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Impact of staggered treatments of novel molecules and ethylene absorbents on postharvest fruit physiology and enzyme activity of 'Santa Rosa' plums

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ABSTRACT

The present investigation was conducted to study the effect of staggered removal of cold stored (2 °C) plums at 7, 14 and 21 days interval and their subsequent treatment with salicylic acid (SA), nitric oxide (NO) and ethylene absorbent (EA) sachets. The fruit were then stored at supermarket conditions (20 ± 1 °C and 90 ± 5% RH) with the objective to know whether delayed EA, SA and NO treatments still have significant beneficial effects on the plum fruit quality. The observations on different physiological and biochemical parameters were taken at 2 days interval. The results showed that staggered treatments enhanced postharvest life and maintained fruit quality. We observed that SA-treated plums showed the highest fruit firmness and lowest decay losses when plums were either removed on 7th, 14th or 21st days of cold storage. Furthermore, SA-treated fruit exhibited lowest rates of respiration and ethylene evolution; phenylalanine ammonia lyase and pectin methyl esterase activities; minimum malondialdehyde content and lowest electrolyte leakage in comparison to those treated either with NO or packed with EA sachets or control fruit. In conclusion, 'Santa Rosa' plum removed after 7th (staggered-I), 14th (staggered-II) and 21st day (staggered-III) from cold storage maintained a shelf life of 10, 6 and 4 days, respectively at subsequent supermarket storage conditions. The overall results submit that even if the plums are not treated immediately or within few days after harvest and placed as such in cold store, they can be still treated with SA, NO or in-package ethylene absorbent (EA) treatment for beneficial postharvest influences.

1. Introduction

The aim of applying any postharvest treatment or technology is to enhance the useful marketing duration of the fruit crop during the handling, storage, transport and distribution to maintain the fruit quality, nutritive quality and the market value of the produce for the final consumption over that attainable by the use of cold storage only. Among many postharvest techniques, salicylic acid, nitric oxide and ethylene absorbent sachets treatments have shown good potential in sustaining the fruit quality and enhancing the postharvest life in many fruit (Huang et al., 2008; Zhang et al., 2008; Singh et al., 2009; Asghari and Aghdam, 2010; Manjunatha et al., 2010; Luo et al., 2011; Sharma et al., 2012a,b; Barman and Asrey, 2014; Barman et al., 2014a,b). Salicylic acid (SA), a well-known endogenous plant growth regulator, is a natural and safe phenolic compound which displays a high potential in controlling postharvest decay losses, maintaining fruit quality and decreasing over all losses of horticultural crops (Asghari and Aghdam, 2010; Luo et al., 2011). The nitric oxide (NO) is a highly reactive free radical gas, which acts as a multifunctional signaling molecule in many of the physiological processes occurring in plants which has also proven to reduce decay losses and extend shelf life of several fruit by various mechanisms (Wendehenne et al., 2004; Manjunatha et al., 2010). The ethylene absorbent (EA) sachets containing $KMnO_4$ in crystals or powder form remove ethylene from the surrounding environment of fruit by absorbing and oxidizing it to produce CO_2 and H_2O.

The plum fruit cultivar 'Santa Rosa' is climacteric in nature (Sharma et al., 2012b). Plums show a rapid softening and deterioration leading to the loss of fruit quality and very limited postharvest life (Singh et al., 2009; Sharma et al., 2012a). The plums are very rich in antioxidants and are termed as super fruit by many due

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to its health benefitting qualities comparable to strawberry and blueberry and higher than even apple and peaches (Wang et al., 1996). The Japanese plums particularly the 'Santa Rosa' cultivar is generally grown in India due to its prolific bearing habit, unique and wholesome taste. However, it has a very limited postharvest life both under ambient conditions and cold storage. The lack of proper postharvest handling and storage infrastructure in India further limits the availability and advances the deterioration of this perishable fruit. In spite of many postharvest technologies for enhancing the shelf-life of the plum fruit, very few or none at all are practically being used on commercial scale by the farmers or the traders in India. Moreover, plums are also affected by chilling injury when stored at low temperatures for long durations depending upon many factors including pre harvest environmental factors, variety, species, storage temperature and duration (Luo et al., 2011). So there is a need to develop other techniques to enhance its postharvest life along with low temperature storage. This study was framed to ascertain whether staggered treatments of plums kept in cold storage (2 ± 1 °C and 90 ± 5% RH) after 7, 14 and 21 days interval with salicylic acid (SA), nitric oxide (NO) and ethylene absorbent sachets (EA), followed by storage at supermarket conditions (20 ± 1 °C and 90 ± 5% RH) show beneficial influences or not and to determine its extent. This is the first report on the effects of staggered treatments of salicylic acid (SA), nitric oxide (NO) and ethylene absorbent sachets (EA) on the postharvest life, decay loss and quality of 'Santa Rosa' plums.

2. Materials and methods

2.1. Fruit material and experimental site

'Santa Rosa' plums were harvested in June 2012–2013 from an orchard at Katrain, Kullu, Himachal Pradesh (latitude 32°11′N and longitude 77°15′E), India. The fruits were sorted and the bruised or diseased fruit were removed. The plums were then packed in corrugated fiber boxes (CFB) and transported to IARI, New Delhi by road, where the fruits were again sorted. The mean temperature during transport was 25 °C and it took about 12 h to reach New Delhi. The plums affected with compression injury, bruising and disease etc., were removed and fruit of uniform size, color and climacteric maturity stage were placed in cold storage (2 °C and 90 ± 5% RH) without any treatment for 21 days. Fruit samples were withdrawn after 7th, 14th and 21st day of storage (Staggered-I, Staggered-II, Staggered-III experiments, respectively), and then treated with EA sachets, SA (2.0 mM) and sodium nitroprusside (0.5 mM). The total number of fruit taken for the experiment was 2160 which were divided into 30 punnets for every treatment, each punnet containing six plums. The experiment was conducted in three replications for every treatment.

2.2. Treatments of plums

For SA treatment, the fruits were dipped in aqueous solution of 2.0 mM SA for 10 min at 20 °C. Similarly, fruits were dipped in aqueous solution of 0.5 mM sodium nitroprusside (SNP, a nitric oxide donor) for 5 min at 20 °C. Both SA and sodium nitroprusside aqueous solutions were made in 10 L plastic bucket. The ethylene absorbent sachets (Bioconservacion) were provided by Poligono Industrial El Regas, Barcelona, Spain. The EA sachets treatment was done by placing two ethylene sachets in each punnet containing six plums. After treatments, the plums were placed in retail supermarket conditions (20 ± 1 °C and 90 ± 5% RH) and observations on different parameters recorded at 2 days interval. Each treatment in every staggered experiment set consisted of 30 punnets each containing six plums.

2.3. Fruit firmness and fruit decay

The fruit firmness in 'Santa Rosa' plums was determined in 3 fruit per treatment after each 2 days interval using a texture analyzer (model: TA + Di, Stable micro systems, UK) by puncture test with a cylindrical probe (2 mm diameter) and expressed in Newton (N). The plum fruit showing any visible symptoms of the disease occurrence regardless of its severity were considered rotten, counted and expressed as percentage of the total number of fruits.

2.4. Respiration and ethylene production rates

The respiration rate was measured as the rate of CO_2 production using the CO_2:O_2 gas analyzer (Model: Checkmate 9900 O_2/CO_2, PBI Dansensor, Denmark) and expressed as mL CO_2 kg^{-1} h^{-1}. It was done by placing two plum fruit each in 3 trap boxes with three replications for each treatment for 1 h duration at 20 °C in 500 mL airtight containers having twist-top lid fitted with a silicone rubber septum at the center of the lid for every staggered treatment. The ethylene production rate was determined by using the static headspace technique (Hewlett Packard gas chromatograph model 5890 Series II equipped with a flame ionization detector (FID), Porapack-N 80/100 mesh packed stainless steel column and a H.P. integrator). The ethylene production rate was determined in 2 fruit in each trap box, replicated three times per staggered treatment and expressed as μL kg^{-1} h^{-1}.

2.5. Electrolyte leakage (EL) determination and malondialdehyde (MDA) content

The electrolyte leakage (EL) and malondialdehyde content (MDA) were determined as described by Luo et al. (2011) with slight changes. The observations were taken after removing the fruit from the cold storage and keeping them at ambient conditions for 3 h for EL. The electrolyte leakage was measured using the conductivity meter (model DB-1040). The fruit discs were excised from the plum fruit with a 2-mm diameter cork borer, incubated in 30 mL of 300 mM mannitol for 3 h in a 50 mL capped centrifuge tube. The conductivity of the solution was measured after 3 h of incubation at 25 °C using a DB-1040 conductivity meter. The disks and bathing solution were then stored at −20 °C for at least 24 h, boiled in water for 30 min, cooled to room temperature, and total electrolyte conductivity was measured once again. It was expressed as percentage for EL. Malondialdehyde content was determined in 3 fruit per treatment at each interval for every staggered experiment set. Fruit (2.0 g) was ground in liquid nitrogen and extracted in 10% (w/v) trichloroacetic acid (TCA). After centrifugation at 10,000 × *g* for 15 min, 2 mL aliquot of the supernatant was mixed with 2 mL 10% (w/v) TCA containing 0.6% (w/v) thiobarbituric acid (TBA) and then subjected to heating up to 100 °C for 20 min, quick cooling and centrifugation at 10,000 × *g* for 10 min. The supernatant was collected and absorbance was recorded at 532, 600 and 450 nm. Finally, malondialdehyde content in fruit was expressed as nmol g^{-1} FW.

2.6. Pectin methyl esterase (PME) and Phenylalanine ammonia lyase (PAL) enzyme activities

The PME enzyme activity in 'Santa Rosa' plums was determined by following the procedure of Hagerman and Austin (1986) and was expressed as μmol min^{-1} g^{-1} FW. Five gram of fruit pulp was homogenized in 15 mL of cold (4 °C) 8.8% sodium chloride solution using pestle and mortar. The homogenate was then centrifuged at 15,000 × *g* for 15 min. The supernatant was collected and its pH was adjusted to 7.5 with NaOH, after which it was used for enzyme assay. In a cuvette, 2.0 mL of pectin was mixed with 0.15 mL of