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Tolerant potato cultivar selection under multiple abiotic stresses

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Abstract

Potato is one of the most important food crops. Numbers of potato genotypes have been cultivated and widely cultured, especially in arid and semi-arid lands, hence, the resistance ability of potato is vital to fight against environmental stresses. In this experiment, we aimed to test five potato cultivars from different regions under drought, salt, high temperature and oxidative stresses in order to select the cultivar which could possess stronger resistance capability when subjected to one or more stresses. According to our result, the Chinese cultivar named LongShu NO.3 (cv. LS) is tolerant to drought, salt and oxidative stress as it hashigher SOD and APX activities, stronger rooting ability and more root dry weight in PEG or NaCl treated groups; however, it is sensitive to high temperature. While the cultivar named ´Superior´ is rather sensitive to drought and salt, compared to other cultivars, its tolerance to high temperature is best with steady growth status, Fv/Fm and water content in leaf, under 42°C for 24 h. The rich anthocyanin contained cultivar JaYoung showed an overall excellent resistant ability to all stresses, and the important antioxidant, anthocyanin, might have played a crucial role.

Key words: Abiotic stress, potato, selection, tolerance.

Introduction

Environmental stresses, including drought, temperature, salinity, air pollution, heavy metals, pesticides and soil pH, are major limiting factors in crop production that affect almost all plant functions [1]. Usually, drought, salt, extreme temperature and oxidative stresses are related to each other, and induce similar cellular damage. For instance, as the typical abiotic stress, drought and salt both cause disorder in cell and in ion distributions [2, 3]. Heat stress induces significant changes in normal physiological processes, such as photosynthesis dark respiration, membrane stability and mitochondrial respiration [4], and oxidative stress, which always couples to other abiotic stresses, always leading to the denaturation of protein structure and function [5]. When plants are exposed to these stresses, the reactive oxygen species (ROS) is generated and it, in turn, has a negative oxidative stress effect on cellular structures and metabolism [6].

In order to adapt to these environmental stresses, a series of biochemical and morphological changes occur spontaneously in plants. Under normal conditions, there is a sustainable balance between the reactive oxygen species (ROS) that was produced by plant cells and their scavenging system. While plants are suffering from environmental stresses, the ROS produced exceeds its scavenging system, resulting in the oxidative damage and impairment of cell functioning [7, 8]. As a defense system, plants in turn enhance the activation of enzymatic and non-enzymatic antioxidants to combat with the stresses. The enzymatic mechanisms include specific antioxidant enzymes, such as superoxide dismutase (SOD), ascorbate peroxidase (APX) and peroxidase (POD), catalase (CAT). The non-enzymatic system contains low-molecular-weight antioxidants, such as ascorbic acid, glutathione, carotenoids and phenolic compounds [9, 10]. SOD converts superoxide radicals (O^{2-}) into hydrogen peroxide (H_2O_2), POD reduces H_2O_2 to water using various substrates as electron donors, APX uses ascorbate as an electron donor to reduce H_2O_2 to water, and CAT dismutates H_2O_2 into water and oxygen. Numerous studies indicate that the activity of antioxidant enzymes is correlated with plant tolerance to abiotic stresses, including drought, high salt and extreme temperature.

Potato is the fourth most important food crop in the world after rice, wheat and maize, and is the only major food crop that is a tuber. Potato is a very efficient food crop and produces more dry matter, protein and minerals per unit area, in comparison to cereals[11]. Potatoes are widely cultivated in many countries, but always in arid and semi-arid regions, where shortage or poor water quality are major factors limiting plant growth and yield [12]. Potato is often considered to be a drought sensitive crop, and it also has been classified as moderately salt-sensitive to moderately salt-tolerant[13]. As numbers of potato genotypes have been cultivated in different regions, different genotypes obtain various agronomic characteristics and show different resistance abilities to stresses.

In this experiment, we took five potato genotypes from different regions and subjected them to different abiotic stresses, including drought, salt, high temperature and oxidative stress, with the aim of selecting the cultivar with stronger resistance ability to specific stresses.

Materials and Methods

Five potato genotypes and their features: Potato (*Solanum tuberosum* L.) cultivars of cv.*LongShu No.3* (Chinese cultivar, in short: LS) and cv. Favorite (Netherlandish cultivar, in short: FR) were stocked in our laboratory (State Key Laboratory of Soil Erosion and Dryland Farming on Loess) in China, and the other cultivars including cv. JaYoung (Korean cultivar, in short JY), cv. Atlantic, (American cultivar, in short AT), and cv. Superior (American cultivar, in short: SP) were provided by Doctor KWAK SangSoo from Korea Research Institute of Bioscience & Biotechnology

Potato propagation: Potato plants were propagated via sub-culturing of shoot tips and stem nodal sections every 3-4 weeks on MS basal medium containing 3% sucrose. All the potato sterile plates were kept in the culture room at 16/8 h light cycle, with a light intensity of 350 $mmol.m^{-2}.s^{-1}$ and a relative humidity of 45% at 25°C.

In order to generate the whole potato plant, we excised the plants in advance, hence, the 30 d old sterile seedlings of each potato cultivar were transferred from MS medium to sterilised soil with full nutrition. Two weeks later, all the individuals were transferred to pots and cultured in a green house with similar environmental conditions. The experiment was composed of different individual experiments (Please check this line again) involving drought, salt, high temperature and MV stress.

PEG treatment: In order to emulate drought stress conditions, the terminal buds from sterile potato seedlings were transferred into MS medium, containing PEG 6000 (polyethylene glycol, Sigma, USA)-infused plates according to a modified version of the method reported in Verslues *et al.* [14]. PEG-infused plates were prepared by dissolving solid PEG in a sterilised solution of half-strength MS medium with 2 mM MES buffer (pH 5.7), followed by overlaying of the PEG solution onto agar-solidified half-strength MS medium plates containing 7% Phyto agar. The agar medium and PEG solution were equilibrated for at least 12 h before the excess PEG solution was removed. Drought stress strength was expressed as the concentration of the overlayed PEG solution: 0% (control) or 2% (drought stress). In order to avoid the osmotic response induced by the high concentration of sucrose, no sucrose was contained in the half-strength MS medium. Each cultivar under each concentration of PEG followed a 15 times repetition. Samples were collected at 10 d and 20 d after the first transfer and then deep-frozen immediately at -80°C before being taken to assay the activities of antioxidant enzymes and Lipid peroxidation.

Extraction and assays of the activities of reactive oxygen-scavenging enzymes: Leaf samples (0.1 g) were frozen in liquid nitrogen and immediately crushed into powder then dissolved in 1 ml of 50 mM potassium phosphate buffer (pH 7.0) that contained 1 mM EDTA, 1 mM ascorbic acid (ASA), 1mM dithiothreitol (DTT), 1mM L-glutathione (GSH) and 5 mM $MgCl_2$. After the homogenate was centrifuged at 20,000 rmp for 15 min at 4°C, the resultant supernatant was deep-frozen at -80°C and used for assays of enzymatic activity. Total protein concentration was determined according to the Bradford method [15] using the Bio-Rad protein assay reagent.

The activity of superoxide dismutase (SOD) was measured according to McCord and Fridovich [39] with slight modification, by immediately monitoring the absorbance at 560 nm due to the reduction of cytochrome *c*. The reaction mixture contained 50 mM phosphate buffer (pH 7.8), 0.1 mM nitrotetrazolium blue chloride (NBT), 0.1 mM EDTA and 13.37 mM methionine.

APX activities were determined with the reaction mixture containing 800 mM HEPES buffer, 50 mM AsA and 100 mM H_2O_2, the reaction was initiated by adding 50 µl plant extract, the APX activities were determined by following the consumption of H_2O_2 at 290 nm for 90 s.

POD activities were determined specifically at 420 nm. The reaction mixture contained 0.4 ml of 100 mM potassium phosphate buffer (pH 6), 0.16 ml of 147 mM H_2O_2, 0.32 ml of 5% pyrogallol and 2.1 ml of DW. The reaction was initiated by adding 20 µl plant extract and after 10 min the POD activity was determined by following the consumption of H_2O_2 (extinction coefficient 39.4 mM^{-1} cm^{-1}) at 420 nm for 20 s.

The activity of catalase (CAT) was assayed by monitoring decreases in absorbance at 240 nm due to the decomposition of H_2O_2. The reaction mixture contained 670 µl potassium phosphate buffer (pH 7.0), 330 µl H_2O_2 and 30 µl of the extract. The CAT activity was determined by following the consumption of H_2O_2 (extinction coefficient 39.4 mM^{-1} cm^{-1}) at 240 nm for 1 min.

Lipid peroxidation: Lipid peroxidation was determined as the amount of malondialdehyde (MDA, e =155 mM±1 cm±1), a product of lipid peroxidation. Of saved supernatant (which has also been used to determinate antioxidant enzyme) 200 µl was mixed with 600 µl reaction buffer, including 5% trichloroacetic acid (TCA) and 0.5% thiobarbituric acid (TBA), and was heated in 100°C water for 15 min, then cooled immediately and centrifuged at 10,000 *g* for 10 min. The absorbance was monitored at 450, 532 and 600 nm.

NaCl treatment: To emulate salt stress, half strength MS medium including 3% sucrose and 7% Phyto agar with 0 (control) or 80 mM NaCl were used. The terminal buds from sterile potato seedlings were transferred to tubes containing MS medium. Each cultivar in each group of NaCl treatment also followed 15 times repeat. Samples were collected at 10 d and 20 d after the first transfer and then deep-frozen immediately at -80°C. Root fresh weight and dry weight were measured and the activities of antioxidant enzymes were assayed as described above.

High temperature treatment: For high temperature stress, five-week-old potato plants growing at 25°C growth chamber were transferred to 42°C for 24 h in the growth chamber. Treated plants were transferred to normal conditions (25°C, 100 mol m^{-2} s^{-1}) for a 12 h period of recovery from the stress. The tolerance of potato plants to high temperature stress was estimated as the photosynthesis activity (Fv/Fm) and the fresh weight of the plants after treatment . The fifth leaves of plants were used to determine the photosynthesis activity with a portable chlorophyll fluorescence meter (Handy PEA, Hansatech, England) and water content at 0, 24 h after treatment and 12 h after recovery from the stress. Water content was estimated by determining the fresh weight before stress, the weight immediately after stress and the dry weight of each sample. Each cv. was repeated 5 times.

MV stress and ion leakage analyses: To assay the oxidative stress tolerance of each cultivar, leaf discs (8 mm in diameter) from 5-week-old plants of each cultivar were taken into plates that contained solution with 0.4% sorbitol and 3 μM MV, as described by Kwon *et al.* [16]. Ion leakage of solution was assessed using an ion conductivity meter (model 455C, Isteck Co., Seoul, Korea).

Results

The tolerance ability of different potato cultivars to drought stress in PEG containing MS medium: To investigate the change of activities of antioxidant enzymes of potato cultivars under drought stress, we analysed the activities of SOD, APX, CAT and POD of each cultivar at 10 d and 20 d that had grown in 0% and 2% PEG contained in 1/2 MS medium. The rooting abilities of each cultivar in medium with 2% PEG were shown in (Fig. 1A), from which we can see that the potato cv. LS and JY can still root in PEG contained in medium, but the other three cultivars were seriously inhibited from rooting. The change of SOD activities in all potato cultivars showed a similar trait (Fig. 1B), that at 10 d in treatment duration they all increased with 2% PEG, compared with the control group, especially cv. LS and cv. JY (the increase rates were 23% and 24%, respectively). However, no significant difference could be found between treatments, 20 d-0% PEG group and 20 d-2% PEG group. The most significant increase in APX activities (Fig. 1C) showed in cv.LS at 10 d (280%) and at 20 d (68%), followed by cv. AT and cv. SP, the increase rates at 10 d and 20 d were 86%, 67% and 78%, 84% respectively. The change tendency of CAT activities showed variously in different cultivars (Fig. 1D), the most significant increases were at 10 d in cv. AT (70%) and in cv. SP (30%), however, except for these two cultivars, the CAT activities all declined, with 2% PEG treatment compared with control treatment. POD activities (Fig. 1E) all increased in the 2% PEG treatment group, compared with the control group. The most significant increase rate of POD activities was obtained by cv. AT, with 81% increase at 10 d and 64% at 20 d, then the 72% increase rate in cv. SP at 10 d and the 30% increase rate in cv. JY at 20 d were also remarkable.

MDA contents have also been analysed in all the potato cultivars at 10 d and 20 d under drought stress (Fig. 2). The MDA content showed almost no change in LS, which was followed by AT, except the increase in 20 d - 2% treatment, the MDA contents in AT were almost at the same level. In cultivar of SP, the MDA contents increased gradually, along with the treatment duration and with, or without, PEG but not significantly. The MDA contents in FR also ascended, and more dramatically, compared with the former three cultivars, which might imply the more serious membrane lipid peroxidation. It is interesting to notice that JY had a very high MDA content, especially the initial MDA content at 10 d of control treatment. Moreover, in 20 d - 2% treatment the MDA content also increased significantly, JY having the highest MDA content among all the cultivars.

The tolerance ability of different potato cultivars to salt stress in NaCl contained MS medium: Salt stress can be imposed by irrigating soil-grown plants with saline solutions or by transferring seedlings or plants to salt-containing media [14]. Here, we simulated salt stress by transfering seedlings to the MS media, that contained 0 mM and 80 mM NaCl, and assayed the activities of antioxidant enzymes. The SOD activities and APX activities (Fig. 3A- B) all increased in 80 mM NaCl treatment compared with control treatment in all potato cultivars, and each cultivar also had different performance. The maximum increase rate of SOD activity was 90% in cv. LS at 10 d, followed by cv. JY with 48%, while at 20 d the maximum increase rate was 40% in cv. JY and about 26% in both cv. LS and cv. AT. At 10 d, the maximum increase rate of APX activities was 205% in cv. LS, then cv. FR with 131%. At the 20 d, the increase rates were higher in cv. AT, JY and LS, and lower in FR and SP. The minimum increase rates of both SOD and APX were possessed by cv. SP. The maximum increase of CAT activities (Fig. 3C) were in cv. LS (84%) and cv. JY (75%) at 10 d, while in cv. AT and SP, the CAT activities declined with added NaCl in media. It was expected that the activity of CAT dropped most significantly in cv. SP. The POD activities (Fig. 3D) all increased at 10 d in NaCl treatment and at 20 d the increase trait remained, except for cv. LS and JY. Potato cv. AT and cv. SP obtained the

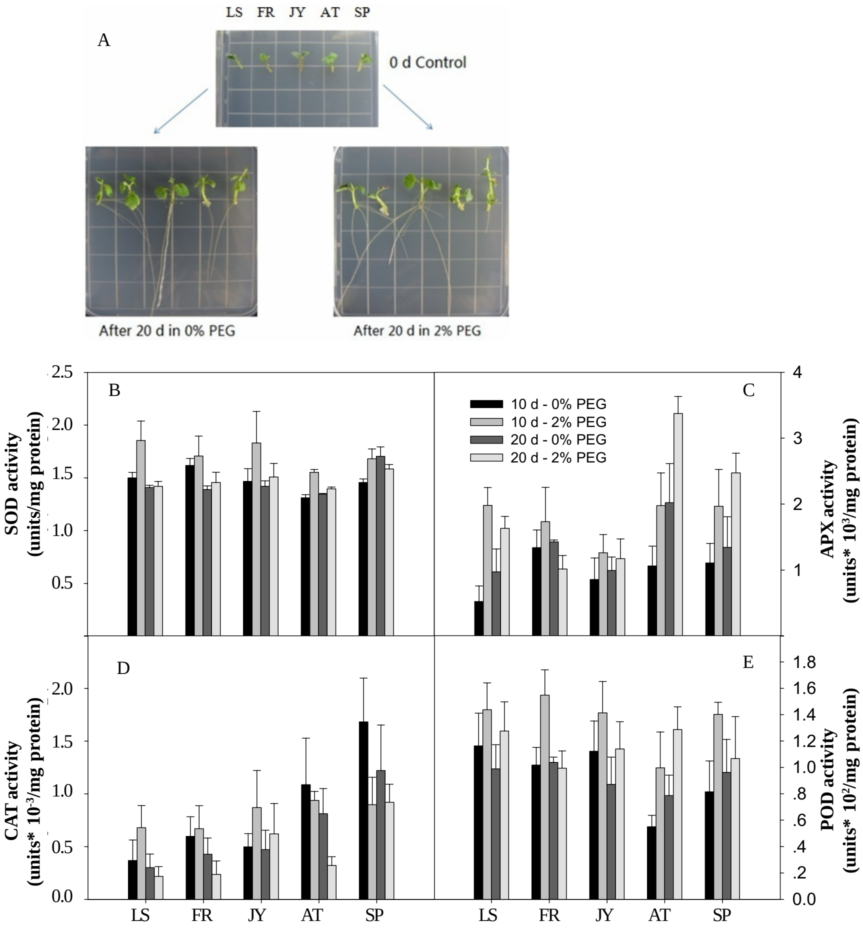

Figure 1. Potato cultivars grown in 1/2 MS medium contained PEG (0% and 2%). The root growth of each cultivar after 20 d of transfer (A); the SOD activities (B), the APX activities (C), the CAT activities (D) and the POD activities (E) at 10 d and at 20 d with/without PEG. Data are expressed as the mean ± standard deviation (SD) of five replicates.

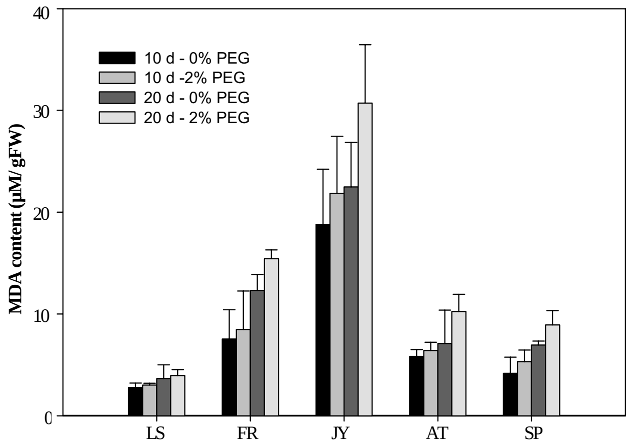

Figure 2. The MDA contents of each potato cultivars after 10 or 20 days transferred to PEG (0% or 2%) containing 1/2MS medium. Data are expressed as the mean ± standard deviation (SD) of five replicates.

maximum POD increase rates at 10 d and 20 d, with 108% and 88%, respectively.

To investigate the physiological changes in potatoes under salt and drought stresses, we analysed the fresh weights and lengths of shoots and roots of potato seedlings at 20 days after transplanting. The root fresh weight and dry weight of all cultivars were all reduced by NaCl (Figs. 4 B- C). However, cv. LS and JY showed an excellent root growth ability under salt stress, after grown in 80 mM NaCl contained media, the root fresh/dry weight decreased by 31% and 47%, 50% and 37% in the two cultivars, respectively. On the other hand, the root fresh/dry weight of cv. SP were reduced most significantly among five cultivars, by 77% in fresh weight and 90% in dry weight.

The tolerance ability of different potato cultivars to high temperature: Tolerance to high temperature stress was assessed in the five potato cultivars. The different morphological status of each cultivar could be observed after 24 h under 42°C temperature and 12 h after recovery, cv. SP appeared to remain healthy while all other cultivars suffered severe wilting especially cv. LS (Fig. 5A). The water content showed a similar trait, 8% of water loss in cv. LS when suffering heat, while in cv. SP it was less than 2% (Fig. 6B). Photosynthetic activity (Fv/Fm) was determined in the fifth leaf from each plant, then, water content was measured. Heat shock reduced the photosynthetic activity of cv. LS and FR by 15% and 19% after 24 h, while the activity of cv. SP only decreased by 6% (Fig. 6C). Following 12 h of recovery at 25°C, except the continued decline in cv. LS, the Fv/Fm of other cultivars had all slightly ascended.

The relative membrane permeability of different potato cultivars in MV treatment: To evaluate tolerance to MV-mediated oxidative stress, the five potato cultivars were cultivated in a growth chamber for 5 weeks and then leaf discs were taken from the fifth healthy leaves for MV treatment. MV is a typical ROS-generating, redox-active compound and has been used as a non-selective herbicide [17]. Leaf discs were treated with 3 uM MV and, after 60 h, the reductions in membrane damage of each cultivar were noted, viz. 49% of cv. LS, 191% of cv. FR, 8% of cv. JY, 40% of cv. AT and 93% of cv. SP (Fig. 6B). The results showed that cv. FR suffered the maximum membrane damage among all the cultivars, and cv. SP was also sensitive to oxidative stress. It is worth noting that, the relative membrane permeability of cv. JY was quite different from that of the other specimens, much higher even in the control treatment without any MV. Based on unique performance, low increase rate of relative membrane permeability was shown with MV treated, compared with the control treatment.

Discussion

In this study, we investigated the change of antioxidant enzymes, and other physiological indices, of all the five potato genotypes under PEG, NaCl, high temperature and MV leaf discs treatments. Among the antioxidant enzymes, SOD is one of the major enzymes that catalyes the dismutation of superoxide into H_2O_2 and O_2 [18], as the first line of defense against ROS, and it is one of the most effective antioxidant enzymes in protecting plants from oxidative damage [10]. POD reduces H_2O_2 to water using various substrates as electron donors, and APX uses ascorbate to reduce H_2O_2 to water. CAT can break H_2O_2 down into water and oxygen [18].

Many researches also have been done to investigate the change of antioxidant enzymes of potato and other plants under different stresses. Under drought stress, the resistance has been associated with either maintenance or increase in antioxidant enzyme activity levels in various plant species. Antioxidant enzyme activity levels have been positively related to drought resistance in some species [18-20]. The levels of antioxidant enzymes are higher in tolerance than in sensitive species under various environmental stresses [21-23].

Potato terminal buds of each potato cultivar were transplanted to 1/2 MS medium with 0% or 2% PEG and 10 or 20 days later, the change of each enzyme was different from other cases, as DaCosta [24] reported that not all antioxidant enzymes change in

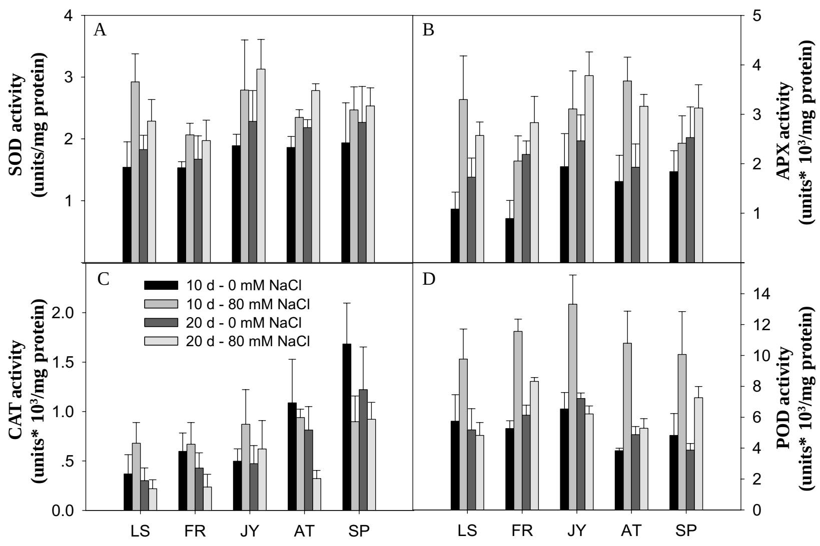

Figure 3. Potato cultivars grown in 1/2 MS medium contained NaCl (0 mM and 80 mM). The SOD activities (A), the APX activities (B), the CAT activities (C) and the POD activities (D) at 10 d and at 20 d with/without NaCl. Data are expressed as the mean ± standard deviation (SD) of five replicates.

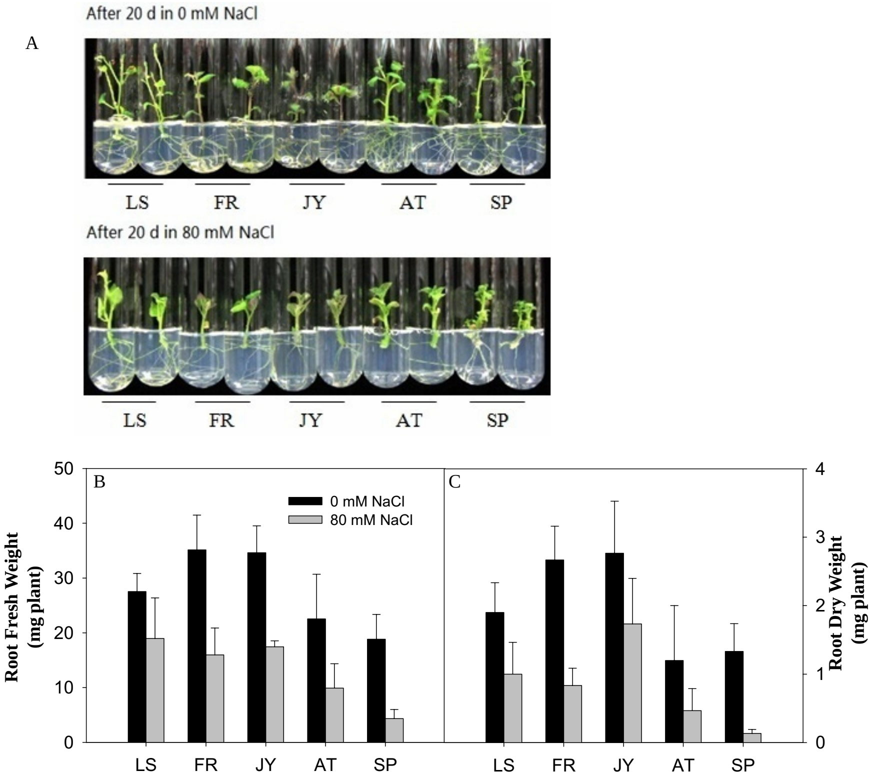

Figure 4. The root growth status (A), root fresh weight (B) and dry weight (C) of each cultivar after 20 days of transfer to NaCl (0 mM or 80 mM) containing MS medium. Data are expressed as the mean ± standard deviation (SD) of five replicates.

their activities at the same time or with the same pattern, in response to drought stress. The SOD was more active in cv. LS and cv. JY than other cultivars, and towards the APX activities, the activities of cv. LS were nearly tripled, being much higher than the second cv. FR, with only 0.8-fold increase. On the contrary, the activities of CAT and POD in cv. LS kept a, generally, similar level with others and even decreased in CAT activities with PEG. With the absence of cv. LS, the cultivar of AT obtained the biggest increase rate among all the cultivars. When taken the MDA into consideration, the lowest increase rate from 0% PEG to 2% PEG treatment was also augmented in cv. LS. Hence, we suppose that cv. LS is the cultivar with he best drought resistance ability among all the five cultivars as it has been frequently reported in many plant species that the drought or salt tolerance cultivar showed higher SOD ability, in a similar manner to potato [12], wheat [25], alfafa[26], bean [27] and tomato [28]. In order to eliminate the superoxide radicals (O^{2-}) and hydrogen peroxide (H_2O_2), the SOD was more responsible in cv. LS and APX was also quickly responsive at 10 d after transfer, better scavenging H_2O_2 as compared with other cultivars. Therefore, the damage to membrane was better prevented with the low MDA contents. As CAT directly breaks H_2O_2 into water and oxygen, it does not require a reducing power and has a high reaction rate, but a low affinity for H_2O_2, thereby, only removing the high concentration of H_2O_2 [29]. Thus, the CAT activities declined in cv. LS, cv. FR and cv. JY, but only increased in cv. AT and cv. SP, which also might have higher H_2O_2 contents and lead to the higher POD activities (Fig. 1 E).

High salt triggers various types of plant stress, including altered nutrition uptake, especially of ions such as K^+ and Ca^+, accumulation of toxic ions, especially Na^+, oxidant stress and osmotic stress [14]. When the five cultivars were exposed to salt stress via transferring their seedlings to 80 mM NaCl containing MS medium, cv. LS and cv. JY obtained the maximum SOD increase rate at 10 d and 20 d, respectively, and cv. LS also showed the highest APX rising tendency. In contrast, in terms of the lowest rate, cultivar of SP possesses the minimum increase. Root dry weight is an important index to evaluate the resistance ability of plant, hence, we assayed the root fresh/dry weight of each cultivar in 0 mM and 80 mM NaCl containing medium. After 20 days under salt stress, the root dry weight of cv. JY and cv. LS reduced by 37% and 47%, respectively, compared with control group while cv. SP reduced 90%. Taking the enzyme activities and root dry weight together into consideration, we can suppose that cv. LS and JY are tolerant to salt stress, but cv. SP is sensitive to salt stress. However, regardless of the drought and salt sensitivity features, cv. SP performed well under high temperature, according to our results (Fig. 5). It not only remain healthy while cv. LS was seriously withered, and the Fv/Fm and water content were both outstanding among all the cultivars with 6% and 2% less, whereas, those in cv. LS were 15% and 6%, respectively. To cope with high temperature, cv. SP is supposed to be a tolerant cultivar, while cv. LS is likely a sensitive cultivar, cv. AT and cv. FR also seem to be sensitive to high temperature, and cv. JY is likely tolerant to high temperature after cv. SP.

The five potato cultivars were taken into oxidative stress that was carried out by leaf discs in 3 μM methyl viologen (MV) containing sorbitol, and the data of relative membrane permeability of each cultivar at 60 h after incubation revealed that cv. AT and cv. LS were assumed to be the tolerant cultivar with the increase of 40% and 49% respectively, followed by cv. SP with 92% and cv. FR was quite sensitive to MV with 2-fold increase in relative membrane permeability.

To sum up all of the results that we have discussed above, cv. LS and cv. JY are assumed to have better resistance ability to drought and salt, while cv. SP is, obviously, a salt sensitive cultivar. Under high temperature stress, cv. SP has the best tolerance ability and it is followed by cv. JY, but cv. LS and cv. AT, which are sensitive cultivars. In relation to oxidative stress, cv. LS and cv. AT are quite tolerant, while cv. FR is sensitive.

It is interesting to notice that, cv. JY has an excellent tolerance ability to drought, salt and high temperature stresses, but the MDA content (under drought stress) and relative membrane permeability (under oxidative stress) are different from the other cultivars and seem quite unusual. This may relate to anthocyanin, the abundant natural pigment that reaches into the whole plant, including leaf and tuber of cv. JY (Fig. 7).

Anthocyanins represents one of the most widely distributed classes of flavonoids in plants. Apart from their colouring effects in fruits, anthocyanins show ability as an important antioxidant in the plant defense system in order to confront environmental stresses [30]. Anthocyanin leads to increase in the tolerance of

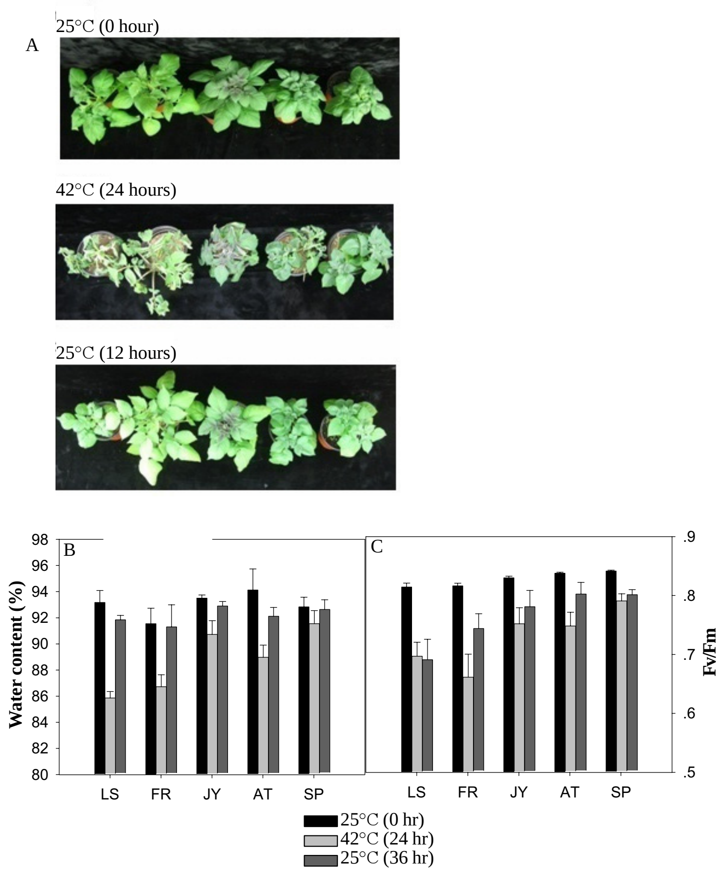

Figure 5. Five-weeks- old whole healthy plant of each cultivar suffered 42°C high temperature for 24 hours and recovered for 12 hours. The growth status of injury and recovery (A), the water content (B) and Fv/Fm values (C). Data are expressed as the mean ± standard deviation (SD) of five replicates.

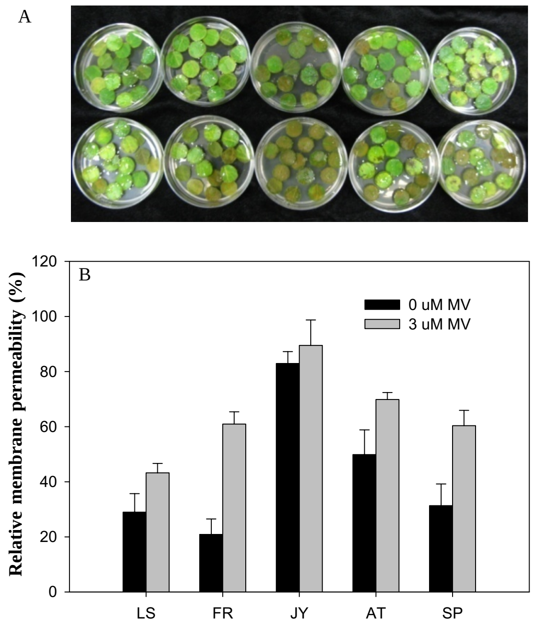

Figure 6. The leaf discs from each cultivar were incubated in MV (0 or 3 μM) contained sorbierite for 60 hours. The picture of all treatments (A), and the results of relative membrane permeability (B). Data are expressed as the mean ± standard deviation (SD) of five replicates

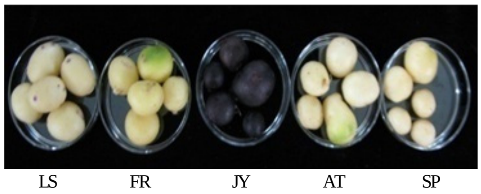

Figure 7. Tubers from each cultivars.

plants when suffering from various stresses, as many researches have proved in rice, Arabidopsis and other plant varieties [31-35]. Many researchers transformed various kinds of genes or transcription factors into different plant species, in order to enhance the anthocyanin content and, therefore, improve the stress resistance ability [36-38].

Conclusions

In this experiment, with exposing to drought, salt and high temperature stresses, cultivars normally have a specific resistance to one or two stresses, such as cv. LS is resistant to drought and salt while insensitive to high temperature; on the other hand, the resistance ability of cv. SP was converse to cv. LS. Among all of the cultivars, JY showed an overall strong resistance capability to each stress and this ability might be, notably, correlated with the lavish anthocyanin content within it. However, as no publication could be found to declare that, if there is any relationship between anthocyanin and plant membrane damage, we are not sure whether or not the special MDA content and the relative membrane permeability of cv. JY are caused by anthocyanin, or they are just the characteristics of the cultivar, therefor, it needs further investigation.

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