stress tolerance and blast disease resistance. Plant J 69:26–36

Bakshi M, Oelmüller R (2014) WRKY transcription factors: jack of many trades in plants. Plant Signal Behav 9:e27700

Birkenbihl RP, Kracher B, Somssich IE, Ziegler J, Liu S (2015) Negative regulation of ABA signaling by WRKY33 is critical for *Arabidopsis* immunity towards *Botrytis cinerea* 2100. e-Life 4:e07295

Cai R, Zhao Y, Wang Y, Lin Y, Cheng B (2014) Overexpression of a maize WRKY58 gene enhances drought and salt tolerance in transgenic rice. Plant Cell Tissue Org 119:565–577

Chen L, Song Y, Li S, Zhang L, Zou C, Yu D (2012) The role of WRKY transcription factors in plant abiotic stresses. Biochim Biophys Acta 1819:120–128

Chen HT, Chen X, Gu HP, Wu BY, Zhang HM, Yuan XX, Cui XY (2014) GmHKT1;4, a novel soybean gene regulating Na+/K+ ratio in roots enhances salt tolerance in transgenic plants. Plant Growth Regul 73:299–308

Diedhiou C, Golldack D (2006) Salt-dependent regulation of chloride channel transcripts in rice. Plant Sci 170:793–800

Ding L, Zhu JK (1997) Reduced Na+ uptake in the NaCl-hypersensitive SOS1 mutant of *Arabidopsis thaliana* Plant Physiol 113:795–799

Ding ZJ, Yan JY, Li GX, Wu ZC, Zhang SQ, Zheng SJ (2014) WRKY41 controls *Arabidopsis* seed dormancy via direct regulation of ABI3 transcript levels not downstream of ABA. Plant J 79:810–823

Gao HM, Wang YF, Xu P, Zhang ZB (2018) Overexpression of a wrky transcription factor TaWRKY2 enhances drought stress tolerance in transgenic wheat. Front Plant Sci 9:997

Guo YY, Tian SS, Liu SS, Wang WQ, Sui N (2017) Energy dissipation and antioxidant enzyme system protect photosystem II of sweet sorghum under drought stress. Photosynthetica 56:861–872

Guo YY, Song YS, Zheng HX, Zhang Y, Guo JR, Sui N (2018) NADP-malate dehydrogenase of sweet sorghum improves salt tolerance of *Arabidopsis thaliana* J Agric Food Chem 66:5992–6002

Han GL, Wang MJ, Yuan F, Sui N, Song J, Wang BS (2014) The CCCH zinc finger protein gene atzfp1 improves salt resistance in *Arabidopsis thaliana* Plant Mol Biol 86:237–253

Han Y, Fan T, Zhu X, Wu X, Cao S (2019) WRKY12 represses GSH1 expression to negatively regulate cadmium tolerance in *Arabidopsis* Plant Mol Biol 99:149–159

Hichri I, Muhovski Y, Žižková E, Dobrev PI, Gharbi E, Jose M, Franco-Zorrilla JM, Lopez-Vidriero I, Solano R, Clippe A, Errachid A, Motyka V, Lutts S (2017) The *Solanum lycopersicum* wrky3 transcription factor slwrky3 is involved in salt stress tolerance in tomato. Front Plant Sci 8:1343

Hu ZM, Yang X, Fromm ME (2006) Activation of the NaCl and drought-induced RD29A and RD29B promoters by constitutively active *Arabidopsis* MAPKK or MAPK proteins. Plant Cell Environ 29:1761–1770

Ibragimova SM, Trifonova EA, Filipenko EA, Shymny VK (2015) Evaluation of salt tolerance of transgenic tobacco plants bearing the P5CS1 gene of *Arabidopsis thaliana* Russ J Genet 51:1181–1188

Ishiguro S, Nakamura K (1994) Characterization of a cDNA encoding a novel DNA-binding protein, SPF1, that recognizes SP8 sequences in the 5' upstream regions of genes coding for sporamin and betaamylase from sweet potato. Mol Gen Genet 244:563–571

Jafarzadeh AA, Aliasgharzad N (2007) Salinity and salt composition effects on seed germination and root length of four sugar beet cultivars. Biologia 62:562–564

Jaime-Pérez N, Pineda B, García-Sogo B, Atares A, Athman A, Byrt CS, Belver A (2017) The sodium transporter encoded by the HKT1; 2 gene modulates sodium/potassium homeostasis in tomato shoots under salinity. Plant Cell Environ 40:658–671

Jiang YZ, Guo L, Liu R, Jiao B, Zhao X, Ling ZY, Luo K (2016) Overexpression of poplar ptrwrky89 in transgenic *Arabidopsis* leads to a reduction of disease resistance by regulating defense-related genes in salicylate- and jasmonate-dependent signaling. PLoS ONE 11:e0149137

Kim KC, Lai Z, Fan B, Chen Z (2008) *Arabidopsis* WRKY38 and WRKY62 transcription factors interact with histone deacetylase 19 in basal defense. Plant Cell Online 20:2357–2371

Lan A, Huang J, Zhao W, Peng Y, Chen Z, Kang D (2013) A salicylic acid-induced rice (*Oryza sativa* L.) transcription factor OsWRKY77 is involved in disease resistance of *Arabidopsis thaliana* Plant Biol 15:452–461

Lee H, Cha J, Choi C, Choi N, Ji HS, Park SR, Lee S, Hwang DJ (2018) Rice WRKY11 plays a role in pathogen defense and drought tolerance. Rice 11:5

Li J, Yan XF, Zu YG (2000) Generation of activated oxygen and change of cell defense enzyme activity in leaves of Korean pine seedling under low temperature. J Integr Plant Biol 42:148–152

Li JG, Pu LJ, Zhu M, Zhang R (2012) The present situation and hot issues in the salt-affected soil research. Acta Geogr Sin 67:1233–1245

Li JG, Pu LJ, Han MF, Zhu M, Zhang RS, Xiang YZ (2014a) Soil salinization research in China: advances and prospects. J Geogr Sci 24:943–960

Li MF, Guo SJ, Xu Y, Meng QW, Li G, Yang XH (2014b) Glycine betaine-mediated potentiation of HSP gene expression involved calcium signaling pathways in tobacco exposed to NaCl stress. Physiol Plant 150:63–75

Li YB, Li T, Han YY, Fan SX (2017) Cloning and function analysis of heat-shock-protein LsHsp70-2711 gene under high temperature stress in leaf lettuce (*Lactuca sativa* L.). Sci Agric Sin 8:12

Li J, Wang YQ, Yu B, Song QP, Liu Y, Chen THH, Li G, Yang XH (2018) Ectopic expression of StCBF1 and ScCBF1 have different functions in response to freezing and drought stresses in *Arabidopsis* Plant Sci 270:221–233

Liu W, Li RJ, Han TT, Cai W, Fu ZW, Lu YT (2015) Salt stress reduces root meristem size by nitric oxide-mediated modulation

of auxin accumulation and signaling in *Arabidopsis* Plant Physiol 168:343–356

Liu QN, Liu Y, Xin ZZ, Zhang DZ, Ge BM, Yang RP, Wang ZF, Yang L, Tang BP, Zhou CL (2017a) Genome-wide identification and characterization of the WRKY gene family in potato (*Solanum tuberosum*). Biochem Syst Ecol 71:212–218

Liu S, Wang WQ, Li M, Wan SB, Sui N (2017b) Antioxidants and unsaturated fatty acids are involved in salt tolerance in peanut. Acta Physiol Plant 39:207

López-Galiano MJ, González-Hernández AI, Crespo-Salvador O, Rausell C, Real MD, Escamilla M, García-Robles I (2018) Epigenetic regulation of the expression of WRKY75 transcription factor in response to biotic and abiotic stresses in Solanaceae plants. Plant Cell Rep 37:167–176

Ma CL (2002) cDNA cloning and gene expression of APX in *Suaeda salsa* in response to salt stress. Acta Pharmacol Sin 28:261–266

Pedro DV, Mohamed F, Gregorio BE, Lydia F, Cesar P, José AH, Lorenzo B (2013) Ectopic expression of cytosolic superoxide dismutase and ascorbate peroxidase leads to salt stress tolerance in transgenic plums. Plant Biotechnol J 11:976–985

Potschin M, Schlienger S, Bieker S, Zentgraf U (2014) Senescence networking: WRKY18 is an upstream regulator, a downstream target gene, and a protein interaction partner of WRKY53. J Plant Growth Regul 33:106–118

Reginato MA, Castagna A, Furlan A, Castro S, Ranieri A, Luna V (2014) Physiological responses of a halophytic shrub to salt stress by Na_2SO_4 and NaCl: oxidative damage and the role of polyphenols in antioxidant protection. AoB Plants 6:plu042

Rinerson CI, Rabara RC, Tripathi P, Shen QJ, Rushton PJ (2015) The evolution of WRKY transcription factors. BMC Plant Biol 15:66

Schnippenkoetter W, Lo C, Liu G, Dibley K, Chan WL, White J, Milne R, Zwart A, Kwong E, Keller B, Godwin I, Krattinger S, Lagudah ES (2017) The wheat Lr34 multipathogen resistance gene confers resistance to anthracnose and rust in sorghum. Plant Biotechnol J 15:1387–1396

Shao Q, Han N, Ding TL, Zhou F, Wang BS (2014) SsHKT1;1 is a potassium transporter of the C_3 halophyte *Suaeda salsa* that is involved in salt tolerance. Funct Plant Biol 41:790

Shi H, Quintero FJ, Pardo JM, Zhu JK (2002) The putative plasma membrane Na/H antiporter sos1 controls long-distance Na^+ transport in plants. Plant Cell 14:465–477

Shi H, Lee BH, Wu SJ, Zhu JK (2003) Overexpression of a plasma membrane Na^+/H^+ antiporter gene improves salt tolerance in *Arabidopsis thaliana* Nat Biotechnol 21:81

Si AJ, Yu Y, Chen H, Tian Q (2017) Functional prediction of stress response GhWRKY2 gene in cotton (*Gossypium hirsutum*). J Agric Biotechnol 25:222–230

Song YS, Li JL, Liu ML, Meng Z, Liu KC, Sui N (2019) The role of nitrogen on maize seedlings under drought stress. Funct Plant Biol 46:350–359

Sui N, Yang Z, Liu ML, Wang BS (2015) Identification and transcriptomic profiling of genes involved in increasing sugar content during salt stress in sweet sorghum leaves. BMC Genomics 16:534

Sui N, Tian SS, Wang WQ, Wang MJ, Fan H (2017) Overexpression of glycerol-3-phosphate acyltransferase from *Suaeda salsa* improves salt tolerance in *Arabidopsis* Front Plant Sci 8:1337

Sui N, Wang Y, Liu SS, Yang Z, Wang F, Wan SB (2018) Transcriptomic and physiological evidence for the relationship between unsaturated fatty acid and salt stress in peanut. Front Plant Sci 9:1–12

Sun C, Palmqvist S, Olsson H, Mats Borén Ahlandsberg S, Jansson C (2003) A novel WRKY transcription factor, SUSIBA2, participates in sugar signaling in barley by binding to the sugar-responsive elements of the iso1 promoter. Plant Cell 15:2076–2092

Syvertsen JP, Garcia-Sanchez F (2014) Multiple abiotic stresses occurring with salinity stress in citrus. Environ Exp Bot 103:128–137

Szekely G, Abraham E, Cseplo A, Rigo G, Zsigmond L, Csisza J, Ayaydin F, Strizhov N, Jasik J, Schmelzer E, Koncz C, Szabados L (2008) Duplicated P5CS genes of *Arabidopsis* play distinct roles in stress regulation and developmental control of proline biosynthesis. Plant J 53:18

Tian XJ, Li XF, Zhou WJ, Ren YK, Wang ZY, Liu ZQ, Tang JQ, Tong HN, Fang J, Bu QY (2017) Transcription factor OsWRKY53 positively regulates brassinosteroid signaling and plant architecture. Plant Physiol 175:1337–1349

Ülker B, Mukhtar MS, Somssich IE (2007) The WRKY70 transcription factor of *Arabidopsis* influences both the plant senescence and defense signaling pathways. Planta 226:125–137

Wang Y, Ying Y, Chen J, Wang X (2004) Transgenic *Arabidopsis* overexpressing Mn–SOD enhanced salt-tolerance. Plant Sci 167:671–677

Wang F, Chen HW, Li QT, Wei W, Li W, Zhang WK, Ma B, Bi YD, Lai YC, Liu XL, Man WQ, Zhang JS, Chen SY (2015) GmWRKY27 interacts with GmMYB174 to reduce expression of GmNAC29 for stress tolerance in soybean plants. Plant J 83:224–236

Wang L, Zhang XL, Wang L, Tian Y, Jia N, Chen S, Shi NB, Huang XM, Zhou C, Yu YW, Zhang ZQ, Pang XQ (2017) Regulation of ethylene-responsive SlWRKYs involved in color change during tomato fruit ripening. Sci Rep 7:16674

Wei DD, Cheng D, Liu WB, Liu T, Yang XH, Zheng YH (2015) Adequate potassium application enhances salt tolerance of moderate-halophyte *Sophora alopecuroides* Plant Soil Environ 61:364–370

Wei DD, Zhang W, Wang CC, Meng QW, Li G, Chen TH, Yang XH (2017) Genetic engineering of the biosynthesis of glycine betaine leads to alleviate salt-induced potassium efflux and enhances salt tolerance in tomato plants. Plant Sci 257:74–83

Wu SJ, Ding L, Zhu JK (1996) SOS1, a genetic locus essential for salt tolerance and potassium acquisition. Plant Cell 8:617–627

Wu H, Shabala L, Zhou M, Shabala S (2014) Durum and bread wheat differ in their ability to retain potassium in leaf mesophyll: implications for salinity stress tolerance. Plant Cell Physiol 55:1749–1762

Xu ZL, Raza Q, Xu L, He XL, Huang YH, Yi JX, Zhang DY, Shao HB, Ma HX, Ali Z (2018a) GmWRKY49, a salt-responsive nuclear protein, improved root length and governed better salinity tolerance in transgenic *Arabidopsis* Front Plant Sci 9:809

Xu Z, Raza Q, Xu L, He X, Huang Y, Yi J, Zhang D, Shao HB, Ma H, Ali Z (2018b) GmWRKY49, a salt-responsive nuclear protein, improved root length and governed better salinity tolerance in transgenic *Arabidopsis* Front Plant Sci 9:809

Yang Q, Chen ZZ, Zhou XF, Yin HB, Li X, Xin XF, Gong Z (2009) Overexpression of SOS (salt overly sensitive) genes increases salt tolerance in transgenic *Arabidopsis* Mol Plant 2:22–31

Yang Y, Chi Y, Wang Z, Zhou Y, Fan B, Chen Z (2016) Functional analysis of structurally related soybean GmWRKY58 and GmWRKY76 in plant growth and development. J Exp Bot 67:4727–4742

Yang Z, Wang Y, Wei XC, Zhao X, Wang BS, Sui N (2017) Transcription profiles of genes related to hormonal regulations under salt stress in sweet sorghum. Plant Mol Biol Rep 35:1–14

Yang Z, Zheng HX, Wei XC, Song J, Wang BS, Sui N (2018) Transcriptome analysis of sweet sorghum inbred lines differing in salt tolerance provides novel insights into salt exclusion by roots. Plant Soil 430:423–439

You LL, Song QP, Wu YY, Li SC, Jiang CM, Chang L, Yang XH, Zhang J (2019) Accumulation of glycine betaine in transplastomic potato plants expressing choline oxidase confers improved drought tolerance. Planta 249:1963–1975

Yue Y, Zhang M, Zhang J, Duan L, Li Z (2012) SOS1 gene overexpression increased salt tolerance in transgenic tobacco by maintaining a higher K^+/Na^+ ratio. J Plant Physiol 169:255–261

Zhang T, Tan D, Zhang L, Zhang X, Han Z (2017) Phylogenetic analysis and drought-responsive expression profiles of the wrky transcription factor family in maize. Agri Gene 3:99–108

Zhao D, Zhao JR, Huang X, Li N, Liu Y, Huang ZJ, Zhang ZY (2011) Functional analysis of TNBL1 gene in wheat defense response to barley yellow dwarf virus using BSMV-VIGS technique. J Crops 37:2106–2110

Zhao X, Wei P, Liu Z, Yu B, Shi H (2017) Soybean Na^+/H^+ antiporter GmsSOS1 enhances antioxidant enzyme activity and reduces Na^+ accumulation in *Arabidopsis* and yeast cells under salt stress. Acta Physiol Plant 39:19

Zheng LY, Guo XS, He B, Sun L, Peng Y, Dong S, Liu T, Jiang S, Ramachandran S, Liu C, Jing H (2011) Genome-wide patterns of genetic variation in sweet and grain sorghum (*Sorghum bicolor*). Genome Biol 12:R114

Zhou QY (2008) Soybean WRKY-type transcription factor genes, GmWRKY13, GmWRKY 21, and GmWRKY 54, confer differential tolerance to abiotic stresses in transgenic *Arabidopsis* plants. Plant Biotechnol J 6:486–503

Zhou L, Wang N, Gong S, Lu R, Li Y, Li X (2015) Overexpression of a cotton (*Gossypium hirsutum*) WRKY gene, GhWRKY34, in *Arabidopsis* enhances salt-tolerance of the transgenic plants. Plant Physiol Biochem 96:311–320

Zou X, Seemann JR, Neuman D, Shen QJ (2004) A WRKY gene from creosote bush encodes an activator of the abscisic acid signaling pathway. J Biol Chem 279:55770–55779

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