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Exogenous application of polyamines alleviates water stress-induced oxidative stress of *Rosa damascena* Miller var. *trigintipetala* Dieck

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ABSTRACT

The growth and development of Damask rose (*Rosa damascena* Miller var. *trigintipetala* Dieck) are drastically inhibited under water stress. Polyamines (PAs) are involved in water stress tolerance. However, very limited information is known concerning the effects of exogenous application of PAs on aromatic plants including Damask rose. This experiment was carried out to study the application of Spermine (Spm) or Spermidine (Spd) on some physiological and biochemical processes to understand the possible mechanisms concerning the water stress alleviation in Damask rose. Plants were exposed to two water conditions: 100% of field capacity (FC) as well watered treatment and 50% of FC as a water stress treatment. Foliar applications of Spm or Spd were applied at 0.5 mM while control plants were sprayed with distilled water. The application of Spm or Spd improved the growth characters, relative water content (RWC), chlorophyll content and stomatal conductance under water stress. Furthermore, proline content and CAT and SOD enzyme activities were also improved by applying Spm or Spd. H_2O_2 production was restricted and MDA accumulation was limited and hence the membrane stability was retained and the water stress damage was alleviated accordingly. In addition, exogenous application of Spm or Spd reduced the endogenous Put level and increased both Spm and Spd levels which suggest that PAs were implicated in water stress adaptation of Damask rose plants. Enhancing Damask rose from water stress not only by activating the antioxidant machinery but also by balancing the PAs metabolism due to exogenous application of Spm or Spd was suggested.

1. Introduction

Water deficit is a main challenge to agricultural production all over the world. Moreover, it is a prime abiotic stress that negatively affects both plant growth as well as productivity (Ribaut et al., 2012). Negative effects of water stress are also associated with changes of different physiological and biochemical factors on aromatic plants and consequently cell turgor loss (Hassan et al., 2013). Moreover, water stress triggers the production of reactive oxygen species (ROS) such as H_2O_2 and O_2 which caused lipids oxidative stress observed by the increment of MDA content (Talaat et al., 2015).

It has been well documented that plants can ameliorate the damage caused by water stress through several physiological and biochemical mechanisms. Mainly by compatible solutes accumulation that lead to osmotic adjustment which resulted in cell turgor maintenance (Marcinska et al., 2013) and promotes the antioxidant enzyme activities like superoxide dismutase (SOD) and catalase (CAT) to detoxify ROS (Arbona and Gómez-Cadenas, 2008). The improvement of antioxidant system was correlated with water stress tolerance in creeping bent grass (Merewitz et al., 2011; Li et al., 2015). Several reports showed that abiotic stresses, especially water stress, elevate polyamines (PAs) biosynthesis (Yang et al., 2007; Kubiś, 2008). Therefore, they may be involved in the plant adaptive mechanism toward different environmental stresses (Bouchereau et al., 1999; Groppa and Benavides, 2008). Several studies concluded that PAs are also implicated in plant growth (Pottosin et al., 2014) and stress tolerance (Farooq et al., 2009; Li et al., 2015).

PAs are ubiquitous, aliphatic amines, polycationic that play important roles in the growth and development of plant (Martin-Tanguy, 2001). Moreover, they modulate several biological processes, including plant stress alleviation due to their effective roles in free radicals scavenging (Roychoudhury et al., 2011), osmotic adjustment and membrane integrity improving (Takahashi and Kakehi, 2009). It has been documented that PAs are involved in different environmental stresses tolerance including water stress (Farooq et al., 2009; Kubiś et al., 2014; Li et al., 2015; Talaat et al., 2015; Sánchez-Rodríguez et al., 2016), salt stress (Zapata et al., 2004; Ghosh et al., 2012; Kamiab et al., 2014) and heavy metal stress (Ding et al., 2010; Xu et al., 2011).

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Foliar application of Spd alleviated the water deficit adverse effects by lowering ROS and MDA contents and increasing RWC, chlorophyll and promoted the antioxidant system relative to untreated control (Nayyar and Chander, 2004; Li et al., 2015). Water stress in cucumber increased membrane damage, MDA and proline accumulation however exogenous application of PAs effectively reduced the membrane injury, lipid peroxidation and elevated the proline accumulation (Kubiś et al., 2014). Sánchez-Rodríguez et al. (2016) suggested that a higher content of PAs (especially Spm) promoted the antioxidant systems since their applications showed a lower ROS generation and higher CAT and SOD enzyme activities which enhanced the dehydration tolerance.

Foliar application of Spm increased the antioxidant enzyme activities which restricted the accumulation of both ROS and MDA, accordingly alleviated the negative effects of water stress in maize plants (Talaat et al., 2015). Furthermore, PAs improved water status (osmotic and turgor potentials as well as RWC) of leaves, photosynthesis and the properties of membrane, and hence protect rice plants from oxidative stress raised by drought (Farooq et al., 2009). Water stress was ameliorated due to Spm foliar application by elevating the antioxidant system and inhibition of ROS and MDA accumulation and consequently the growth and plant production were enhanced (Farooq et al., 2009; Radhakrishnan and Lee, 2013). With the same line, Spm and Spd application also decreased the production of ROS as well as MDA content which reflected on reducing the plasma membrane damage (Xu et al., 2011).

Although, PAs improve several abiotic stresses tolerance in plants mainly by inhibition of oxidative damage, another mode of actions during foliar application are to be explored to improve water stress tolerance in various species. Many studies of PAs have performed on various crops (Ding et al., 2010; Talaat et al., 2015; Sánchez-Rodríguez et al., 2016) however; the available information concerning the effects of PAs on aromatic plants is very limited. *Rosa damascena* Miller var. *trigintipetala* Dieck (Damask rose) which belongs to Rosaceae family is very important species of Rosa genus. This aromatic plant has several products such as rose oil, concrete and rose water which is widely used not only in perfume but also in pharmaceutical, cosmetic, and food industries (Ali et al., 2014; Abdel-Hameed et al., 2016). Additionally, Damask rose was also used as antioxidant, anti-bacterial, anti-diabetic, anti-HIV and anti-inflammatory activity (Boskabady et al., 2011; Baydar and Baydar, 2013).

Interestingly, although the importance of Damask rose, the studies about water stress alleviation in rose is very scarce. Pinior et al. (2005) used arbuscular mycorrhizal fungus and Chen et al. (2016) used gene pyramiding to improve the drought tolerance of roses. Additionally, there is no information about the efficacy of exogenous PAs application to enhance water stress tolerance in this plant especially with the fact that the shortage of fresh water supply is increasing seriously. Because of its economic value, it is a suitable plant for this experimental research. Therefore, the exogenous application of Spm or Spd on some physiological and biochemical processes was targeted to understand the possible mechanisms concerning the water stress alleviation in Damask rose.

2. Materials and methods

2.1. Soil preparation and plant cultivation

This pot experiment was conducted in the greenhouse area of Faculty of Science, Taif University, KSA (21°26′02.4″N 40°29′36.9″E) through 2015 and 2016 seasons. Uniform rooted stem cuttings of *Rosa damascena* Miller var. *trigintipetala* Dieck were cultivated in plastic pots 30 cm filled with sandy soil. The soil physical properties were (sand, 80.26%, silt 6.88% and clay 12.86%) and chemical attributes were (EC, 2.13 dSm^{-1}, pH 8.16, OM, 0.12%, Total $CaCO_3$, 0.82%, Ca^{+2}, 42.69 ($meqL^{-1}$), Na^+, 3.52 ($meqL^{-1}$), SO_4^{-2}, 47.73 ($meqL^{-1}$), HCO_3, 2.15 ($meqL^{-1}$), Cl^-, 0.58 ($meqL^{-1}$), total N^+, PO_4^{-3}, K^+ were 0.16, 0.043 and 0.051%, respectively). All pots were received a compound chemical fertilizer containing NPK (17:17:17) at constant level of 3 g/pot according to manufacturer recommendation. The average temperature, relative humidity and rainfall were 23.9 °C, 35.57 and 12.84 mM, respectively during the study.

2.2. Water stress treatments

After three weeks from transplanting, the plants were exposed to two water conditions: 100% of field capacity (FC) as well watered treatment and 50% of FC as a water stress treatment. The soil water content (SWC) was measured according to the method of Coombs et al. (1987).

2.3. Polyamines treatments (PAs)

After three weeks from the beginning of water stress treatments, foliar applications with PAs i.e. Spermine (Spm) and Spermidine (Spd) were applied at 0.5 mM till run off. A preliminary investigation was performed to determine the exact concentration of PAs applied. The plants were weekly sprayed for two months. A surfactant of Tween 80 (0.5% v/v) was used. Control treatment was sprayed with distilled water containing the same surfactant. A completely randomize design was applied to arrange the pots in two way factorial experiment (Snedecor and Cochran, 1980). Each treatment had four replicates with five pots each.

2.4. Growth parameters

By the end of PAs treatments, the vegetative growth characters were assessed and the samples were taken for the subsequent required analysis. The same leaf part and the leaves of the same sizes were used for measurements. Plant height was measured and presented as (cm). The plants were removed from the pots and the fresh weight (g) was directly determined and the dry weight was measured (g) after drying at 70 °C for 48 h.

2.5. Relative water content (RWC)

Leaf midday RWC was measured by the method of Weatherley (1950).

2.6. Stomatal conductance

Leaf porometer (Delta T AP4, UK) was used to determine leaf stomatal conductance (mol H_2O m^{-2} s^{-1}).

2.7. Chlorophyll determination

The total chlorophyll content in rose leaves was spectrophotometrically determined according to Metzner et al. (1965). The values of chlorophyll were expressed as mg g^{-1} FW.

2.8. Proline analysis

The free content of leaf proline was measured as described by Bates et al. (1973). The proline concentration was calculated based on a standard curve and was expressed as μmol g^{-1} FW.

2.9. Membrane stability index (MSI)

MSI was measured by the level of ions leakage as described by Sairam et al. (1997).

2.10. Lipid peroxidation determination

Lipid peroxidation was spectrophotometrically assessed by the level of Malondialdehyde determination (MDA) as mentioned by Hodges et al. (1999). The MDA content was expressed as μmol ml^{-1}.