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RESEARCH ARTICLE

Effect of pre-harvest spray of putrescine on shelflife and quality of tomato during storage

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Putrescine (PUT) is a polyamine responsible for plant growth and development, fruit ripening, stress response, senescence and in higher quantities to maintain post-harvest quality in vegetables. Tomato (cv. Punjab Ratta) crop grown during 2012-13 and 2013-14 at Vegetable Research Farm, Punjab Agricultural University, Ludhiana, Punjab, India replicated thrice treated two times at 120 and 130 days after transplanting with putrescine at 1 mmol/L, 2 mmol/L and 3 mmol/L concentrations and water sprayed in control corresponding to T₁, T₂, T₃ and T₄ respectively. On following the day of second spray, fruits of uniform size and weight at mature green to breaker stage were picked, taken in plastic crates and stored at 13±2°C and relative humidity of 85-90% for further storage studies. To find out the effect of pre-harvest application of putrescine on tomato during storage, representative sample fruits from the treatments were analysed at 5 day interval i.e at 1st, 6th, 11th, 16th, 21st and 26th day of storage for various quality parameters and average of two year readings was presented. T₁ (PUT at 1 mmol/L) treatment showed significant lowest cumulative physiological loss in weight % compared to other putrescine treatments at all the days of analysis except at 6th day after storage. However, at the end of the storage period, putrescine treatments T₁ (0.58%) and T₂ (0.58%) were significantly higher in titratable acidity than control. All the putrescine treatments able to maintain decay % below threshold level (10%) upto 16th day of storage while, in control it was upto 11th day of storage only.

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Introduction

Tomato (*Solanum lycopersicum* L.) is one of the most important vegetable crops cultivated all over the world for its fleshy fruits. Tomato is considered as protective as well as productive food because of its special nutritive value and also wide spread production. It is a rich source of lycopene, ascorbic acid, β-carotene and minerals and tomatoes ranked number one among vegetables contributing vitamins and minerals (Rick, 1978). It has attracted the attention due to the anti-carcinogenic and antioxidant property of lycopene and ascorbic acid. Lycopene of tomato is an efficient quencher of singlet oxygen and free radicals which provides protection against a broad range of epithelial cancers (Mascio *et al.*, 1991).

Tomato being a climacteric fruit, the start of ripening is accompanied by a rapid rise in respiration rate called 'respiratory climacteric' during which oxidative breakdown of complex substrates occur, ageing follows, leading to product deterioration. Also tomatoes being fleshy fruits, continue to lose water after harvest. This results in a wilted, dull appearance that reduces the eye appeal and freshness and eventually becomes unmarketable. Several growth regulators and other chemicals have been reported to delay the ripening and extend the post-harvest life of

many fruits (Khader *et al.*, 1988). Polyamines have been found to be anti-senescence agents, with changes in their levels considered as a protective mechanism in plant tissues. Usually, the concentration of polyamines decreases during tissue senescence with accelerated ethylene production (Valero *et al.*, 2002). Polyamine compounds present in living cells have been involved in plant growth and development, fruit ripening, stress response and senescence (Malik and Singh, 2003). Exogenous application of polyamines delay the fruit senescence and physiological processes leading to the fruit ripening. Putrescine ($C_4H_{12}N_2$) is one of polyamine compounds present in living cells and a higher endogenous level of putrescine (PUT) is associated with delayed fruit ripening (Dibbille *et al.*, 1988). The reduced rate of softening of long-keeping tomato cultivars has been correlated with elevated putrescine (PUT) (Dibbille *et al.*, 1988; Saftner and Baldi, 1990). It is reported that comparatively higher levels of polyamines and lower level of ethylene have been found in long keeping tomato (Dibbille *et al.*, 1988). The present study was carried out to find out the effect of pre-harvest application of putrescine at different concentrations during storage of fresh tomatoes at $13\pm 2^\circ C$ and relative humidity of 85-90%.

The retardation of chlorophyll loss in muskmelon with exogenous application of polyamines has been attributed to reduced hydrolytic activities acting on chloroplast thylakoid membranes (Lester, 2000). Exogenous application of putrescine on strawberry, apricot and sweet cherry fruits resulted in reduced ethylene production that prevented softening of fruit tissues during storage period (Khosroshahi and Ashari, 2008). Similarly, Law *et al.* (1991) reported that tomato fruits treated with putrescine showed an increase in fruit firmness and delayed ethylene production.

Materials and Methods

An experimental plot of tomato (cv. Punjab Ratta) was grown at Vegetable Research Farm, Punjab Agricultural University, Ludhiana, Punjab, India during 2012-13 and 2013-14 and replicated thrice in the field. Aqueous solutions of putrescine prepared at 1 mmol/L, 2 mmol/L and 3 mmol/L concentrations corresponding to T_1 , T_2 and T_3 respectively and distilled water in control (T_4) were used to spray two times at 120 and 130 days after transplanting. Harvesting of fruits was done one day after second spray of treatment imposition. Fruits of uniform size and weight picked at mature green to breaker stage from the treated plots brought to the laboratory and washed properly with normal tap water to remove the field heat and blemishes on the fruits followed by drying with whatmann paper. Later, fruits taken in plastic crates stored at $13\pm 2^\circ C$ and relative humidity of 85-90% for further storage studies at Post-harvest laboratory of the Department of Fruit Science, PAU, Ludhiana. In each treatment, 5kg fruits were taken and representative sample of 3 fruits were used for analysis at 5 day interval i.e at 6th, 11th, 16th, 21st and 26th days of storage for various quality parameters like physiological loss in weight (%), titratable acidity (% citric acid), ripening index (TSS/TA), firmness (Kgf) and decay % and data obtained was presented after taking the average of two years.

Physiological loss in weight % (PLW %)

A separate set of fruits were stored in plastic crates and undisturbed throughout the storage period and physiological loss in weight (PLW) of fruits was calculated on initial weight basis. The per cent loss in weight after each storage interval was calculated by subtracting final weight from the initial weight of the fruits and then converted into percentage value. The cumulative loss in weight was calculated on fresh weight basis.

$$\text{Physiological loss in weight \% (PLW \%)} = \frac{(\text{Initial fruit weight} - \text{Final fruit weight})}{\text{Initial fruit weight}} \times 100$$

Titrateable acidity (% citric acid) and Ripening Index (TSS/TA)

Titrateable acidity was determined by titrating the 2ml juice sample against 0.1N NaOH along with phenolphthalein as an indicator and expressed as % citric acid per 100g fresh weight. A ratio was calculated by dividing the values of TSS and that of corresponding values of titrateable acidity and regarded as ripening index (Sabir and Agar, 2011).

Fruit firmness (kgf)

Firmness of randomly selected fruits (three from each replication) was measured with the help of fruit pressure tester (Model FT-327, USA) on two opposite sides of the equatorial axis of fruit and average values expressed in terms of kgf.

Decay %

The decay percentage of fruits was calculated on number basis by counting the spoiled fruits in each replication and total number of fruits per replication.

$$\text{Decay \%} = (\text{Number of spoiled fruits} / \text{Total number of fruits}) \times 100$$

Experimental design and statistical analysis:

The experiment was arranged in Completely Randomized Design and statistical analysis was done using the SAS (v.9.3, SAS Institute, Cary, NC, USA) and least squares means separated by the Tukey's test at 1% significance level.

Results and Discussion

Physiological Loss in Weight % (PLW %)

In all the treatments studied, cumulative physiological loss in weight % increased progressively with the increase in storage period (Table 1). Significant differences were observed between putrescine treatments and control throughout the storage period. However, among the putrescine treatments, T₁ (PUT at 1 mmol/L) treatment showed significant lowest cumulative physiological loss in weight % compared to other putrescine treatments at all the days of analysis except at 6th day after storage. But, less than threshold level of PLW% (5%) was recorded in T₁ treatment upto 16th day of storage, T₂ and T₃ treatments upto 11th day of storage while in control more than threshold level was recorded at 6th days of storage only indicating treatment with putrescine reduced the moisture loss and probably reduced the respiration rate also during storage and thus further helping in extension of storage life of the tomato fruits. However, Kaur (2011) reported that pre-harvest spray of putrescine at 3 mmol/L and 2 mmol/L on peach fruits of cv. 'Shan-i-Punjab' reduced the physiological loss in weight during cold storage.

Titratable Acidity (% citric acid) and Ripening index (TSS/TA)

The data in table 2 indicated that there were significant differences in titratable acidity between putrescine treatments and control at all the days of analysis. However, at the end of the storage period, putrescine treatments T₁ (0.58 %) and T₂ (0.58 %) were significantly higher in titratable acidity than control. The higher titratable acidity in putrescine treated fruits may be due to the decreased hydrolysis of organic acids and subsequent accumulation of organic acids (Pool *et al.*, 1972). Pre-harvest application of putrescine on 'Kensington Pride' mango resulted in high acidity as compared to control up to 20 days (Malik *et al.*, 2003). Similarly, putrescine treated fruits of 'Angelino' plum as pre-harvest application showed higher titratable acidity than the control fruits up to six weeks of storage (Khan and Singh, 2008).

Likewise, in ripening index significant differences were observed between putrescine treatments and control at all the days of analysis except at 1st and 11th day of storage (Table 3). At the end of storage, treatments T₁ and T₂ showed significant lower ripening index than control which might be due to delayed reduction of acids to sugars by putrescine. Similar kind of results reported by Khan and Singh (2010) wherein application of 2 mM putrescine on Japanese plum one week before anticipated commercial harvest resulted in reduced total soluble solids: titratable acidity ratio during storage.

Firmness (Kgf)

A rapid decrease in firmness in control while, relatively slower decrease in firmness in putrescine treatments was observed (Table 4). A significant higher firmness was recorded in putrescine treatments over control at all the days of analysis except by T₂ and T₃ treatments at 6th day of storage and T₁ treatment at 21st day of storage. Though all the treatments on 1st day of storage non-significantly differed in initial firmness, at the end of the storage all the putrescine treatments showed higher significant firmness than control. Pre-harvest application of putrescine has resulted in retardation of plum fruit softening during low temperature storage through suppressed ethylene biosynthesis and reduced activities of fruit softening enzymes (Khan *et al.*, 2007).

Decay %

All the putrescine treatments showed significant lower cumulative decay % over control throughout the storage period except at 6th day of storage and T₁ treatment at 11th day of storage (Table 5). Cumulative decay % of fruits increased with the progression of the storage period. It was observed that all the putrescine treatments able to maintain decay % below threshold level (10%) upto 16th day of storage while, in control it was upto 11th day of storage only. Pre-harvest treatments with putrescine was done to enhance shelf life of 'Navel Oranges' resulted in the lowest fruit decay per cent as compared to control (Marzouk and Kassem, 2011).

Table 1: Effect of pre-harvest application of putrescine on physiological loss in weight % (PLW %) of fresh tomatoes during storage at 13±2^oC and RH of 85-90%

Treatments	Particulars	Days of storage					
		1 st	6 th	11 th	16 th	21 st	26 th
T ₁	Putrescine 1 mmol/L	0	1.55 ^b	3.45 ^c	4.59 ^d	5.53 ^d	6.51 ^d
T ₂	Putrescine 2 mmol/L	0	1.66 ^b	4.53 ^b	5.75 ^c	8.35 ^b	9.79 ^b
T ₃	Putrescine 3 mmol/L	0	1.92 ^b	4.79 ^b	6.96 ^b	7.86 ^c	9.04 ^c
T ₄	Water spray (control)	0	5.19 ^a	7.55 ^a	8.90 ^a	9.74 ^a	11.31 ^a

(Data in column followed by different letter superscripts are significantly different at $P \leq 1\%$)

Table 2: Effect of pre-harvest application of putrescine on titratable acidity (% citric acid) of fresh tomatoes during storage at 13±2 °C and RH of 85-90%

Treatments	Particulars	Days of storage					
		1 st	6 th	11 th	16 th	21 st	26 th
T1	Putrescine 1 mmol/L	1.03 ^a	0.79 ^b	0.83 ^a	0.77 ^{ab}	0.45 ^d	0.58 ^a
T2	Putrescine 2 mmol/L	1.02 ^a	0.77 ^{bc}	0.81 ^{ab}	0.81 ^a	0.58 ^b	0.58 ^a
T3	Putrescine 3 mmol/L	1.05 ^a	1.07 ^a	0.45 ^c	0.66 ^c	0.68 ^a	0.41 ^c
T4	Water spray (control)	1.03 ^a	0.74 ^c	0.71 ^b	0.73 ^b	0.52 ^c	0.45 ^b

(Data in column followed by different letter superscripts are significantly different at $P \leq 1\%$)

Table 3: Effect of pre-harvest application of putrescine on ripening index of fresh tomatoes during storage at 13±2°C and RH of 85-90%

Treatments	Particulars	Days of storage					
		1 st	6 th	11 th	16 th	21 st	26 th
T1	Putrescine 1 mmol/L	4.90 ^a	9.19 ^a	7.45 ^b	7.21 ^{bc}	9.94 ^b	8.26 ^c
T2	Putrescine 2 mmol/L	4.95 ^a	9.02 ^a	7.82 ^b	6.76 ^c	8.70 ^c	8.25 ^c
T3	Putrescine 3 mmol/L	4.85 ^a	6.18 ^b	13.54 ^a	8.31 ^a	8.54 ^c	11.78 ^a
T4	Water spray (control)	4.90 ^a	9.16 ^a	8.31 ^b	7.73 ^b	10.74 ^a	10.21 ^b

(Data in column followed by different letter superscripts are significantly different at $P \leq 1\%$)

Table 4: Effect of pre-harvest application of putrescine on firmness (Kgf) of fresh tomatoes during storage at 13±2°C and RH of 85-90%

Treatments	Particulars	Days of storage					
		1 st	6 th	11 th	16 th	21 st	26 th
T1	Putrescine 1 mmol/L	51.50 ^a	49.47 ^a	50.24 ^a	46.39 ^{ab}	31.05 ^{bc}	28.64 ^a
T2	Putrescine 2 mmol/L	52.05 ^a	48.20 ^b	46.75 ^b	47.22 ^a	34.94 ^a	25.42 ^b
T3	Putrescine 3 mmol/L	51.85 ^a	45.81 ^c	46.61 ^b	45.64 ^b	31.89 ^b	29.64 ^a
T4	Water spray (control)	51.66 ^a	47.21 ^b	38.81 ^c	33.11 ^c	29.08 ^c	19.78 ^c

(Data in column followed by different letter superscripts are significantly different at $P \leq 1\%$)

Table 5: Effect of pre-harvest application of putrescine on decay % of fresh tomatoes during storage at 13±2°C and RH of 85-90%

Treatments	Particulars	Days of storage					
		1 st	6 th	11 th	16 th	21 st	26 th
T1	Putrescine 1 mmol/L	0	0.84 ^a	7.63 ^a	9.69 ^b	11.40 ^{bc}	18.14 ^b
T2	Putrescine 2 mmol/L	0	0.50 ^a	4.04 ^b	8.48 ^b	11.05 ^c	17.54 ^b
T3	Putrescine 3 mmol/L	0	1.70 ^a	4.70 ^b	8.58 ^b	14.21 ^b	19.80 ^b
T4	Water spray (control)	0	2.66 ^a	8.17 ^a	14.99 ^a	23.41 ^a	32.97 ^a

(Data in column followed by different letter superscripts are significantly different at $P \leq 1\%$)

Conclusion

Tomato fruits treated with PUT at 1 mmol/L, 2 mmol/L and 3 mmol/L are observed with significant lower cumulative physiological loss in weight % compared to control while, all the PUT treatments retained cumulative decay % below 10% threshold level upto 16th day of storage. However, all the PUT treatments maintained significant higher firmness over control (water spray) at the end of the storage (26 days).

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