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ROLE OF ABSCISIC ACID (ABA) IN MODULATING THE RESPONSES OF TWO APPLE ROOTSTOCKS TO DROUGHT STRESS

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Abstract

Drought stress is considered as the main limiting factor for apple (*Malus domestica* L.) production in some semi-arid areas of China. In this study, we investigated the modulation role of abscisic acid (ABA) and fluridone (ABA synthesis inhibitor) on water relations and antioxidant enzyme system in 2-year-old seedlings of two apple rootstocks i.e. *Malus sieversii* (Ledeb.) Roem. (MS) and *Malus hupehensis* (Pamp.) Rehd. (MH). Drought stress induced ion leakage, accumulation of malondiadehyde (MDA) and decreases in leaf water potential and relative water content (RWC) in both rootstocks, which were significantly alleviated by exogenous ABA application. Drought stress also induced markedly increases in endogenous ABA content and activities of superoxide dismutase (SOD), peroxidase (POD), catalase (CAT), ascorbate peroxidase (APX), dehydroascorbate reductase (DHAR), monodehydroascorbate reductase (MDHAR), and glutathione reductase (GR), to a greater magnitude in MS as compared to MH rootstock. Concentration of 100μmol/L and 50μmol/L ABA had the most positive effects on drought-stressed rootstocks of MS and MH, respectively. Spraying optimum exogenous ABA contributed to enhancement in most of the above antioxidant enzymes activities but reduction in content of MDA and maintained the appropriate leaf water potential and RWC in both rootstocks. Pretreatment with fluridone aggravated ion leakage and the accumulation of MDA in two apple rootstocks under drought stress, which was overcome by exogenous ABA application to some extent. In conclusion, the endogenous ABA was probably involved in the regulation of two apple rootstocks in responses to drought stress.

Introduction

China is a major contributor to apple (*Malus domestica*) industry in the world and accounts for about 40% of the world production. The Loess Plateau area has become the most suitable for apple cultivation in China. However, in recent years, this region has been underdoing frequent and severe drought stress due to climate change, which is impacting sustainable development of apple industry (Zhang *et al*., 2007; Cao *et al*., 2012; Zhang *et al*., 2013; Zhao *et al*., 2013). Drought stress induces a series of biochemical and physiological responses such as inducing ion leakage, lipid peroxidation and leaf dehydration status resulting in accumulation of malondiadehyde (MDA) and reductions in leaf water potential (Apel *et al*., 2004; Ashraf, 2010). It has been suggested that DS also caused protein degradation by enhancing hydrolysis protease activity resulting in an increase in soluble protein (SP) content (Ashraf, 2010). To mitigate the detrimental effect of DS on plants, various strategies have been conducted to contend with this problem. Increasing crop resistance to drought stress by exploring genetics of enhanced water use efficiency is highly desirable for improving productivity and reducing agricultural reliance on fresh-water resources (Chaves *et al*., 2002; Xiong *et al*., 2006; Ashraf, 2010). Many studies has been carried out to understand the mechanisms in increasing drought tolerance of crop plants, including elucidating biochemical responses of apple cultivars to drought tolerance (Li *et al*., 2012), selection of drought-resistant rootstock (Liu *et al*., 2011), promoting drought gene expression (Li *et al*., 2010; Wang *et al*., 2011), and application of exogenous substances to enhance drought tolerance of plants (Zhu *et al*., 2004; Ma *et al*., 2008).

Abscisic acid (ABA) is considered as the most important signal substance, which plays an important role in physiological adaptation of plants to environmental stresses such as cold, salt, drought, heat and phosphate deficiency in plants (Hsu *et al*., 2003; Travaglia *et al*., 2010; Jia *et al*., 2013). It has been noted that application of exogenous ABA on leaves could create a wide array of adaptive changes to water deficit including accumulation of endogenous ABA and enhancement in activities of antioxidant enzymes including glutathione reductase (GR), superoxide dismutase (SOD), ascorbate peroxidase (APX) and catalase (CAT) (Duan *et al*., 2007; Ricardo *et al*., 2008; Zhang *et al*., 2006). In addition, reactive oxygen species (ROS) could be removed by exogenous ABA to enhance the cell membrane stability (CMS) to alleviate stress damage to plants (Zhang *et al*., 2001; Liu *et al*., 2005; Ricardo *et al*., 2008).

Many studies support that spraying exogenous ABA can enhance plants' stress resistance in various crop species (Tony & Gusta, 1983; Brigitte & Mauch, 2005; Nese *et al*., 2012). However, studies are seriously lacking on the evaluation of apple responses of different apple rootstocks to drought stress by application of exogenous ABA and/or fluridone (ABA synthesis inhibitor). The objective of this study was to evaluate the role of exogenous ABA in modulation the responses of two apple rootstocks i.e., two *Malus* species MH and MS in view of leaf water relations, antioxidant defense system and lipid peroxidation in an effort to develop practical guidelines to mitigate the negative effects of drought stress on apple.

Materials and Methods

Plant material and treatments: Seeds of *M. hupehensis* (Pamp) Rehd. (MH, drought sensitive) and *M. sievers*ii (Ledeb) Roem (MS, drought tolerant) (Bai *et al*., 2011; Liu *et al*., 2011) were collected from Pingyi, Shandong (35°07′N, 117°25′E) and Gongliu, Xinjiang (42°07′N, 86°37′E), China, respectively.

The experiment was conducted at the Northwest A&F University, Yangling (34°20′N, 108°24′E), China. Seeds were stratified with sand at 4°C for 35–40 days and then planted in plastic pots (12 cm×12 cm) filled with sand. The plastic pots were placed in a greenhouse under natural light and temperature conditions. At the two-leaf stage, the seedlings were replanted in plastic pots (25cm×35cm) filled with mixture of soil and matrix (ratio of 1:1) The field moisture capacity of the mixed soil was 28.4%. The tested soil was collected from the 0-20cm loam soil in the local farm field. In March 2012, 48 MH seedlings and 48 MS seedlings, which had the similar growth vigor, were replanted in other plastic pots (40cm×35cm) filled with the same mixture of soil and matrix as the described above. Each plastic pot was filled with 1.4 kg mixed soil and placed in a rain shelter under natural environment. Soil moisture treatments were divided into 2 levels by weighting method, i.e., 70-75% of soil moisture capacity (control, CK) and 40-45% of soil moisture capacity (drought stress) according to Shao, (2006). The white plastic bag was covered on the upper of pot to prevent the evaporation of soil moisture through the surface. The external part of pot was packaged with reflective film to prevent the soil from heating up too much.

Experiment 1: Different concentrations of ABA (0, 25, 50, 100, 200, 300μmol·L^{-1} ABA) were sprayed on both surfaces of leaves of each seedlings at 9:00 am, 12 June 2012. Each concentration of the solution was applied on leaves 5 times, and the interval time was 20 minutes. Before the treatment, the soil moisture was 72.1% of field moisture capacity, and then decreased to 40-45% of that after 7 days by weighting method. The plants grew in the natural humidity environment i.e., 70-75% of soil moisture capacity were treated as CK. All treatments were replicated four times, with a completely randomized block design. The leaves were sampled at the same age and different position of the plants after 7 days. The sampled leaves were detached and wrapped with wet absorbent gauze and taken to the laboratory timely to determine membrane permeability (MP) and MDA content.

Experiment 2: The optimum concentrations of ABA (determined from experiment 1) and fluridone were sprayed on both surfaces of leaves of other seedlings at 9:00 am, 22 July 2012. Application of drought stress and control (CK) treatments and ABA spraying method were the same as mentioned above. The seedlings were divided into five treatments groups: (1) control (CK): no drought; (2) drought stress; (3) drought+ABA; (4) drought + fluridone; (5) drought +ABA + fluridone. After 7 days, leaf samples at the same age and different position of the plant were collected. Some samples were wrapped with wet absorbent gauze and taken to the laboratory timely, and the others were quickly frozen in liquid nitrogen and stored at -80°C.

Measurements of MP, MDA content and soluble protein content: MP was determined according to Sairam & Srivastava (2005) with some modifications. The fresh leaves were punched with hole puncher, then collected randomly 20 pieces into a glass beaker containing deionized water. The beakers were incubated at 25°C for 2h, and then the conductivity of the solution was measured with a conductivity meter (HI 8633, Beijing Hanna Instruments Science & Technology Co., Ltd., China). After boiling the samples for 15 min, their conductivity was measured again when the solution was cooled to room temperature. The percentage of electrolyte leakage was calculated by using the formulae, EC (%) = (C1 /C2) × 100, where C1 and C2 are the electrolyte conductivities measured before and after boiling, respectively. MDA content was determined following the procedure of Hodges *et al*., (2000). The leaves were extracted with 10% trichloroacetic acid and absorbance measured at 450, 532 and 600 nm with 0.6% thiobarbituric acid. SP content was determined with coomassie blue staining according to the method described by Cusido *et al*., (1987), using bovine serum albumin as a standard.

Measurements of leaf water potential and relative water content (RWC): Leaf water potentials were measured in a pressure chamber (MODEL-100, PMS Instrument, Corvallis, OH, USA). The leaves were sampled on the outside of the crown in the middle of an annual shoot and the measurements were carried out in 9:00 am after 7 days after spraying ABA and fluridone under drought stress. Leaf RWC was determined by rapidly weighing (three leaves per plant), re-weighing them after allowing them to hydrate fully by floating them for 3h on deionized water, and then drying the leaves to a constant weight at 65°C.

Extraction and assay of antioxidant enzymes: Enzymes were extracted according to Grace & Logan (1996) with some modifications. Frozen leaves (0.5g) were homogenized in 100 mM potassium phosphate buffer (pH 7.0) containing 1 mM EDTA, 0.1% Triton-X-100, and 2% (w/v) PVP in a chilled mortar and pestle. The homogenate was centrifuged at 14000×*g* for 30 min at 4°C and the supernatant was used for the following enzyme assays.

SOD activity was estimated by measuring its ability to inhibit the photochemical reduction of nitroblue tetrazolium (NBT) (Dhindsa *et al*., 1981). One unit of SOD was considered to be the amount of enzyme required to inhibit tetrazolium (NBT) reduction by 50% in absorbance at 560 nm. Peroxidase (POD) activity was assayed as described by Ngo & Lenhoff (1980). POD activity was measured specifically with guaiacol at 470 nm and one unit of enzyme activity was taken as the rate of guaiacol which was oxidized in three minutes. CAT activity was determined by measuring the decreasing rate in the absorbance of H_2O_2 at 240 nm (Deng *et al*., 2012). One unit was defined as the amount of enzyme catalyzing the decomposition of 1 μmol $L^{-1}H_2O_2$ per minute. APX and GR activities were assayed using the method described by Cheng & Ma (2004). One unit of APX activity was the amount of APX catalyzes the oxidation of

1mmol ascorbate in absorbance at 290 nm per min. One unit of GR activity was defined as the reduction of 1 mmol oxidized glutathione (GSSG) per min at 340 nm. Monodehydroascorbate reductase (MDHAR) activity was assayed at 340 nm in 1 ml reaction mixture containing 50 mM phosphate buffered saline (PBS) (pH 7.8), 1mM coenzyme (NADH), 2.5 mM ascorbic acid (AsA), 25 units AsA oxidase and enzyme extract. The reaction was initiated by adding AsA oxidase (Miyake & Asada, 1992). One unit of MDHAR activity was defined as the amount of enzyme that oxidizes 1mmol NADH per minute. Dehydroascorbate reductase (DHAR) activity was measured at 265 nm in 1ml assay solution containing 50mM PBS (pH 7.8), 20 mM reduced glutathione (GSH), and 2 mM dihexyl adipate (DHA), 1 mM EDTA-Na_2. The reaction was initiated by adding DHA (Dalton *et al*., 1986). One unit of DHAR activity was defined as the amount of enzyme that produces 1 m mol AsA per minute. The specific enzyme activity for all the above enzymes was expressed as units (U) g^{-1} fresh weight.

ABA determination: The extraction, purification and measurement of endogenous levels of ABA were conducted by an indirect ELISA technique described by Yang *et al*., (2001) and Ali & Asharf, (2011) with some modification. Briefly, the frozen leaf samples (0.5g) were homogenized in liquid nitrogen and extracted in cold 80% (v/v) methanol containing 1mm butylated hydroxytoluence as an antioxidant. The extract was incubated at 4 ^oC for 4 h and centrifuged at 4,000×*g* for 15 min at the same temperature. The supernatant was passed through Chromosep C18 columns (C18 Sep-Park Cartridge, Waters Corp., Millford, MA, USA), prewashed with 10mL 100% (w/v) and 5 mL 80% (v/v) methanol, respectively. The hormone fractions from the columns were eluted with 10mL 100% (v/v) methanol and 10 mL ether. The resulting elution was dried under N_2 gas, and dissolved in 2 mL phosphate buffer saline (PBS) containing 0.1% (v/v) Tween 20 and 0.1% (w/v) gelatin (pH 7.5) for ELISA analysis. The mouse monoclonal antigen and antibody against ABA used in the ELISA were produced at the Phytohormones Research Institute, China Agricultural University, Beijing.

Statistical analysis: Each treatment included 4 replications (flasks). Analysis of variance was calculated using the SPSS-17 statistical software. The analysis of variance (ANOVA) was followed by the least significance test to determine the significant difference among the mean values at 95% confidence limit.

Results

Effect of different ABA concentrations on MP and MDA content in two apple rootstocks (MS and MH) under drought stress: MP and MDA content were increased by 18% and 41% in MS, and by 75% and 85% in MH leaves, respectively, when exposed to drought stress as compared to CK. MP and MDA content of two apple rootstocks changed dependant on pretreatment of different ABA concentrations of 25 to 300μmol·L^{-1} (Fig. 1A, B). As shown in Fig. 1A, B, 100 μmol/L and 50 μmol/L ABA had the most obvious positive effects on MS and MH respectively due to their lowest MP and MDA content.

Effects of ABA and fluridone on leaf water potential and RWC in two apple species (MS and MH) under drought stress: Fig. 2A, B shows the effects of exogenous ABA and fluridone on leaf water potential and relative water content of two apple species i.e., two *Malus* species MH and MS. Leaf water potential and RWC of MS were both greater than MH under drought stress regardless of application of exogenous ABA and/or fluridone. Spraying optimal exogenous ABA increased leaf water potential and RWC in two rootstocks under drought stress. Such, exogenous ABA could alleviate the leaf dehydration status to some extent. However, this function could be inhibited by fluridone which can suppress the formation of endogenous ABA.

Effects of ABA and fluridone on MP and contents of SP and MDA in two apple rootstocks (MS and MH) under drought stress: Drought stress increased MP and contents of SP and MDA in both rootstocks as compared to CK. Exogenous ABA could decrease SP and MDA contents in both rootstocks under drought stress. However, application of ABA decreased MP in both MS and MH under drought stress, but remained the similar level in MH as the control plants. Fluridone significantly increased enhanced SP as compared to that in all other treatments in both rootstocks. The MP and MDA content were greater in the plants treated with fluridone as compared to those in the plants treated with drought combined with ABA in both rootstocks as well as those in the plants treated with drought only in MS. The MDA contents of both rootstocks treated with combination of ABA and fluridone were lower than those treated with single fluridone (Fig. 3 A, B, C).

Effects of ABA and fluridone on antioxidant enzymes activities in two apple rootstocks (MS and MH) under drought stress: Drought stress increased SOD activity by 5 and 9% in MS and MH rootstocks respectively, as compared to that in control plants. SOD activity was greater in MS leaves as compared to that in MH leaves across all treatments. Exogenous ABA significantly increased SOD activity of MS rootstock as compared to that of the plants under drought stress alone. Fluridone decreased SOD activity, which was overcome by exogenous ABA (Fig. 4A). Similar responses were also obtained for other enzymes activities including POD, CAT, APX, DHAR, GR and MDHAR with few exceptions (Fig. 4B, C, D, E, F, G).

Effects of ABA and fluridone on endogenous ABA content in two apple rootstocks (MS and MH) under drought stress: ABA contents were increased to 169 and 107 ng g^{-1} FW in the MS and MH leaves respectively due to drought treatment. Endogenous ABA content was greater in MS leaves as compared to that in MH leaves under drought stress (Fig. 5). Application of exogenous ABA increased but fluridone decreased endogenous ABA content in both rootstocks under drought stress. However, application of exogenous ABA plus fluridone decreased endogenous ABA content in MS rootstock and remained unchanged pattern of endogenous ABA in MH rootstock under drought stress (Fig. 5).

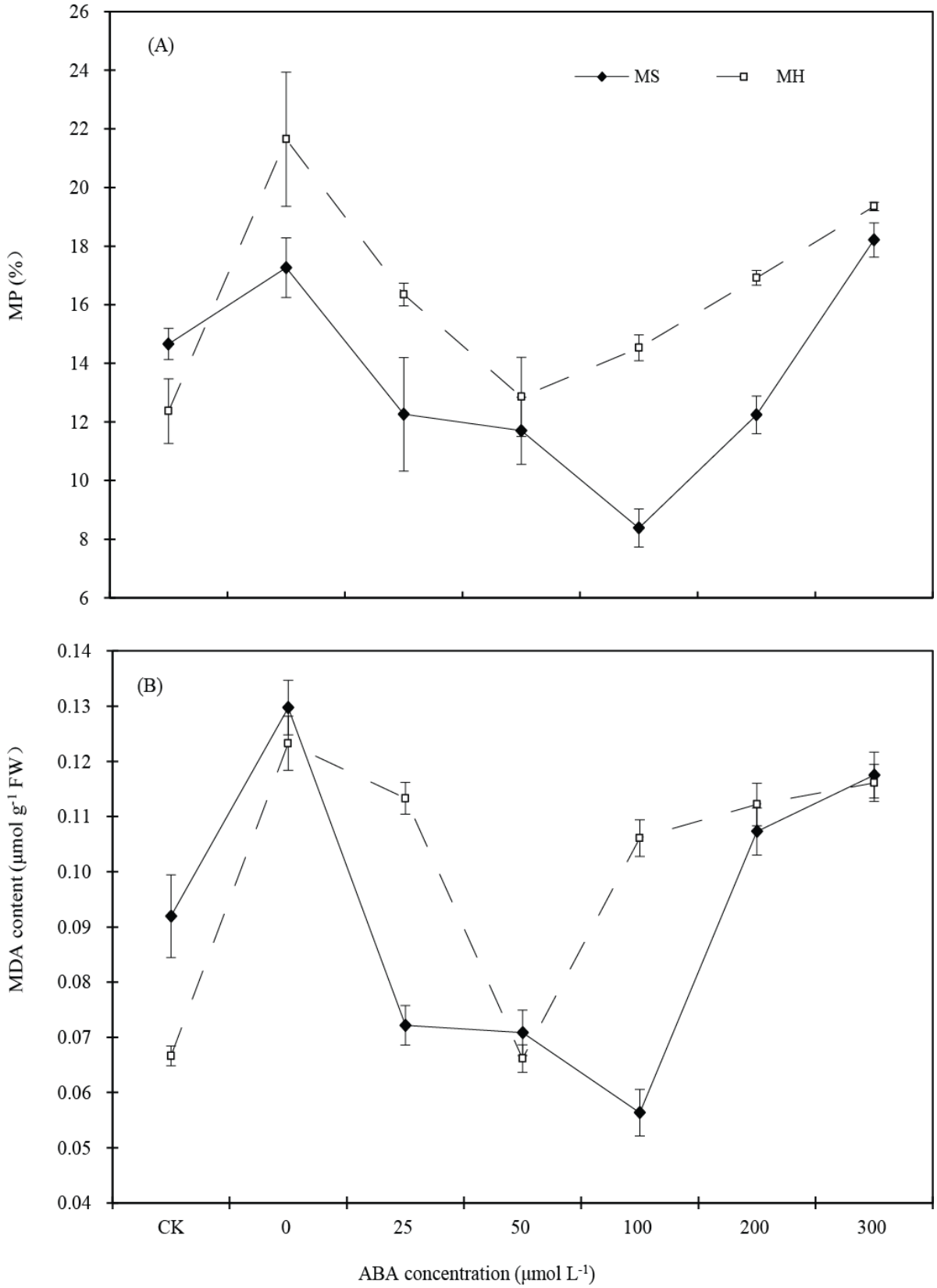

Fig. 1. Effect of different ABA concentrations on membrane permeability (MP) (A) and malondiadehyde (MDA) content (B) in leaves of MH (*Malus. hupehensis* (Pamp) Rehd) and MS (*Malus. sievers*ii (Ledeb) Roem) rootstocks under drought stress. Values are means ± S.E. (*n* =4).

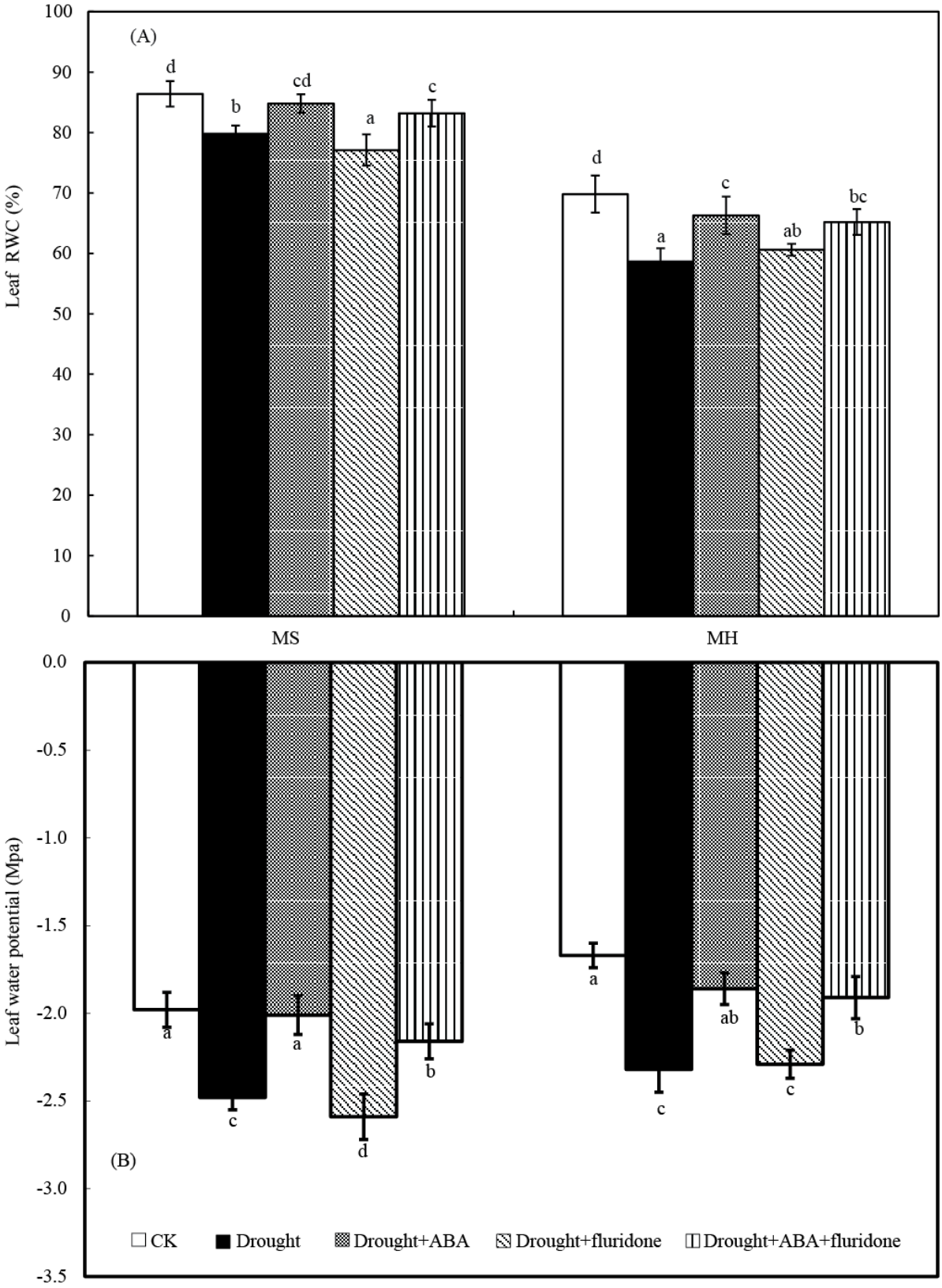

Fig. 2. Leaf relative water content (RWC) (A) and Leaf water potential (B) in leaves of MH (*Malus. hupehensis* (Pamp) Rehd) and MS (*Malus. sievers*ii (Ledeb) Roem) rootstocks under five treatments i.e. control (CK), drought, drought + ABA, drought + fluridone and drought + ABA + fluridone. Values are means ± S.E. (n =4). At the top of bars, different letters indicate significant differences among the mean values (p<0.05), by each *Malus* rootstock species.

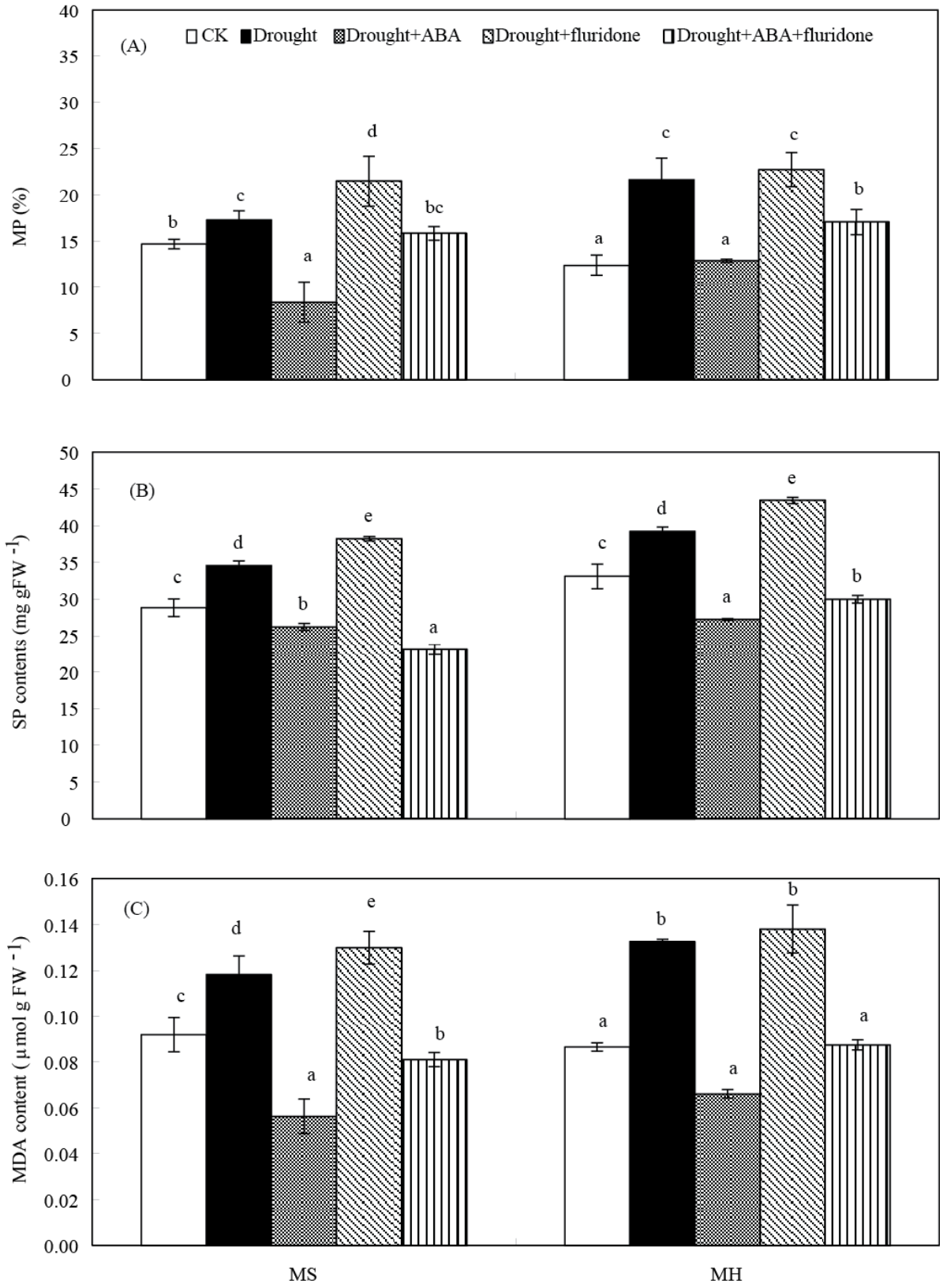

Fig. 3. Membrane permeability (MP) (A), soluble protein (SP) content (B) and malondiadehyde (MDA) content (C) in leaves of MH (*Malus. hupehensis* (Pamp) Rehd) and MS (*Malus. sievers*ii (Ledeb) Roem) rootstocks under five treatments i.e. control (CK), drought, drought + ABA, drought + fluridone and drought + ABA + fluridone. Values are means ± S.E. (n =4). At the top of bars, different letters indicate significant differences among the mean values (p<0.05), by each rootstock.

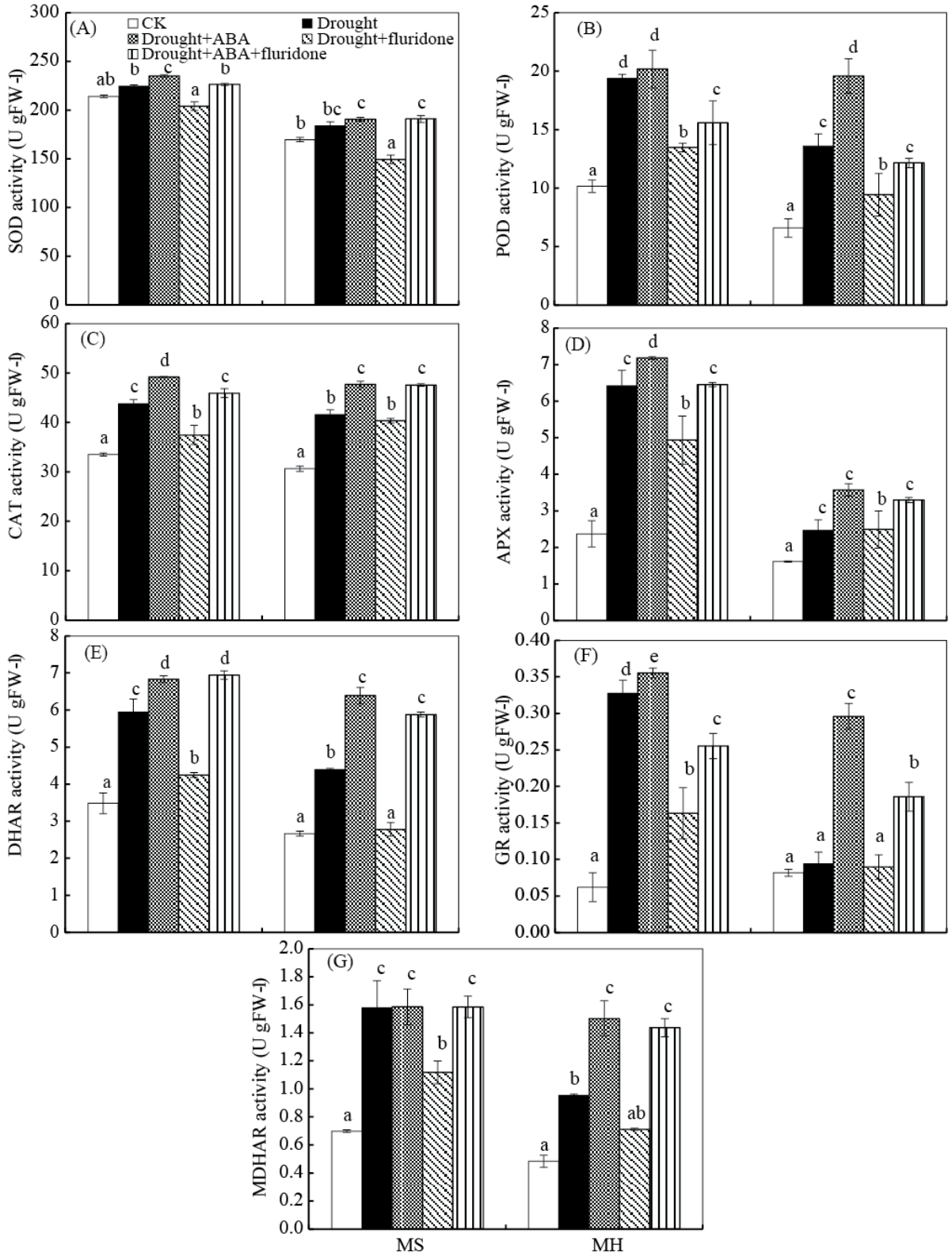

Fig. 4. Activities of superoxide dismutase (SOD) (A), peroxidase (POD) (B), catalase (CAT) (C), ascorbate peroxidase (APX) (D), dehydroascorbate reductase (DHAR) (E), glutathione reductase (GR) (F) and monodehydroascorbate reductase (MDHAR) (G) in leaves of MH (*Malus. hupehensis* (Pamp) Rehd) and MS (*Malus. sievers*ii (Ledeb) Roem) rootstocks under five treatments i.e. control (CK), drought, drought + ABA, drought + fluridone and drought + ABA + fluridon. Values are means ± S.E. (n =4). At the top of bars, different letters indicate significant differences among the mean values ($p<0.05$), by each rootstock.

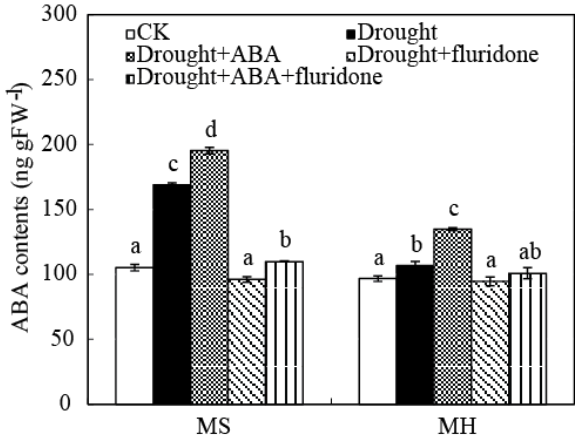

Fig. 5. Endogenous ABA content in leaves of MH (*Malus. hupehensis* (Pamp) Rehd) and MS (*Malus. sievers*ii (Ledeb) Roem) rootstocks under five treatments i.e. control (CK), drought, drought + ABA, drought + fluridone and drought + ABA + fluridone. Values are means ± S.E. (n =4). At the top of bars, different letters indicate significant differences among the mean values (p<0.05), by each rootstock.

Analysis of variance for antioxidant enzymes activities parameters of two apple rootstocks i.e., two *Malus* species (MS and MH): Analysis of variance showed that the effects of different treatments (T) and species (S) were significant on leaf water potential, RWC, SP, MDA and most antioxidant enzymes activities parameters measured, except the effects of S on CAT and MDHAR activities (Table 1). Two-way interactions were also significant for activities of SOD, POD, APX, GR and MDHAR. This study showed that choice of species and spraying exogenous optimum ABA is important to overcome the adverse effects of drought stress in terms of plant antioxidant responses.

Discussion

Drought stress is one of the major constrains of production systems in many parts of the world including China. Water shortages and soil water losses due to environmental and land use changes are challenges to fruit production (Xia *et al*., 2007; Ashraf, 2010; Ahmed *et al*., 2013). Previous studies have indicated that drought stress usually induces the accumulation of reactive oxygen species (ROS), which causes oxidative damage to plants (Apel & Hirt, 2004; Papadakis *et al*., 2005), resulting in membrane lipid peroxidation, membrane disruption and growth suppression (Liu *et al*., 2000; Jiang & Huang, 2001). MP and membrane lipid peroxidation product MDA have been commonly used to distinguish drought-tolerant and drought-sensitive genotypes of many plants (Zhang *et al*., 2007; Bai *et al*., 2010). In the current study, MP and MDA content increased in both rootstocks after the period (7 days) of moisture stress. Greater accumulation of MDA and MP in MH leaves as compared to MS leaves demonstrated that MS leaves were relatively drought-tolerance as compared to the former rootstock. This agrees with the conclusion of Bai *et al*., (2011) and Wang *et al*., (2012). Additionally, the effects of exogenously applied ABA on the above mentioned parameters were highly significant. It is, therefore, suggested that the optimal application of ABA can benefit plant growth under drought stress, but this response is cultivar-specific and dose-specific as evident from data in Table 1 and Fig. 1 A, B, respectively.

ABA is a commonly occurring plant growth hormone in plants actively involved in plant growth and development under drought conditions (Zhang *et al*., 2006). The present study showed that optimum exogenous ABA could enhance drought-tolerance in both rootstocks by alleviating leaf dehydration status, ion leakage, lipid peroxidation and leaf water potential induced by drought stress (Figs. 1-5). The endogenous ABA content in both MS and MH rootstocks increased rapidly under drought compared to control (CK). The ABA content in MS leaves was higher than that in MH. This study used a direct inhibitor of ABA synthesis i.e. fluridone to elucidate further the physiological role of endogenous ABA in modulation of drought-tolerance of two apple rootstocks under drought stress. Results showed that treatment with fluridone had significant impacts on aggravating membrane damage and lipid peroxidation. These responses were greater in MS leaves as compared to those in MH leaves. The large-scale synthesis of ABA in MS leaves induced by drought stress might explain greater tolerance of this rootstock to drought stress as compared to MH rootstock. Application of exogenous ABA to both rootstocks significantly reduced ion leakage, lipid peroxidation and SP content induced by drought stress, and increased leaf water potential and RWC in leaves as well as activities of SOD, CAT, POD, APX, DHAR, MDHAR and GR, thus improved leaf dehydration status and scavenged ROS. These physiological changes in turn enhanced drought tolerance. However, application of fluridone inhibited the beneficial effects of ABA, thus, decreased drought tolerance of plants. Therefore, ABA produced in two apple rootstocks under drought stress might serve as a signal for the inducing drought-tolerance of plants.

Previous research has shown that ABA has an affect on stomatal closure in *Arabidopsis* (Bright *et al*., 2006). Similarly, ABA might be acting as a signaling substance under drought stress (Carlos & Lamattina, 2002). Plant responses to both biotic and abiotic stresses require action of common signaling component which involve ABA (Hancock *et al*., 2011). ABA has an influence on physiological changes in response to stresses such as drought, cold and salt, i.e. stomatal movements, leaf growth and expansion, the control of water movements and gene expression. Theses mechanisms were mediated by shorter term effects on signalling components such as phosphatases and thus the alteration of cellular phosphorylation levels (Wilkinson *et al*., 2010; Zhang *et al*., 2006; Parent *et al*., 2009; Hubbard *et al*., 2010; Jia *et al*., 2013). So far, although application of ABA under salt, osmotic and drought stresses have been reported (Sarath *et al*., 2007), little is known about the optimum exogenous ABA concentration applied on two apple rootstocks which are widely used in the Loess Plateau of China under drought stress. This study concluded that the ABA acted as signaling transduction substance during adaptive plant responses to drought, and spraying optimum exogenous ABA could act as an effective management tool to mitigate drought stress in apple production in the arid regions.

Table 1. *F* values of the effects of endogenous ABA treatments (T), species (S), and their interactions on all parameters measured in this study.

Source of variation	Endogenous ABA treatments (T)	Species (S)	T×S
LWP	4.033*	5.302*	0.077
LRWC	16.065***	358.124***	0.353
MP	5.426**	0.279	1.079
SP content	38.894***	26.451***	1.117
MDA content	79.801***	86.064***	89.950***
SOD activity	36.109***	196.498***	3.138*
POD activity	58.635***	43.902***	2.906*
CAT activity	42.179***	0.13	1.813
APX activity	13.923***	76.549***	3.159*
DHAR activity	44.445***	24.106***	0.888
GR activity	14.550***	13.329**	3.243*
MDHAR activity	6.025**	0.604	4.514**

*, **, *** significance at 5%, 1% and 0.1 % level of significance, respectively. LWP, leaf water potential; LRWC, leaf relative water content; MP, membrane permeability; MDA, malondiadehyde concentration; SP, soluble protein content; SOD, superoxide dismutase; POD, peroxidase; CAT, catalase; APX, ascorbate peroxidase; DHAR, dehydroascorbate reductase; GR, glutathione reductase; MDHAR, monodehydroascorbate reductase

Conclusion

In summary, drought stress exerted adverse effects on leaf water attributes of the two apple rootstock i.e. two *Malus* species. However, exogenous application of ABA was effective in enhancing stress tolerance of two apple rootstocks by increasing activities of antioxidant enzyme (SOD, CAT, POD, APX, DHAR, MDHAR and GR) and water relations performance under drought conditions. The above positive roles of ABA were inhibited by fluridone which can suppress the synthesis of endogenous ABA. Such, It is, therefore, concluded that endogenous ABA was probably involved in modulation the responses of two apple rootstock under drought stress.

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