Effects of Tea Polyphenols on the Activities of Antioxidant Enzymes and the Expression of Related Gene in the Leaves of Wheat Seedlings Under Salt Stress

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Abstract

Longchun 30, a new wheat variety, was used to investigate seedling growth, element absorption and antioxidant response under 150 mM NaCl and tea polyphenols (TP) (25 and 100 mg L⁻¹) treatments alone or in combination, thus revealing TP-alleviating mechanism on the salt damage to plants. 150 mM NaCl stress alone inhibited the seedling growth, increased sodium content and reactive oxygen species (ROS) accumulation, but reduced potassium (K) and calcium (Ca) levels at different culture times, thus resulting in the oxidative damage to the leaves. Even though TP treatment alone led to the significant increase of ROS generation, TP-treated leaves exhibited the reduction of relative electrical conductivity and no change of malondialdehyde content. Moreover, high TP concentration alone stimulated the seedling growth. In addition, the activities and gene expression of superoxide dismutase, catalase and peroxidase (POD) as well as diamine oxidase and polyamine oxidase were changed to different degrees due to NaCl or TP treatment alone. Further study showed that the presence of TP promoted the seedling growth, increased K⁺ and Ca²⁺ contents, and led to the reduction of ROS accumulation. Taken together, salinity-inhibitory effect on the growth of wheat seedlings might be associated with salt-induced imbalance of element content and the increase of oxidative damage resulting from ROS accumulation, while the application of TP effectively alleviated salinity-inhibitory effect on the seedling growth and improved the tolerance of wheat seedlings to salt environment, which might be associated with the increases of K⁺ and Ca²⁺ contents as well as the reduction of oxidative damage in the leaves of wheat seedlings under NaCl and TP treatment in combination.

Introduction

Tea, a natural health drink originated in China for about 5,000 years, is one of the world's three largest non-alcoholic drinks, along with coffee and cocoa. Plant leaves are the natural sources of antioxidants such as pigments, phenolics and flavonoids (Sarker et al. 2018b; Sarker and Oba 2020a; Sarker and Oba 2020b). As the major chemical component in tea, tea polyphenols (TP) are the general term of polyphenols, including catechines, phenolic acids, flavonoids and anthocyanins. Among them, catechins in flavanols are the most important, and about 70% of TP are catechins (Khan and Mukhtar 2018). TP can be used as good antioxidant because of their ability to scavenge free radicals. Chen et al. (2016) reported that TP exhibited antioxidant properties and had a certain activation effect on antioxidant enzyme system of lychee fruits during storage, which could be used to delay the browning of fruits. In a recent study, we found that the addition of TP alleviated the accumulation of osmotic regulators in salt-treated wheat roots (Yang et al. 2017). The study of Si et al. (2020) indicated that TP application effectively blocked salinity-induced reduction of chlorophyll content and the transport efficiency of photosynthetic electron, which might help to strengthen photosynthetic ability and improve the tolerance of wheat seedlings to salt stress. Recently, Zhao et al. (2018) found that TP increased superoxide dismutase (SOD) activity but reduced glutathione (GSH) contents in a D-galactose-induced oxidative mouse model. However, no relevant studies have revealed that TP are involved in antioxidant stress when plants were exposed to salt environment.

Soil salinization is an increasingly serious environmental problem. About 7% of the world's land surface is threatened by salinization, affecting about 8.31×10^8 hectares of soil resources worldwide. At the same time, the area of secondary salinization is about 7.7×10^7 hectares, 58% of which is in irrigated agricultural areas (Huang et al. 2019). Soil salinization not only leads to the great reduction of agricultural output and the waste of land resources, but also causes the deterioration of ecological environment. Salinity disrupts most physiological progresses, thus inhibiting plant growth (Sarker and Oba 2018e; Sarker and Oba 2020b) and even reducing crop yield and quality (Almeida et al. 2017). Literatures showed that salinity treatment increased the amount of sodium (Na) and calcium (Ca) but decreased potassium (K) content in various plants (Sarker et al. 2018a; Zhang et al. 2018). Differently, the content of K and Ca decreased in the leaves and roots of maize as well as in the leaves of sorghum under salt stress (Huang 2018; Li et al. 2020). Thus, the regulation of Na^+ , K^+ and Ca^{2+} uptake in different plants and even different organs of the same plant is different in response to salt stress. There is no report about the effect of TP on the amount of Na^+ , K^+ and Ca^{2+} in plants tissues.

Besides, changes in reactive oxygen species (ROS) generation are one of the responses of plants to adversity stress. ROS mainly include singlet oxygen, superoxide anion ($\text{O}_2^{\cdot-}$), hydrogen peroxide (H_2O_2) and hydroxyl free radical ($\cdot\text{OH}$). Adversity stress can induce excessive ROS accumulation in plants, thus leading to oxidative damage. At the same time, extreme environmental may induce the changes of a variety of antioxidant enzymes, which is to resist oxidative damage and lethal effects in response to ROS accumulation (Hossain et al. 2017). Antioxidant enzymes, which are composed of SOD, peroxidase (POD) and catalase (CAT), can effectively remove high ROS concentration, prevent membrane peroxidation and maintain normal growth of plants under environment stress. Recent studies showed that salt stress caused the increased activities of antioxidant enzymes such as CAT, SOD and POD with the increase of salt concentration in various plants (Sarker and Oba 2018c; Yin et al. 2019; Datir 2020). Razavizadeh et al. (2020) observed that the activities of CAT and POD in *Carum copticum* L. seedlings increased first and then decreased with the increase of salt concentration. Hu et al. (2020) found that salt stress significantly increased POD activity in the leaves of some rice varieties, thus enhancing rice resistance to high salinity stress.

As the major staple crops around the world, wheat (*Triticum aestivum* L.) planting area is over 220 million hectares of soil, the annual output is nearly 750 metric tons (Balfourier et al. 2019). Soil salinization is the main restricting factor of semiarid ecosystem agriculture, which leads to the decline of soil fertility and restricts the sustainable development of agriculture (Szoboszlay et al. 2019). Therefore, it is of practical significance to study the damage mechanism of salt stress on plants or to find effective ways to alleviate salt stressed-induced damage. The new spring wheat variety Longchun 30, bred by the Wheat Research Institute of GanSu Academy of Agricultural Sciences, was mainly planted in some arid and saline areas such as JiuQuan, ZhangYe and WuWei in the HeXi corridor of GanSu Province, China (Si et al. 2020). Recently, different salt concentrations (25, 100 and 200 mM) were selected to treat wheat cultivar Longchun 30, and the results showed that 100 and 200 mM salt concentration induced significant inhibition of seedling growth (Xu et al. 2020). In this study, wheat Longchun 30 was used to investigate

the effects of TP on the balance of Na⁺ and K⁺, antioxidant response and the expression of related enzyme genes under 150 mM NaCl stress, aiming to reveal the regulatory mechanisms of TP to alleviate salinity-induced stress damage and provide the theoretical basis for further elucidating the salt resistance mechanism of wheat seedlings.

Materials And Methods

Culture and treatments of experimental materials

Wheat Longchun 30 seeds were purchased from GanSu Academy of Agricultural Sciences. TP was provided by GanSu Characteristic Plant Effective Ingredient Products Engineering Technology Research Center.

Wheat seeds were surface-sterilized with 0.1% HgCl₂ for 10 min, rinsed with running water for 10 h, and germinated overnight in darkness. Plump and uniformly germinated seeds were selected and placed neatly in the petri dishes. The seedlings were treated with 1/4 Hoagland solution containing 0, 150 mM NaCl, 25 or 100 mg L⁻¹ TP, 150 mM NaCl + 25 mg L⁻¹ TP or 150 mM NaCl + 100 mg L⁻¹ TP, with at least three replicates for each treatment. The seedlings were cultured at 25 ± 0.5°C and 12 h light/12 h dark under a light irradiance of 300 μmol m⁻² s⁻¹ in the incubator.

After 6 days of growth, 30 wheat seedlings in each group were randomly selected, the length of their shoots and roots was measured. In addition, the above-ground and under-ground parts of the seedlings were washed with deionized water, sucked the surface water with a filter paper and weighed the fresh weight (Fw). And then, dry weight (Dw) was weighted after plant material was dried the seedlings at 105°C for 10 min and then at 80 °C for 6 h.

Determination Of Relative Electrical Conductivity And Lipid Peroxidation

Seedling leaves of the same size were thoroughly rinsed with distilled water, the surface water was sucked with filter paper, and three parts of the same mass were quickly weighed and soaked in distilled water at 25°C for 24 h respectively and the initial conductivity (S1) was measured with conductivity meter. Then it was bathed in boiled water for 30 min and taken out. After it cooled and shaken well. The final conductivity (S2) was measured with distilled water as the reference. The relative electrical conductivity (REC) was calculated as follows: REC% = {(S1/S2)×100%}.

The method of Li et al. (2014) was employed to detect the amount of malondialdehyde (MDA). Wheat leaves (0.5 g) were homogenized with the extracted solution containing 0.25% thiobarbituric acid (TBA) and 10% trichloroacetic acid (TCA). The mixture was bathed in boiled water for 30 min, cooled

immediately and centrifuged at $1000 \times g$ for 10 min. The absorption value of the obtained supernatant was detected at wavelengths of 450, 530 and 600 nm.

Measurement Of Element Content

The amount of Na^+ , K^+ and Ca^{2+} in wheat leaves were determined by referring to Achary et al. (2012). Wheat leaves cleaned with deionized water and dried naturally. The dry sample was cut into pieces, put into the microwave digestion tank containing 8 mL concentrated nitric acid (HNO_3), and then acidified for 1.5 h. After the sample solution was cooled, the acid was transferred to the crucible for further heating. The residue was dissolved with 1 M HNO_3 for a constant volume of 50 mL. Na^+ , K^+ and Ca^{2+} content was measured with atomic absorption spectrophotometer.

Analyses Of Antioxidant Enzyme Activities

0.5 g of plant material was ground with 1 mL phosphate buffer (PBS) (50 mM, pH 7.8) containing 0.1 mM ethylenediaminetetraacetic acid disodium salt (EDTA-Na_2) and 1% polyvinyl pyrrolidone (PVP). After the homogenate was centrifugated at $15000 \times g$ for 30 min, and the supernatant was used to detect antioxidant enzyme activities.

SOD activity was measured with reference to the method of Sarker and Oba (2018c). The reaction mixture consisted of 50 μL enzyme extract and 50 mM PBS (pH 7.8) containing 13 mM methionine, 0.1 mM EDTA-Na_2 and 75 M nitrotetrazolium blue chloride (NBT). And then, riboflavin with a final concentration of 2 μM was added to initiate the reaction. After it was illuminated for 30 min at 25°C , the absorbance was recorded at 560 nm. One unit of SOD was defined as the amount of enzyme in the condition that NBT reduction inhibition was 50%, and SOD activity was expressed as units U mg^{-1} protein.

CAT activity was detected according to Abdel et al. (2019). In 3 mL 50 mM PBS buffer (pH 7.0), 100 μL of enzyme extract was added and incubated at 25°C for 5 min. Then the reaction was started with 40 μL 0.05% H_2O_2 , and the scan was performed at an interval of 20 s at 240 nm for 2 min. CAT activity was expressed as units U mg^{-1} protein.

POD activity was measured based on the guaiacol method of Rao et al. (1996). 5 μL enzyme extract was added to 3 mL enzyme reaction solution containing 50 mM PBS buffer (pH 7.0) and 20 mM guaiacol, and the reaction was initiated with 40 μL 0.05% H_2O_2 . The scan was manipulated at an interval of 20 s at 470 nm for 2 min. POD activity was expressed as units U mg^{-1} protein.

Detection Of Relative Gene Expression

Total RNA extraction

Wheat leaves were ground in liquid nitrogen, and 1 mL RNAiso Plus reagent was added. The solution was shaken well and placed at room temperature for 10 min. After the solution was centrifuged at $12000 \times g$ for 10 min at 4°C, the supernatant was transferred to the centrifuge tube and then 200 µL chloroform was added. After being shaken well and placed for 10 min, it was centrifuged again, the supernatant was transferred and the equal amount of isopropyl-ketone was added. It was placed at room temperature for 10 min and the white precipitate was obtained by centrifugation. 1 mL 75% alcohol containing diethyl pyrocarbonate (DEPC) was added and it was centrifuged at $8000 \times g$ for 5 min at 4°C. The precipitate was collected, and 50 µL RNase-free water was added to dissolve the RNA.

The synthesis of cDNA was performed using the PrimeScript™ RT reagent Kit and gDNA Eraser (Perfect Real Time) Kit.

RT-PCR amplification system and conditions

The reverse transcriptional cDNA due to different treatments was used as the template and *Triticum aestivum* glyceraldehyde-3-phosphatedehydrogenase (*TaGAPDH*) was selected as the official gene. The reaction system was configured according to the instructions of SYBR Premix Ex Taq™ Kit of TaKaRa company. All reactions were performed under the following condition: Pre denaturation at 95°C for 30 s, 40 cycles of denaturation at 95°C for 5 s, and annealing at 60°C for 20 s; From 55°C to 95°C, there were 81 cycles with 0.5 increase per 30 s. Three parallel samples were set for each sample, the relative expression level was calculated with the $2^{-\Delta\Delta C_t}$ method and the real-time quantitative PCR experiment was carried out three times.

Wheat related sequences were searched on NCBI, and gene-specific primers (Table 1) were designed by using Primer Premier 5.0 software in combination with the design requirements of real-time fluorescent quantitative PCR primers, and then sent to Shanghai ShengGong Biological Engineering CO., LTD for synthesis.

Table 1
Primers used for RT-qPCR

Primer	Serial number	Sequence(5'-3')
<i>TaGAPDH</i>	AF251217	F: TTAGACTTGCGAAGCCAGCA R: AAATGCCCTTGAGGTTTCCC
<i>TaCu/ZnSOD</i>	TAU69632	F: TGTCGATAGCCAGATTCCTTTGA R: AGTCTTCCACCAGCATTTCCAG
<i>TaMnSOD</i>	AF092524	F: CTGGAAGAACCTCAAGCCTATC R: GATCCTTGTAAGCACCTCTG
<i>TaCAT</i>	E16461	F: CAACCACTACGACGGGCTCA R: GGGCTGCTTGAAGTTGTTCTCC
<i>TaPOD</i>	AY857755	F: CTTGACAACGCCTACTACAC R: GGTTGGACGCAAAGTTCC

Analysis Of Ros Content

The detection of $O_2^{\cdot-}$ content was based on the method of Kiba et al. (1997). 0.5 g of wheat leaves was added to 50 mM Tris-HCl buffer solution (pH 6.4) containing 0.2 mM NBT, 0.2 mM nicotinamide adenine dinucleotide (NADH) and 250 mM sucrose. The mixture solution was vacuum-infiltrated for 10–15 min and illuminated for 24 h to develop color. The absorbance was determined at 530 nm and the amount of $O_2^{\cdot-}$ was expressed as $\mu\text{mol g}^{-1}$ Fw.

The method of Sarker and Oba (2018d) was used to analyze of H_2O_2 level. Wheat leaves (0.5 g) were homogenized with 5 mL 0.1% TCA. After the homogenate was centrifuged at $12000 \times g$ for 20 min, 0.7 mL supernatant were added to 0.7 mL 10 mM PBS and 1.4 mL 1 M potassium iodide. The absorbance was measured at 390 nm and the unit of H_2O_2 level was expressed in ng g^{-1} Fw.

$\cdot\text{OH}$ content was determined by the reference to Halliwell et al. (1987). 0.5 g of plant materials was homogenized with 10 mM PBS buffer (pH 7.4) containing 2 mM 2-Deoxy-D-ribose, and the homogenate was centrifuged at $12000 \times g$ for 15 min. The obtained supernatant was added to 0.5% TBA and 1 mL glacial acetic acid containing 2.5 mM NaOH. The reaction mixture was boiled for 30 min, the absorbance was measured at 532 nm. $\cdot\text{OH}$ content was expressed as nmol g^{-1} Fw.

Measurement of diamine oxidase (DAO) and polyamine oxidase (PAO) activities

DAO activity was determined according to Naik et al. (1981). 1 g of wheat leaves was ground with 50 mM PBS buffer (pH 7.0) containing 20 mM guaiacol, and the homogenate was centrifuged at $16000 \times g$ for 20 min. The supernatant was added into 50 mM PBS (pH 7.8) containing 0.1 mM pyridoxal phosphate and 10 mM humic acid (HA), and incubated at 30°C for 1 h. The reaction was stopped with 1 mL 20% TCA. 30 min later, the reaction mixture was centrifuged at $5000 \times g$ for 15 min. The supernatant was added to 1 mL of the intermixture containing 250 mg of ninhydrin, 6 mL of acetic acid and 4 mL of phosphoric acid, and the intermixture was boiled for 30 min. Finally, 1 mL of glacial acetic acid was added and the absorbance was determined at 510 nm.

PAO activity was detected by referring to Asthir et al. (2002). 1 g of wheat leaves was homogenized with 2 mL 100 mM PBS buffer (pH 7.0) containing 5 mM dithiothreitol, and the homogenate was centrifuged at $16000 \times g$ for 20 min. After the supernatant was removed, the precipitate was washed with 100 mM PBS buffer (pH 7.0) containing 1 mM NaCl, and then it was centrifuged at $16000 \times g$ for 20 min. The reaction mixture consisted of 450 μ L enzyme solution, 0.1% 2-aminobenzaldehyde, 10 mM spermidine and 50 U CAT. It was incubated for 3 h at 30°C, the reaction was terminated with 1 mL 10% perchloric acid, and then it was centrifuged for 10 min at $6500 \times g$. The absorbance at 430 nm was determined with the supernatant.

Determination Of Protein Content

According to Bradford (1976) method, bovine serum albumin (BSA) was used as the standard to calculate protein content.

Data analysis

All values were replicated at least three times, and the data was expressed as the mean $\pm$ standard error. Statistical analyses were performed with the SPSS 17.0 software. The data were then analyzed using one-way analysis of variance (ANOVA) followed by Duncan's multiple comparison test. The differences were considered significant at a probability level of $P \leq 0.05$.

Results

Changes of biomass, REC and lipid peroxidation in wheat seedlings under different treatments

As indicated in Fig. 1a, the shoots and roots of wheat seedlings were significantly shorter when wheat seedlings were subjected to 150 mM NaCl stress. In comparison with the control, 25 mg L⁻¹ TP treatment resulted in the notable reduction of shoot length and the insignificant change of root length. Shoot and root lengths in 100 mg L⁻¹ TP-treated seedlings increased to 111% and 129% of the control, respectively. When 25 or 100 mg L⁻¹ TP was added, the length of shoots and roots in salt-treated seedlings increased significantly compared with those of the seedlings treated with salt stress alone.

Compared with the control, the Fw of leaves and roots in response to salt treatment decreased by 61% and 47%, and the Dw decreased by 50% and 30%, respectively (Fig. 1b and 1c). 25 mg L⁻¹ TP treatment alone did not affect the Fw of leaves and roots, but increased the Dw. By contrast, these parameters increased to 1.18, 1.13, 1.14 and 1.16 times of the control, respectively, under 100 mg L⁻¹ TP treatment alone. Compared with salt treatment alone, the Fw of leaves and roots as well as the Dw of leaves increased significantly in salt stressed seedlings with the addition of TP at different concentrations. However, TP application had no obvious effect on the Dw of roots under NaCl treatment (Fig. 1b and 1c).

A significant elevation of REC due to 150 mM NaCl stress was observed, whereas different TP concentrations resulted in the notable decrease, with about 37% and 27% reduction due to 25 mg L⁻¹ or 100 mg L⁻¹ TP treatment, respectively, in wheat leaves in comparison with the control. The addition of 25 mg L⁻¹ or 100 mg L⁻¹ TP effectively weakened the salinity-induced enhancement of REC, by about 34% and 19% decrease compared with salt stress alone, respectively (Fig. 2a).

As shown in Fig. 2b, MDA content increased by about 78% in the leaves of salt-stress seedlings, in comparison with untreated seedlings, and this parameter was not affected by different TP concentrations (25 and 100 mg L⁻¹). Leaf MDA content due to 25 mg L⁻¹ TP + 150 mM NaCl and 100 mg L⁻¹ TP + 150 mM NaCl treatments decreased by about 17% and 20% in comparison with salt stress alone.

Changes of Na⁺, K⁺ and Ca²⁺ content in wheat leaves under different treatments

Compared with the control, leaf Na⁺ content increased to 2.74 times in wheat seedlings treated with 150 mM NaCl. Additionally, low TP concentration did not affect leaf Na content, whereas a large amount of Na⁺ was found due to high TP concentration (Fig. 3a). Compared with salt stress alone, the addition of 25 and 100 mg L⁻¹ TP effectively alleviated the salt-induced increase in leaf Na⁺ level, and the reduction was about 31% and 24%, respectively.

150 mM NaCl or 25 mg L⁻¹ TP treatment alone decreased K⁺ content in wheat leaves by 71% and 66% of the control, respectively. Nevertheless, 100 mg L⁻¹ TP treatment alone did not affect K⁺ content. Compared with salinity-alone-treated seedlings, leaf K⁺ content in NaCl-stressed ones enhanced significantly by about 38% and 36% in the presence of 25 or 100 mg L⁻¹ TP, respectively (Fig. 3b).

In comparison with the untreated seedlings, leaf Ca²⁺ content decreased by 35% under 150 mM NaCl stress. Differently, 25 and 100 mg L⁻¹ TP treatment resulted in the significant elevation of Ca²⁺ content by 13% and 38%, respectively. In addition, leaf Ca²⁺ level in response to 25 mg L⁻¹ TP + 150 mM NaCl and 100 mg L⁻¹ TP + 150 mM NaCl treatments notably increased by about 45% and 51% compared with that under salt stress alone (Fig. 3c).

Changes of antioxidant enzyme activities in wheat leaves under different treatments

As shown in Fig. 4a, leaf SOD activity increased initially, followed by decrease with the extension of growth time of wheat seedlings. Compared with the control, the activity of SOD was not affected by salt stress alone on the 2 days, and obviously increased to 123% and 113% of the control group on the 4 and 6 days. This enzyme activity in response to 100 mg L⁻¹ TP treatment alone significantly decreased in comparison with the control, but did not alter under 25 mg L⁻¹ TP treatment on the 2 and 4 days, whereas notably reduced for 6 days. Compared with salt treatment alone, the presence of two TP concentrations had no significant effect on the change of leaf SOD activity induced by salt stress.

Compared with the control, leaf CAT activity decreased by about 30% and 14%, respectively, when the seedlings were stressed with 150 mM NaCl on the 2 and 4 days, but there was no evident change on the 6 days (Fig. 4b). In addition, leaf CAT activity under 25 mg L⁻¹ TP treatment alone for 2 days significantly decreased, but notably increased by about 28% and 23% under the treatment with 100 mg L⁻¹ TP for 4 and 6 days. In comparison with salt stress alone, the addition of 25 mg L⁻¹ TP did not affect salt-induced change of CAT activity, while the further decrease in this parameter was found in wheat leaves treated by salinity together with 100 mg L⁻¹ TP, which was reduced by about 10%, 19% and 38% on the 2, 4 and 6 days, respectively (Fig. 4b).

In comparison with the control, the activity of POD in wheat leaves reduced with the extension of salt treatment time, and decreased by about 31%, 40% and 11% on the 2, 4 and 6 days, respectively (Fig. 4c). Moreover, the changes of leaf POD activity under TP treatment were similar to those under salt stress. For example, this enzyme activity under 25 mg L⁻¹ TP treatment on the 2, 4 and 6 days decreased by about 31%, 53% and 32%, respectively, compared with the control. The presence of two TP concentrations did not affect salinity-induced change of POD activity on the 2 days, but this enzyme activity significantly increased by about 45% and 40% at 4 days as well as 27% and 28% on the 6 days, respectively, due to NaCl + 25 mg L⁻¹ TP and NaCl + 100 mg L⁻¹ TP treatment in comparison with salt stress alone.

Changes of antioxidant enzyme gene expression in wheat leaves under different treatments

As shown in Fig. 5a, the expression level of *TaCu/Zn-SOD* significantly increased when the seedlings exposed to NaCl stress alone on the 2 and 4 days, which were 1.68 and 1.34 times higher than those of the control, whereas its expression was significantly lower than that of the control on the 6 days. Compared with the control of the same period, the expression of *TaCu/Zn-SOD* did not change significantly in response to 100 mg L⁻¹ TP treatment. Compared with salt stress alone in the same period, this parameter expression increased by about 19% on the 2 days after NaCl + TP treatment, and the change of the gene expression on the 4 days was not significant, but it was significantly lower at the 6 days of treatment.

It can be seen from the Fig. 5b that the expression of *TaMn-SOD* under salt treatment alone increased significantly to 149%, 238% and 156% of the control on the 2, 4 and 6 days, respectively. This gene expression increased to 1.08 times as much as that of the control in wheat leaves when treated with TP on the 2 days, whereas the effect of TP treatment of this gene significantly reduced on the 4 and 6 days.

NaCl + TP treatment had no effect on the expression of *TaMn-SOD* on the 2 days, while it was reduced by about 53% and 67% of the salt treatment alone on the 4 and 6 days.

Compared with the control, the expression of *TaCAT* in wheat leaves reduced by about 12%, 22% and 53% on the 2, 4 and 6 days of salt treatment alone, respectively (Fig. 5c). This parameter expression tended to decrease with the increase of TP treatment time, but it was remarkably higher than control 1.98 and 1.39 times on the 2 and 4 days, respectively, whereas reduced about 41% on the 6 days. In comparison with the control, the presence of TP did not affect the expression of *TaCAT* on the 2 days, but resulted in notable reduction of this gene expression on the 4 and 6 days by about 23% and 39%.

As shown in Fig. 5d, exposure to salt for 2 days did not affect *TaPOD* expression, but remarkably inhibited on the 4 and 6 days, with reduction by about 51% and 77% compared with the control, respectively. The *TaPOD* expression in wheat leaves evidently decreased due to 100 mg L⁻¹ TP treatment, which was 0.71, 0.72 and 0.34 times of that of the control on the 2, 4 and 6 days. Compared with salt treatment alone, the application of 100 mg L⁻¹ TP significantly increased this gene expression on the 2, 4 and 6 days by about 41%, 86% and 143%, respectively.

Changes of ROS content in wheat leaves under different treatments

As can be seen from the Table 2, with the extension of growth time, leaf O₂^{•-} production in unstressed seedlings decreased gradually. The amount of O₂^{•-} in wheat leaves increased with salt stress alone, which was 1.58, 1.16 and 1.53 times of the control values on the 2, 4 and 6 days, respectively. Compared with the control, leaf O₂^{•-} content increased by about 11% when the seedlings were exposed to 25 mg L⁻¹ TP on the 2 days, while did not significantly change on the 4 and 6 days. Differently, this parameter in response to 100 mg L⁻¹ TP treatment significantly rose, with about 56%, 39% and 15% enhancement on the 2, 4 and 6 days, respectively. In addition, 25 mg L⁻¹ TP did not affect NaCl-induced changes of O₂^{•-} production in wheat leaves, whereas the addition of 100 mg L⁻¹ TP remarkably reduced O₂^{•-} accumulation on the 4 and 6 days of salt treatment.

Table 2
Changes of reactive oxygen species (ROS) content in wheat leaves under different treatments

	Treatments	Two days	Four days	Six days
O ₂ ^{•-} content (μmol g ⁻¹ Fw)	CK	2.05 ± 0.15 ^a	1.60 ± 0.10 ^a	1.43 ± 0.01 ^a
	150 mM NaCl	3.23 ± 0.09 ^c	1.86 ± 0.13 ^b	2.19 ± 0.01 ^c
	25 mg L ⁻¹ TP	2.28 ± 0.12 ^b	1.59 ± 0.15 ^a	1.51 ± 0.02 ^{ab}
	100 mg L ⁻¹ TP	3.20 ± 0.05 ^c	2.22 ± 0.15 ^c	1.65 ± 0.03 ^b
	150 mM NaCl + 25 mg L ⁻¹ TP	3.02 ± 0.17 ^c	1.83 ± 0.17 ^b	2.25 ± 0.03 ^c
	150 mM NaCl + 100 mg L ⁻¹ TP	3.06 ± 0.25 ^c	1.63 ± 0.12 ^a	1.43 ± 0.01 ^a
H ₂ O ₂ content (ng g ⁻¹ Fw)	CK	347.05 ± 1.96 ^b	453.36 ± 1.11 ^a	536.17 ± 2.05 ^a
	150 mM NaCl	601.23 ± 1.36 ^d	872.62 ± 1.25 ^e	819.34 ± 5.80 ^d
	25 mg L ⁻¹ TP	310.38 ± 1.82 ^a	779.36 ± 1.67 ^d	613.21 ± 3.29 ^b
	100 mg L ⁻¹ TP	466.05 ± 1.19 ^c	681.48 ± 1.26 ^c	587.30 ± 2.24 ^{ab}
	150 mM NaCl + 25 mg L ⁻¹ TP	345.66 ± 1.57 ^b	753.67 ± 2.09 ^d	708.12 ± 6.59 ^c
	150 mM NaCl + 100 mg L ⁻¹ TP	498.44 ± 1.04 ^{cd}	552.14 ± 2.57 ^b	614.60 ± 5.60 ^b
·OH content (nmol g ⁻¹ Fw)	CK	27.94 ± 1.39 ^a	53.31 ± 1.79 ^a	72.23 ± 2.36 ^b
	150 mM NaCl	34.23 ± 1.62 ^b	69.12 ± 1.85 ^{bc}	93.91 ± 3.72 ^c
	25 mg L ⁻¹ TP	31.20 ± 1.06 ^b	65.04 ± 1.72 ^b	54.68 ± 2.40 ^a
	100 mg L ⁻¹ TP	28.62 ± 1.09 ^{ab}	75.98 ± 1.95 ^c	52.32 ± 2.92 ^a
	150 mM NaCl + 25 mg L ⁻¹ TP	30.80 ± 1.64 ^{ab}	67.82 ± 1.56 ^{bc}	87.06 ± 2.05 ^c
	150 mM NaCl + 100 mg L ⁻¹ TP	28.28 ± 1.10 ^{ab}	72.28 ± 2.21 ^c	55.26 ± 2.96 ^a
Values represent mean ± SE of at least three experiments with replicated measurements; Different small letters mean significant difference at 0.05 levels. TP: Tea polyphenols				

Leaf H_2O_2 level due to salt or TP treatment alone showed a trend of increased initially, followed by decrease with the extension of seedlings growth time, and reached the peak on the 4 days of growth (Table 2). In comparison with the control, 150 mM NaCl stress induced a notable elevation in H_2O_2 content, which was 173%, 192% and 153% of the control on the 2, 4 and 6 days, respectively. After the seedlings were exposed to 25 mg L^{-1} TP alone on 2 days, the amount of H_2O_2 showed no obvious change, while on the 4 and 6 days, this parameter enhanced to 1.72 and 1.14 times of the control. Differently, leaf H_2O_2 content due to 100 mg L^{-1} TP treatment on the 2 and 4 days increased to 1.34 and 1.50 times, whereas 100 mg L^{-1} TP was treated on the 6 days, there was no significant change in this parameter, in comparison with that to the control. Additionally, when 25 mg L^{-1} TP was added on the 2, 4 and 6 days, leaf H_2O_2 content in salt-stressed seedlings decreased by about 43%, 14% and 14%, respectively, in comparison with salinity-alone stressed ones. Similar, the application of 100 mg L^{-1} TP on the 2, 4 and 6 days, also decreased this parameter in salinity-stressed seedlings.

Leaf $\cdot\text{OH}$ production in untreated seedlings enhanced with the extension of plant growth time. This parameter due to 150 mM NaCl treatment enhanced by about 23%, 30% and 30% on the 2, 4 and 6 days, respectively, in comparison with the control. Similarly, leaf $\cdot\text{OH}$ content elevated by about 12% and 22% due to 25 mg L^{-1} TP treatment on the 2 and 4 days, respectively, as well as increased by about 43% to 100 mg L^{-1} TP on the 4 days. Wheat seedlings treated by both TP concentrations on the 6 days, exhibited in the reduction of leaf $\cdot\text{OH}$ content. In addition, the application of TP on the 2 and 4 days did not affect leaf $\cdot\text{OH}$ production in salt-treated seedlings, but salt-induced $\cdot\text{OH}$ generation significantly reduced at 6 days of 100 mg L^{-1} TP treatment, compared with salt treatment alone (Table 2).

Changes of DAO and PAO activities in wheat leaves under different treatments

As shown in Table 3, DAO activity in response to 150 mM NaCl stress alone notably increased on the 4 and 6 days, but no obviously changed on the 2 days, in comparison with the control. In the early and middle stage of 25 mg L^{-1} TP treatment (2 and 4 days), leaf DAO activity decreased initially, followed by an increase. Differently, in the middle and late stage of 100 mg L^{-1} TP treatment (4 and 6 days), this parameter showed the changes of increased initially, followed by decrease, in comparison with the control. Compared with the salt treatment alone, leaf DAO activity in salt-treated seedlings decreased by 51%, 33% and 35% in the presence of 25 mg L^{-1} TP on the 2, 4 and 6 days, respectively. After the seedlings were treated with NaCl + 100 mg L^{-1} TP on the 2 days, the activity of DAO reduced by about 32%, but did not significantly alter on the 4 and 6 days, compared to salt treatment alone.

Table 3
Changes of diamine oxidase (DAO) and polyamine oxidase (PAO) activities in wheat leaves under different treatments

	Treatments	Two days	Four days	Six days
DAO activity (U mg ⁻¹ protein)	CK	0.84 ± 0.07 ^c	1.07 ± 0.02 ^a	2.97 ± 0.09 ^c
	150 mM NaCl	0.91 ± 0.01 ^c	1.79 ± 0.01 ^c	3.39 ± 0.01 ^d
	25 mg L ⁻¹ TP	0.67 ± 0.02 ^b	1.71 ± 0.09 ^c	2.95 ± 0.04 ^c
	100 mg L ⁻¹ TP	0.83 ± 0.04 ^c	1.92 ± 0.06 ^c	1.95 ± 0.02 ^a
	150 mM NaCl + 25 mg L ⁻¹ TP	0.45 ± 0.02 ^a	1.20 ± 0.01 ^b	2.27 ± 0.03 ^b
	150 mM NaCl + 100 mg L ⁻¹ TP	0.62 ± 0.04 ^b	1.85 ± 0.01 ^c	3.38 ± 0.03 ^d
PAO activity (U mg ⁻¹ protein)	CK	5.79 ± 0.44 ^e	3.73 ± 0.30 ^c	1.76 ± 0.02 ^{bc}
	150 mM NaCl	5.65 ± 0.27 ^{de}	4.62 ± 0.29 ^{cd}	2.31 ± 0.01 ^d
	25 mg L ⁻¹ TP	4.63 ± 0.17 ^c	2.70 ± 0.24 ^a	2.05 ± 0.04 ^{cd}
	100 mg L ⁻¹ TP	5.16 ± 0.19 ^d	3.15 ± 0.33 ^b	1.28 ± 0.03 ^a
	150 mM NaCl + 25 mg L ⁻¹ TP	3.04 ± 0.25 ^a	4.01 ± 0.33 ^d	1.60 ± 0.07 ^b
	150 mM NaCl + 100 mg L ⁻¹ TP	4.09 ± 0.10 ^b	3.85 ± 0.12 ^c	2.08 ± 0.03 ^{cd}
Values represent mean ± SE of at least three experiments with replicated measurements; Different small letters mean significant difference at 0.05 levels. TP: Tea polyphenols				

Leaf PAO activity of 150 mM NaCl stress on the 2 days showed no significant change, but significantly increased by about 24% and 31% on the 4 and 6 days, compared with that of the control. Treatment with 25 mg L⁻¹ TP alone on the 2 and 4 days resulted in significant inhibition of leaf PAO activity, while this parameter decreased to 89%, 84% and 73% of the control on the 2, 4 and 6 days of 100 mg L⁻¹ TP treatment, respectively. The application of TP effectively alleviated salinity-induced effect on the activity of PAO. For example, when 25 mg L⁻¹ TP was added on the 2, 4 and 6 days, leaf PAO activity in salt-treated seedlings decreased by about 46%, 13% and 31%, respectively, in comparison with salt-alone-stressed ones (Table 3).

Discussion

Changes of seedlings growth and ion content under different treatments

Soil salinization is one of the major abiotic stresses in worldwide agricultural production. Salt environment can change the physiological, biochemical and molecular characteristics of plants, which eventually affects plant growth and reduces crop production. Growth inhibition and biomass reduction are the main response of plants to salinity environment (Guo et al. 2017; Abdel et al. 2019; Datir et al. 2020; Sarker and Oba 2020b). The present study supported this viewpoint, because 150 mM NaCl stress inhibited root and shoot growth of wheat seedlings, and reduced both Fw and Dw of leaves and roots. There were no reports about the influence of TP on plant growth and development. In this study, we also observed that the growth of wheat seedlings was promoted by 100 TP mg L⁻¹ treatment alone, and in particular, 100 mg L⁻¹ TP significantly increased the Fw and Dw of leaves and roots. In addition, the presence of TP partly blocked the growth inhibition and the biomass reduction in salt-stressed wheat seedlings, suggesting that TP application might enhance the tolerance of wheat seedlings to salt environment.

Salt-induced inhibition of plant growth can be attributed to ion toxicity. The normal growth of plants is inseparable from the balanced absorption of various ions in the vivo. Excessive salt ions in the environment may compete with nutrient elements, thus affecting the selective absorption of nutrient elements. It was found that different plants exposed to salt stress exhibited the increased level of cellular Na⁺ and restricted the absorption of K⁺ (AbdElgawad et al. 2016; Guo et al. 2017; Sarker et al. 2018a; Jadamba et al. 2020). In the present study, salinity stress alone increased Na⁺ content but significantly decreased K⁺ level in wheat leaves. Similarly, the amount of Na⁺ increased but K⁺ content decreased in response to different NaCl concentrations in the roots and leaves of various plants (Batista et al. 2019; Ma et al. 2021; Nazar et al. 2011). These changes in Na⁺ and K⁺ contents could destroy the K⁺/Na⁺ ratio and resulted in the disruption of the distribution of mineral elements in plant tissues, which might also cause salt damage and affect plant growth. Ca²⁺ is a large element necessary for the normal growth and development of plants. High salinity can also exert its effects on plants by regulating Ca²⁺ concentration (Halperin et al. 1997). Increased Ca²⁺ content in plants during salt stress has been reported by different authors (Manishankar et al. 2018; Todea et al. 2020). In a previous study, we also observed a significant elevation of total Ca in wheat roots after NaCl exposure (Yang et al. 2007). In contrast to these conclusions, Huang et al. (2020) observed that increasing NaCl concentration resulted in a fast decrease in the amount of Ca²⁺ in rice. The present data showed that Ca²⁺ content in wheat leaves significantly decreased due to salinity stress alone, but increased to treatment with TP alone. Epstein and Rains (1987) believed that the Ca content became insufficient and could not meet the need of the normal growth of plants under salt stress, which might be related to salt-induced inhibition of absorption and utilization of Ca²⁺ in plants. The damage of environment stress to plants was partly due to the obstruction of Ca signaling system (Lynch et al. 1989). Furthermore, Ca²⁺ can effectively resist the toxic effect of Na⁺ (Tester and Davenport 2003) and maintain the balance of plant mineral ions (Kurusu et al. 2013). Therefore, imbalance of Na⁺, K⁺ and Ca²⁺ contents in the leaves might be the reason why salt stress significantly inhibited the growth of wheat seedlings, and the increase of Ca²⁺ content might be associated with TP-induced promoting effect on seedling growth. Additionally, the reduction of Na⁺

accumulation in plant leaves has been considered as an important mechanism of tolerance against salt stress (Tester and Davenport 2003). There are few reports about the effect of TP on the regulation of various ions contents in plant tissues. In the present study, even though 100 mg L^{-1} TP led to significant increase in the amount of Na^+ and 25 mg L^{-1} TP reduced K^+ level, the addition of two TP concentrations reduced the amount of Na^+ but increased both K^+ and Ca^{2+} contents in the leaves of salt-treated seedlings, which might be related to the alleviating effects of TP on salt-induced inhibition of seedling growth. And also, exogenous salicy acid (SA) treatment alone resulted in a decrease of K^+ content, but NaCl and SA stress in combination strongly induced the increase of K^+ content in both leaves and roots of *Egletes viscosaboth* (Batista et al. 2019). Differently, high SA (1.0 mM) concentration alone did not influence Na^+ content, but 1.0 mM SA + NaCl treatment increased this parameter (Nazar et al. 2011). It can be seen that the transportation of Na^+ and K^+ in plants is very complex. Why the changes of Na^+ and K^+ content in wheat leaves are opposite due to TP treatment alone and in combination with salt, which needs to be further studied.

Changes of oxidative damage and ROS accumulation under different treatments

Salinity stress can disrupt ROS metabolism system and cause excessive ROS accumulation, which may lead to oxidative damage of plant cells. Oxidative damage is associated with the aggravation of membrane lipid peroxidation and the leakage of a large number of organic matter and inorganic ions in plant cells (Ren et al. 2020). MDA, the product of membrane lipid peroxidation, is commonly used to mark oxidative stress when plants are subjected to abiotic stress. REC was also related to oxidative damage in plants exposed to salt environment (Sarker and Oba 2018d; Yu et al. 2020). Salinity stress has been reported to disturb the integrity of cell membranes in wheat associated with the increase of membrane permeability and the decrease of lipid partiality (Guo et al. 2017). Previous studies have indicated that these parameters are often consistent with stress-induced ROS accumulation (Guo et al. 2017; Chen et al. 2020). The effect of salt stress in this study was consistent with these studies. This was because salt treatment alone caused the significant increases of MDA content and REC, which was accompanied by the accumulation of ROS including $\text{O}_2^{\cdot-}$, $\cdot\text{OH}$ and H_2O_2 (Table 2). These results suggested that excessive ROS generation induced by salt stress resulted in oxidative damage to wheat seedlings. Differently, TP-induced ROS accumulation in wheat leaves was not accompanied by oxidative damage, because no significant change of MDA content and the reduction of REC were observed when wheat seedlings were treated with TP alone. These results seemed to indicate that TP treatment alone might enhance the tolerance of wheat seedlings to excessive ROS generation in leaf tissue. Consistent with this study, Paul et al. (2017) found that spermidine or salt application alone induced H_2O_2 accumulation, but that the combination treatments of both significantly decreased this parameter. The study of Chen et al. (2014) also indicated that H_2O_2 contents increased in response to salt stress alone, but decreased by adding sodium nitroprusside in the leaves of NaCl-treated *A. corniculatum*.

Even though H_2O_2 content is generally considered to be an indicator of oxidative stress caused by abiotic stress (Yen et al. 2013), $\cdot\text{OH}$ is more toxic to biological tissues than H_2O_2 . This is because $\cdot\text{OH}$ is the

known ROS with the highest reactivity, and there are no enzymatic mechanisms to eliminate this highly reactive ROS (Xie et al. 2019). Furthermore, $\bullet\text{OH}$ is considered to be the main reason for mediating oxidative damage in vivo (You and Chang 2015). In this study, although the addition of TP at different concentrations led to the decrease of H_2O_2 content and the elevation of $\text{O}_2^{\cdot-}$ and $\bullet\text{OH}$ accumulation in salt-stressed seedlings, it effectively alleviated salt stress-induced increases of MDA content and REC in wheat leaves. These results further indicated that TP might provide plant tissues with a protective pathway different from ROS scavenging to alleviate salt stress-caused oxidative damage to membrane tissues. At the same time, the present data also indicated that there might be differences between the TP-alone-induced protective mechanism on biofilms and the TP-alleviated effect on salt-induced oxidation damage. Whether TP directly acts on biofilms or indirectly enhances the resistance of biofilms to excessive ROS accumulation, this needs to be further explored.

Change of enzyme activities and related gene expression associated with different treatment-induced ROS accumulation

ROS accumulation in plant tissues is related to scavenging systems such as antioxidant enzymes and to complex ROS production networks. In addition to increased SOD activity, salt stress resulted in the accumulation of H_2O_2 in wheat leaves resulted from weakening the activities of H_2O_2 -scavenging enzymes POD and CAT and from enhancing the activities of H_2O_2 -producing enzymes PAO and DAO. Similar to some of the present results, significant decreases of POD and CAT activities were found in wheat and rice under salt stress (Guo et al. 2017; Jadamba et al. 2020). However, salinity treatment significantly increased the activities of these two enzymes in various plants (Ali et al. 2017; Abd Allah et al. 2017; Sarker and Oba 2018e; Yin et al. 2019). When wheat seedlings were exposed to 25 mg L^{-1} TP, the accumulation of H_2O_2 was contributed to the reduction of POD activity, but not to the decrease of PAO and DAO activities. Differently, 100 mg L^{-1} TP treatment alone led to increased H_2O_2 generation in wheat leaves by reducing POD activity and increasing PAO and DAO activities. Therefore, the way of ROS accumulation in wheat leaves due to NaCl or TP treatment alone might be different. At the same time, we also found that the presence of different TP concentrations effectively blocked salinity-inhibitory effect on POD activity and salinity-excitatory effect on PAO and DAO, which was also the reason why TP application partly reduced the amount of H_2O_2 in response to salinity stress. POD is believed to be the main enzyme that protects cells from H_2O_2 damage in plants (Rodrigues et al. 2020). Furthermore, salt stress-induced stimulation of CAT and POD is considered the evidence for the salt tolerance of wheat varieties (Datir et al. 2020). Therefore, TP-induced increase of POD activity as well as decreases of PAO and DAO activities suggested an important defense mechanism against lipid peroxidation in wheat leaves under salinity treatment, weakening the oxidative damage caused by salt stress.

Several studies have indicated that overexpression of SOD, CAT and POD genes can confer resistance to various abiotic stresses, such as chilling, salinity, drought and heavy metals, in plants (Li et al. 2010; Kaouthar et al. 2016; Han et al. 2019). It was considered that the elevated expression of antioxidant system components has been linked to the decrease of oxidative damage under different stresses (Wang

et al. 2010). For example, the calli of *P. pratensis* suffered and survived from the salinity-induced oxidative stress by increasing the expression of antioxidant enzymes gene (Luo et al. 2020). In this experiment, except for down-regulating *TaCu/ZnSOD* expression at 6 days, the expression of *TaCu/ZnSOD* and *TaMnSOD* up-regulated in response to salinity stress alone, suggesting that the increase of SOD activity was due to the expression of these genes in the leaves of wheat seedlings under salinity stress. Similarly, an increase in SOD activity was directly related to the expression of *Cu/ZnSOD* and *MnSOD* in rice exposed to salinity (Mishra et al. 2013). Additionally, the present data showed that salinity stress resulted in the significant down-regulation of *TaCAT* and *TaPOD* expression in wheat leaves in a concentration-dependent manner. Furthermore, the reduction of CAT and POD activities was consistent with the changes of related gene expression in the leaves of salinity-stress seedlings, indicating that the activities of CAT and POD might be regulated at the transcriptional levels. In Kentucky bluegrass (*Poa pratensis* L.) cultivars, the expression of CAT and POD genes also down-regulated in response to salinity stress (Puyang et al. 2015). Differently, the expression of *Cu/ZnSOD* showed no significant change, while CAT gene expression up-regulated in leaves of rice with the extension of salt treatment time (Rossatto et al. 2017). Qian et al. (2020) also observed that salt stress promoted the activity of antioxidant enzymes and up-regulated the expression of *CAT* and *POD* in *Gladiolus gandavensis* seedlings. Therefore, the expression of antioxidant enzyme genes due to salt stress was different in various plants. There was no report about the effect of TP treatment on the expression of antioxidant enzyme genes. In this study, 100 mg L⁻¹ TP treatment alone did not affect the expression of *TaCu/ZnSOD*, while the decreases of *TaMnSOD* and *TaPOD* expression was greater with the extension of treatment time, which was consistent with the change trend of SOD and POD activities. Differently, *TaCAT* expression up-regulated in wheat leaves at 2 and 4 days of 100 mg L⁻¹ TP treatment alone but down-regulated at 6 days. In addition, the presence of TP significantly reduced the expression of NaCl-induced *TaCu/ZnSOD* and *TaMnSOD*, but did not affect the expression of *TaCAT* in the leaves of early salt-treated seedlings. Furthermore, the addition of TP effectively alleviated salt-inhibitory effect on the expression of *TaPOD*, which might be the reason why TP enhanced POD activity in salt-treated wheat leaves.

Conclusion

In conclusion, salt stress inhibited the growth of wheat seedlings, which might be related to the nutrient imbalance of Na⁺, K⁺ and Ca²⁺ as well as the oxidative damage resulting from excessive ROS accumulation in wheat leaves under salt treatment. Salt-induced ROS accumulation was associated with the inhibition of H₂O₂-scavenging enzymes POD and CAT and with the stimulation of SOD and H₂O₂-producing enzymes PAO and DAO. Differently, TP-induced ROS accumulation was accompanied by the reduction of REC, no change of MDA content and the increase of Ca²⁺ level, implying that TP treatment alone might provide a protective mechanism different from salt stress against ROS accumulation in wheat seedlings. In addition, TP application partly blocked the nutrient imbalance, the oxidative damage resulting from excessive ROS accumulation and the inhibition of POD activity and gene expression in wheat leaves under salinity stress, thus enhancing the tolerance of wheat seedlings to salt environment and facilitating the growth of salt-treated seedlings.

Declarations

Ethics approval and consent to participate (Not applicable)

Consent for publication (Not applicable)

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

YZ performed the detection of biomass, relative conductivity, malondialdehyde and element content, was a major contributor in writing the manuscript. GYL and LBS detected the changes of antioxidant enzyme activities and related gene expression in wheat under different treatments. NL was involved in the modification of the manuscript. YLY and TPG mainly participated in the design of experimental scheme and also provided corresponding technical support. In addition, YLY was the corresponding author of this manuscript.

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Figures

Figure 1

Growth, Fresh weight (FW) and Dry weight (DW) in wheat seedlings exposed to different treatments. Bars are the means of five replicates $\pm$ standard deviation ($n=5$), and difference letters on bars indicate significant difference ($p < 0.05$). TP: Tea polyphenols

Figure 2

Malondialdehyde (MDA) content and relative electrical conductivity (b) in wheat leaves exposed to different treatment. Bars are the means of five replicates $\pm$ standard deviation ($n=5$), and difference letters on bars indicate significant difference ($p < 0.05$). TP: Tea polyphenols

Figure 3

Changes of sodium (Na), potassium (k), and calcium (Ca) contents in wheat leaves in response to different treatments. Bars are the means of five replicates $\pm$ standard deviation ($n=5$), and difference letters on bars indicate significant difference ($p < 0.05$). TP: Tea polyphenols

Figure 4

Superoxide dismutase (SOD), catalase (CAT) and peroxidase (POD) activities in wheat leaves under different treatments. Bars are the means of five replicates $\pm$ standard deviation ($n=5$), and difference letters on bars indicate significant difference ($p < 0.05$). TP: Tea polyphenols

Figure 5

The expression of related antioxidant enzymes genes in wheat leaves under different treatments. Bars are the means of five replicates $\pm$ standard deviation ($n=5$), and difference letters on bars indicate significant difference ($p < 0.05$). TP: Tea polyphenols