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Foliar applications of abscisic acid decrease the incidence of blossom-end rot in tomato fruit☆

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ABSTRACT

Various environmental stress factors, such as drought and high relative humidity, can cause calcium (Ca) deficiency and lead to physiological disorders such as blossom-end rot (BER) in tomato (*Lycopersicon esculentum*) fruit. Recent studies demonstrate that abscisic acid (ABA) triggers whole-plant and fruit-specific mechanisms to increase fruit Ca uptake and prevent BER development. The objective of this study was to examine the effects of foliar application of ABA and hydroponic Ca treatments in fertilizer solution on localized deficiency of Ca causing BER in tomato fruit. The application of 500 mg L^{-1} ABA foliar spray treatment significantly decreased Ca in the leaf tissue. Ca decreased by 10.7% in the leaf tissue when comparing the foliar application of ABA to the control. In addition, decreasing Ca treatments from the 180 mg L^{-1} to the Ca deficient treatment of 60 mg L^{-1} decreased Ca concentration in the leaf tissue by 39.5%. The interaction of ABA and Ca treatments had a significant effect on Ca concentration in the fruit tissue. Ca concentration increased 25.7% when comparing the 180 mg L^{-1} Ca and 0.0 mg L^{-1} ABA treatment to the combination treatment of 180 mg L^{-1} Ca and 500 mg L^{-1} ABA treatment. In addition, ABA treatments had statistically significant effect on Ca in tomato fruit proximal and distal tissue and increased the concentrations 14.7% and 34.6% in, respectively. The incidence of BER in tomato fruit tissue was lowest with ABA treatments and Ca treatment of 180 mg L^{-1}. The incidence of BER decreased by 86.2% from the combination of 180 mg L^{-1} Ca and 0.0 mg L^{-1} ABA treatment to the combination treatment of 180 mg L^{-1} Ca and 500 mg L^{-1} ABA treatment. The results demonstrate that the application of ABA increased Ca in the fruit tissue as Ca treatments were decreased from the 180 mg L^{-1} Ca. Furthermore, our results demonstrate that, despite reducing total plant Ca uptake in the leaf tissue, the 500 mg L^{-1} ABA foliar spray treatments significantly reduced the incidence of BER development in tomato fruit. Thus, ABA could be an alternative treatment to increase Ca uptake into fruit and distribution into the distal tissue of the fruit relative to leaf uptake.

1. Introduction

The plant transports Ca from tissue to tissue through uptake and distribution. On the molecular level, Ca is transported through the symplast. On the physiological level, Ca, which originates at the root, is transported from the root cortex through the xylem tissue and distributed to vegetative and fruit tissue. The uptake of Ca is higher in apical than in basal root zones (Haussling et al., 1988; Ferguson and Clarkson, 1976). Root apical zones take up high amounts of Ca. Part of the Ca remains in the root while the rest is delivered to the shoots (Clarkson, 1984). Calcium distribution in the plant can be negatively affected by environmental stresses, such as light, temperature and humidity. The negative impact on Ca distribution in the plant can cause Ca deficiencies and can have severe physiological dysfunctions that decrease tomato yield and fruit quality. Perhaps the most deleterious of these disorders is blossom end rot (BER) which is induced by Ca deficiency and plant stress.

BER primarily occurs because of the local deficiency of Ca in the distal end of tomato fruit (Suzuki et al., 2003; Adams and Ho, 1993). BER is generally attributed to an inadequacy of Ca in the fruits, and it is therefore called a 'Ca-related disorder' (Shear, 1975). This disorder causes cells near the blossom end of the fruit to die, giving the tissue a water-soaked appearance that can cover half of the fruit surface (Abdal and Suleiman, 2005). Although it can be caused by

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inadequate supply of Ca in the root zone, it frequently occurs when substrate moisture and Ca content is at adequate levels for normal plant growth. In this situation, the most likely causes of this physiological disorder are poor Ca uptake by the roots and insufficient distribution of Ca to the fruit during a period of high Ca demand. Research on greenhouse tomato production has demonstrated that insufficient Ca supplied to the plants in the fertilizer solution rarely causes BER. More often, BER occurs in plants with an adequate Ca supply when grown in environmental conditions that reduce transport of Ca to rapidly growing distal fruit tissue (Saure, 2001; Ho and White, 2005). In addition, incidences of BER may occur during increased demand of distal fruit tissue for Ca in early stages of fruit development (Ho et al., 1993; Ho and White, 2005).

Plant growth and development are regulated by internal and external signals. One important regulator that coordinates these changes is the plant hormone abscisic acid (ABA). ABA can trigger oscillation in the cytosolic Ca concentration, which is then perceived by Ca binding proteins to initiate a series of signaling cascades that control many physiological processes, including adaptation to environmental stress (Guo et al., 2002). Recent studies demonstrate that ABA triggers whole-plant and fruit-specific mechanisms to increase fruit Ca uptake and prevent BER development. For example, de Freitas et al. (2011) found that ABA induced lower leaf stomatal conductance and water loss, which resulted in increased Ca concentrations in the fruit and lower Ca levels in the leaves. The role of ABA as a stress hormone makes it an attractive and novel treatment to improve Ca uptake and distribution within tomato fruit, which could increase Ca concentrations in situations where Ca distribution into the fruit is low.

The objective of this study was to examine the effects of foliar application of ABA on localized deficiency of Ca causing BER in tomato fruit. In addition, we examined how foliar spray ABA applications at different Ca fertility levels affected the partitioning of Ca between leaves and fruit of tomato plants, especially in the distal tissue.

2. Materials and methods

2.1. Plant culture and harvest

Seeds of 'Mountain Fresh Plus' tomato (Johnny's Selected Seed, Waterville, ME) were sown into Pro-Mix BX soilless medium (Premier Tech Horticulture, Québec, Canada) and germinated in the greenhouse (Knoxville, TN; 35°N Lat.) at 25/20 °C (day/night) under a 16 h supplemental light at an average of 850 μmol m^{-2} s^{-1}. At 30 days after seeding, the plantlets were transferred to 11-L Dutch pots (Tek Supply, Dyersville, IA) filled with Sunshine® Pro Soil Conditioner (Sungro Horticulture, Agawam, MA). Tomato plants were grown hydroponically with a tomato fertilization program developed at the University of Tennessee. Elemental concentrations of the nutrient solutions were (mg L^{-1}): nitrogen (N; 180), phosphorus (P; 93.0), potassium (K; 203.3), magnesium (Mg; 48.6), sulfur (S; 96.3), iron (Fe; 1.0), boron (B; 0.25), manganese (Mn; 0.25), zinc (Zn; 0.025), copper (Cu; 0.01), and molybdenum (Mo; 0.005). Two identical experiments were conducted. The first experiment was completed in fall 2011 and the second in spring 2012. The experimental design was a randomized complete block with a 3×2 factorial, which consisted of six blocks and two replications of each treatment with individual pots representing an experimental unit. There were four tomato plants per treatment per replications. Ca was applied at three different treatment levels of 60, 90, or 180 mg L^{-1}. Ca treatments were applied to the plants via irrigation lines. ABA (Valent BioScienes, Inc., Libertyville, IL, USA) treatments were applied as a foliar spray at concentrations of 0.0 and 500 mg L^{-1}. Concentrations of the ABA treatments were determined by Valent BioScience from previous independent research data. ABA treatments were mixed with Latron B1956 surfactant was added to treatments at 0.5 mL L^{-1}. ABA spray treatments were applied each week until dripping from the foliage. No ABA reached the root zone. Fruit tissues were harvested 84–90 days after seeding. Subsequently, fruit were sorted by the use of USDA tomato color for red ripe (U.S.D.A, 1975) and size classification into extra-large (XL), large (L), medium (M) and small (S) fruit (U.S.D.A, 2007). Tomato fruit with BER were categorized separately. Fruit from each treatment were separated by replication and were weighed for yield. At least three fruit from two clusters for each experimental unit were separated into proximal and distal fractions in preparation for elemental nutrient analysis. Harvested fruit samples were stored at −80 °C prior to analysis. Leaf samples were taken from two clusters per plant at the last harvest for analysis of Ca.

2.2. Elemental nutrient determination

Nutrient analysis was conducted according to Barickman et al. (2014). Briefly, analysis of samples was performed using a 5.0 g subsample of fresh fruit tissue, which was combined with 10 ml of 70% HNO_3 and digested in a microwave digestion unit (Model: Ethos, Milestone Inc., Shelton, CT). The microwave temperature was ramped to 140 °C for 5 min at 1000 W and 2000 kPa, followed by an increase to 210 °C for 10 min at 1000 W and 3000 kPa. Furthermore, microwave temperature was held at 210 °C for 10 min at 1000 W and 4000 kPa and cooled for 10 min at 0 W and 2000 kPa. The digest was then allowed to cool to 20 °C. A 100 μL subsample of the digest was diluted with 9900 μL of ICP-MS matrix consisting of 2% HNO_3 and 0.5% HCl (v/v). Leaves were collected and triple rinsed with de-ionized water and dried for 48 h in a forced air oven (model large; Fisher Scientific, Atlanta, GA) at 65 °C. Dried samples were ground to homogeneity using liquid nitrogen, and 0.5 g sub-samples were weighed for analysis. Samples were microwave digested, and a 100 μL aliquot of the digested sample was diluted with 9900 μL of ICP-MS matrix for analysis. Nutrient analysis was conducted using an inductively coupled plasma mass spectrometer (ICP-MS; Agilent Technologies, Inc., Wilmington, DE). The ICP-MS system was equipped with an octapole collision/reaction cell, Agilent 7500 ICP-MS ChemStation software, a Micromist nebulizer, a water-cooled quartz spray chamber, and a CETAC (ASX-510, CETAC Inc., Omaha, NE) autosampler. The instrument was optimized daily in terms of sensitivity (Li, Y, Tl), level of oxide (Ce), and doubly charged ion (Ce) using a tuning solution containing 10 μg l^{-1} of Li, Y, Tl, Ce, and Co in a 2% HNO_3/0.5% HCl (v/v) matrix. Tissue nutrient concentrations are expressed on a dry weight (DW) basis.

There were no statistical differences in the data between the two experiments. Therefore, the data was pooled and analyzed together for treatment means. The experimental design was a randomized complete block in a factorial arrangement. The three Ca treatment concentrations were subdivided into ABA and non-ABA treated plants. Three separate clusters of tomato fruits were collected within each factorial arrangement and measured for total tissue Ca in the proximal and distal portions. Furthermore, leaves from the three clusters were analyzed for total tissue Ca. Statistical analysis of data was performed using SAS (version 9.3; SAS Institute, Cary, NC). Data were analyzed using the PROC GLIMMIXED analysis of variance.

3. Results

The statistical analysis of the results indicated that there was no interaction between ABA and Ca treatments on tomato leaf tissue Ca concentrations. Therefore, the following results are presented