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Preharvest applications of putrescine influences the storage life and quality of pear fruit

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ABSTRACT

Pre-harvest foliar applications of putrescine (PUT) were given to extend the storage life and maintain the quality of pear fruit *cv*. Punjab Beauty during the cold storage. Pre-harvest sprays of 1mM, 2mM & 3mM PUT were applied 7 days before harvest (DBH) and 14DBH. Harvested fruit were stored at 0-1°C and 90-95% RH for 75 days. Fruit were analysed on 0, 15th, 30th, 45th, 60th, 67th & 75th day for various physico-chemical and enzymatic changes. The minimum weight loss (5.47%) and highest average sensory quality (SQ) (7.25), starch content (0.97 mg/g FW) & titratable acidity (0.23%) were maintained in 3mM PUT (14DBH) treated fruit at the end of storage, which was at par with 2mM PUT (14DBH) treatment. These treatments were also helpful to delay the changes in colour and enzymatic activities [Pectin methyl esterase (PME) & cellulase activity] during cold storage and retained higher soluble solids content (SSC) at the end of storage. The results revealed that, 2mM & 3mM PUT (14DBH) applications were effective to extend the storage life and maintain the fruit quality as compared to the control during storage.

Key word: Cellulase, fruit quality, pear, PME, putrescine.

INTRODUCTION

Pear (*Pyrus* spp) is one the most important temperate zone fruit crop of India having high economic value. In north-western plains of India, some low chilling semi-soft cultivars of pear are commercially recommended. 'Punjab Beauty' is a promising cultivar of semi-soft pear (Singh and Dhillon, 16). It matures in the third week of July when the temperature and humidity are very high that lead to a reduction in the shelf life of the fruit. Fresh pear fruit have 85-90% moisture content which prone to lose after harvest by transpiration and respiration mechanism, cause visual degradation, loss of succulence and firmness due to shrivelling (Xanthopoulos, 18). Proper storage, various chemical treatments (fungicides, growth regulators and nutrients), waxing and packaging can check the respiration, transpiration and delay any objectionable disease infection and biochemical changes.

Polyamines having poly-cationic nature and small organic metabolites, interact with phospholipids, proteins and nucleic acid which are negatively charged molecules leads to antioxidant properties and enhance the ability of the cell to protect from abiotic stress (Kusano *et al.*, 12). These cationic aliphatic amines are the antagonist to ethylene production, as they share the common biosynthesis precursor i.e. *S*-Adenosyl Methionine. Earlier studies revealed that PUT significantly improved the quality and enhanced the storage life of apricot (Davarynejad *et al.*, 5), mango (Razzaq *et al.*, 15) and pear (Hosseini *et al.*, 8). Keeping in view the effects of PUT, the present research was conducted to inspect the effect of pre-harvest application of PUT on the extension of storage life and storage behaviour of pear [*Pyrus pyrifolia* (Burm) Nakai] fruit *cv*. Punjab Beauty under cold storage conditions.

MATERIALS AND METHODS

Pre-harvest applications of PUT were given at 7 & 14DBH on uniform and healthy 24 plants of pear *cv*. Punjab Beauty during 2016 and 2017, while four control plants were sprayed with water at the Fruit Research Farm, Punjab Agricultural University, Ludhiana. The experiment was comprised of seven treatments *viz*. 1mM, 2mM & 3mM PUT sprays 7DBH and same concentration 14DBH, while controls with water. Fruit from the experimental plants were harvested at a mature stage (69.68±5.00N firmness, 12.08±0.50% SSC) and shifted instantly in PVC crates to the Post-harvest, Laboratory for storage studies. Harvested fruit were disinfected with 100ppm chlorinated water before packaging in 3 ply CFB boxes (5% ventilation). One kilogram pear fruit were packed for storage study from each replication of every treatment. Packed fruit were kept in cold storage (0-1°C & 90-95% RH) and analysed for physico-chemical and enzymatic changes on 0, 15th, 30th, 45th, 60th, 67th and 75th days of storage.

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The weight loss of fruit was calculated on the initial weight basis and expressed in per cent. The SQ of fruit was evaluated by a panel of 5 judges on the basis of general appearance, taste, texture and flavour of fruit on Hedonic scale (1-9) as described by Amerine *et al.* (1). Firmness of ten randomly selected fruit was measured with the help of a stand mounted penetrometer (Model FT-327, USA) using stainless steel probe at two opposite points on the fruit's equator and expressed in terms of newton (N) force. The spoilage per cent of fruit was calculated on the number basis by counting the spoiled fruit at each storage interval and expressed in per cent. SSC of juice was *determined with the help of ATAGO digital hand refractometer in terms of Brix (%). These readings were corrected with the help of temperature correction chart at 20 °C temperature. Titratable acidity was recorded as per* AOAC (2) *and expressed in per cent of malic acid.* Pear skin colour was recorded from opposite positions of each fruit in *Commission International de L'Eclairage* (CIE) units by using a Color Flex 45°/0° spectrophotometer (Hunter Lab Color Flex, Hunter Associates Inc., Reston, VA, USA) and expressed' as *b** hunter 'colour value. The PME and cellulase activities were estimated as per the method given by Mahadevan and Sridhar (13).

The data were pooled and analyzed by one-way analysis of variance (ANOVA) as per Randomized Complete Block Design with four replications and means ± SD were separated using LSD test. Differences were considered statistically significant at the level $p<0.05$ using statistical software SAS (version 9.3 for windows).

RESULTS AND DISCUSSION

Weight loss of the fruit increased with the progression of the storage period in all the treatments (table 1A). However, lowest weight loss was recorded in 3mM PUT (14DBH) application followed by 2mM PUT (14DBH) application. The highest weight loss was recorded in the control during the entire storage period. At the end of storage, 3mM PUT (14DBH) treated fruit recorded 14.13% less weight loss in comparison to the untreated fruit. Kader (9) suggested that, weight loss more than 5% is consider to be loss in the quality during storage. Similarly, Hosseini *et al* (8) reported lowest weight loss in the foliar application of 2mM PUT of banana. Cell membrane phospholipids conjugation with polyamines led to cell membrane integrity may be ascribed to lower weight loss in PUT treated fruit (Enas *et al.*, 6).

SQ of the fruit in all the treatments increased during the initial period of storage (table 1B). At the time of storage to 45th day of storage, there was no significant variation in SQ among the treatments. SQ increased up to 60th day of storage in 2mM & 3mM PUT applied 7DBH & 14DBH, while in other treatment, it increased up to 45th day of storage. On 67th day of storage, SQ was declined in all the applications; however, the highest SQ (7.59) was observed in 3mM PUT (14DBH) application and lowest (6.10) in the control. At the end of storage fruit treated with 3mM PUT (14DBH) recorded the highest SQ (4.33) which was followed by 2mM PUT treatment while lowest in the control. Similarly, Hosseini *et al* (8) reported that pre-harvest treatment of PUT (2mM) in banana fruit substantially exhibited the higher hedonic score as compared to the control fruit at the end of storage. Valero et al (17) suggested that the anti-senescence action of PUT by preventing transcription, synthesis and activity of 1-aminocyclopropane-1carboxylate-synthase and binding with pectin molecule maintained the higher sensory quality during storage of fruit.

Firmness is the major limiting factor in the storage life of pear fruit. Data presented in Fig. 1A elucidated that fruit firmness decreased with storage in all the PUT applications. At the time of storage, the highest firmness (68.77N) was recorded in 3mM PUT (14DBH) application, which was equivalent to 2mM PUT (14DBH) application, while lowest (65.32) in the control. This firmness behaviour observed throughout the storage period. At the end of storage, 3mM PUT (14DBH) treated fruit registered 50.57% higher firmness as compared to the control. Similar results were reported by Kaur and Jawandha (10) in peaches. PUT treatment maintains the higher firmness may be due to reduction in activity of cell wall degrading the enzymatic activity of pectin esterase, PME and poly-galactouronase which degrade the pectic substances in the cell wall (Valero *et al*., 17).

During storage of pear fruit, no spoilage was recorded up to 45th day in all the treatments, however, spoilage of 2% was observed in the control on the 60th day (table 1C). Results further showed that, PUT applications of 2mM & 3mM (14DBH) registered no spoilage up to 67th day. At the end of storage, spoilage in fruit was recorded in all the treatments. However, 3mM PUT (14DBH) registered the lowest spoilage of 2.55%. Similarly, Hosseini *et al.* (7) reported the lowest spoilage in pre-harvest application of 2mM PUT on pear *cv*. Spadona at the end of storage. The anti-pathogenic properties of PUT might be reduce the decay per cent of the fruit (Bal, 3).

The important quality attributes for post-harvest quality of climacteric fruit are SSC and titratable acidity. SSC of the fruit generally increased during

Table 1. Variation in weight loss (A), sensory quality (B) and spoilage (c) of pear fruit during cold storage in relation to different pre-harvest PUT treatments.

Parameters & PUT applications	Storage period days						
	0	15	30	45	60	67	75
A) Physiological loss in weight							
1mM (14DBH)	-	1.66±.11^{b}	3.06±.14^{b}	4.30±.14^{b}	5.15±.14^{b}	5.55±.15^{b}	6.08±.18^{bc}
2mM (14DBH)	-	1.28±.13^{d}	2.56±.14^{d}	3.75±.17^{d}	4.65±.18^{d}	5.10±.15^{cd}	5.58±.19de
3mM (14DBH)	-	1.18±.12^{d}	2.54±.20^{d}	3.75±.19^{d}	4.52±.14^{d}	4.98±.12^{d}	5.47±.21^{e}
1mM (7DBH)	-	1.77±.16^{ab}	3.23±.11^{a}	4.48±.13^{a}	5.35±.12^{a}	5.74±.21^{a}	6.28±.20^{ab}
2mM (7DBH)	-	1.50±.14^{c}	2.83±.18^{c}	4.12±.11^{c}	4.92±.17^{c}	5.41±.22^{b}	5.92±.20^{c}
3mM (7DBH)	-	1.29±.16^{d}	2.73±.13^{c}	3.85±.16^{d}	4.81±.17^{c}	5.20±.15^{c}	5.67±.21^{d}
Control	-	1.90±.14^{a}	3.31±.15^{a}	4.56±.16^{s}	5.45±.16^{a}	5.80±.22^{a}	6.37±.21^{a}
P	-	0.000	0.000	0.000	0.000	0.000	0.000
B) Sensory quality (1-9)							
1mM (14DBH)	7.22±.20^{a}	7.68±.35^{a}	8.21±.27^{a}	8.38±.27^{a}	8.19±.35^{ab}	7.02±.47^{bc}	3.52±.51^{cd}
2mM (14DBH)	7.12±.26^{a}	7.52±.47^{a}	8.1b±.34^{a}	8.23±.32^{a}	8.37±.27^{a}	7.39±.37^{ab}	4.41±.43^{ab}
3mM (14DBH)	7.08±.26^{a}	7.41±.40^{a}	7.72±.63^{a}	8.12±.30^{a}	8.44±.20^{a}	7.59±.55^{a}	4.33±.45^{a}
1mM (7DBH)	7.24±.24^{a}	7.73±.37^{a}	8.27±.26^{a}	8.41±.20^{a}	8.04±.24^{b}	6.67±.48^{c}	3.82±.38^{bc}
2mM (7DBH)	7.19±.30^{a}	7.63±.43^{a}	8.08±.22^{a}	8.31±.28^{a}	8.35±.26^{a}	7.23±.26^{ab}	3.7±.50^{bc}
3mM (7DBH)	7.14±.23^{a}	7.6±.52^{a}	7.99±.25^{a}	8.27±.28^{a}	8.32±.32^{ab}	7.23±.20^{ab}	4.08±.42^{ab}
Control	7.27±.21^{a}	7.84±.47^{a}	8.1±.37^{a}	8.21±.33^{a}	7.39±.54^{c}	6.1±.43^{d}	3.14±.49^{d}
P	0.648	0.515	0.053	0.426	0.000	0.000	0.000
C) Spoilage (%)							
1mM (14DBH)	0.00	0.00	0.00	0.00	0.00	1.95±.52^{b}	0.66±.64^{bc}
2mM (14DBH)	0.00	0.00	0.00	0.00	0.00	0.00	2.87±.47^{cd}
3mM (14DBH)	0.00	0.00	0.00	0.00	0.00	0.00	2.55±.59^{d}
1mM (7DBH)	0.00	0.00	0.00	0.00	0.00	2.17±.51^{b}	3.61±.63^{ab}
2mM (7DBH)	0.00	0.00	0.00	0.00	0.00	0.66±.76^{c}	3.16±.49^{bc}
3mM (7DBH)	0.00	0.00	0.00	0.00	0.00	0.44±.48^{cd}	3.05±.45^{cd}
Control	0.00	0.00	0.00	0.00	2.00±.38^{a}	3.34±.64^{a}	4.09±.43^{a}
P	-	-	-	-	0.000	0.000	0.000

Mean values followed by same letters within a column are not significantly different at *$p \leq 0.05$. n = 4 replications

the initial period of storage. In freshly harvested fruit, lowest SSC (12.08%) was recorded in 3mM PUT (14DBH) application which was statistically not different from 2mM PUT (14DBH) application (Fig. 1C). it was observed that SSC of pear fruit was increased slowly in 2mM & 3mM PUT sprayed at 7DBH & 14DBH up to 67th day; however, it increased sharply in untreated fruit up to 45th day and afterwards declined. At the end of storage, highest SSC (13.25%) was retained by 3mM PUT (14DBH) application which did not differ from 2mM PUT application, while lowest SSC registered in the control. SSC of fruit increased during storage as a result of the conversion of polysaccharides into soluble solids by the process of dehydration and hydrolysis. Razzaq *et al.* (15) also reported in mango that SSC content during storage was increased, while PUT treated fruit recorded lower increase in SSC as 1.20 fold lower than control, which may be ascribed to ethylene suppression that further affected the acid and sugar metabolism.

Titratable acidity continuously decreased during the storage of fruit (Fig. 2A). At the time of storage, highest titratable acidity (0.39%) was recorded in 3mM PUT (14DBH) application, while the lowest (0.32%) in the control. At the end of storage, highest titratable acidity was registered in 3mM PUT (14DBH) application which was not significantly ($p \leq 0.05$) from

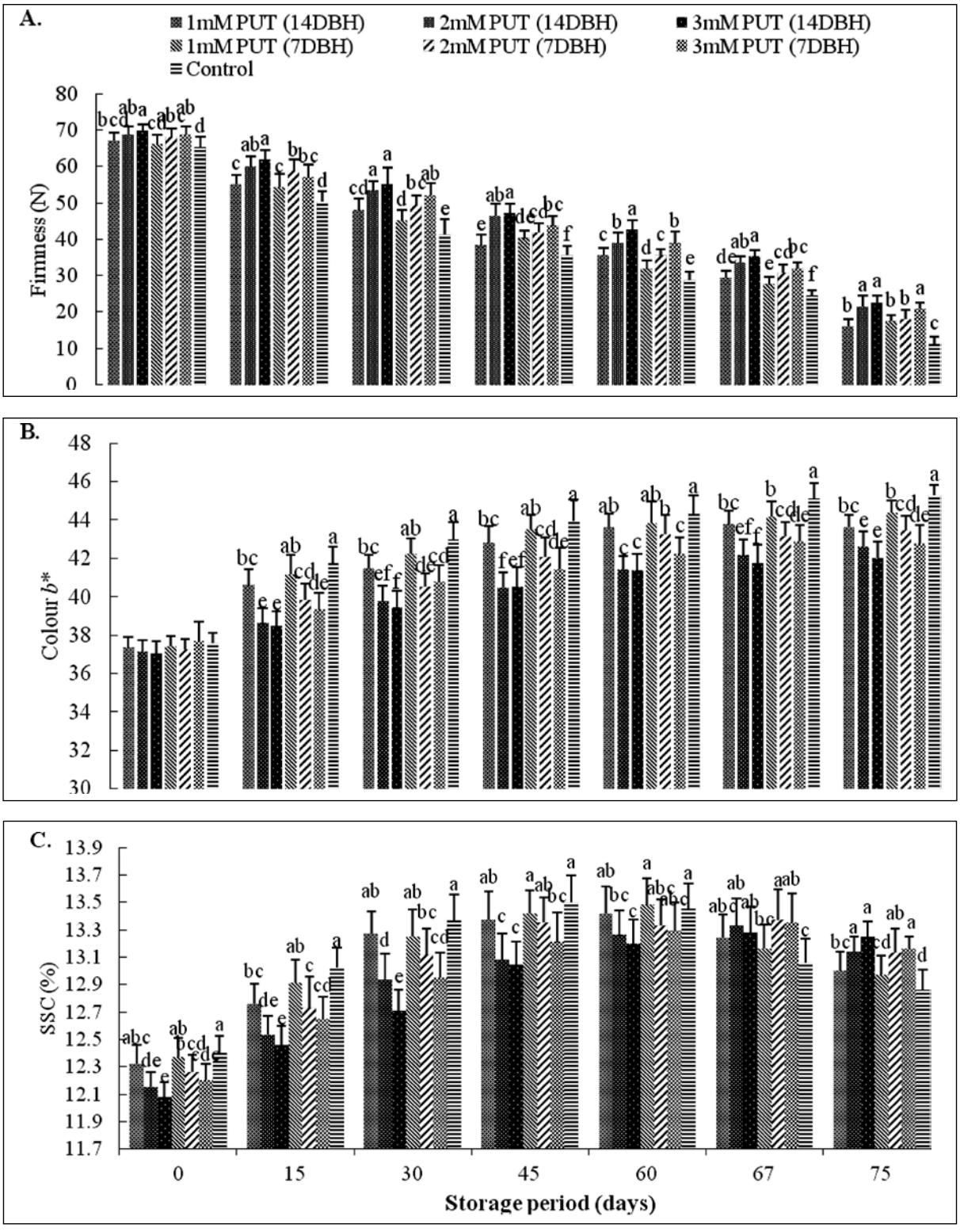

Fig. 1. Variation in firmness (A), colour *b** (B) and SSC (C) of pear fruit during cold storage in relation to different pre-harvest PUT applications. Vertical bars represent ± S.D. of means for 4 replicates. Mean values followed by same letters within a column are not significantly different at $^{*}p \leq 0.05$.

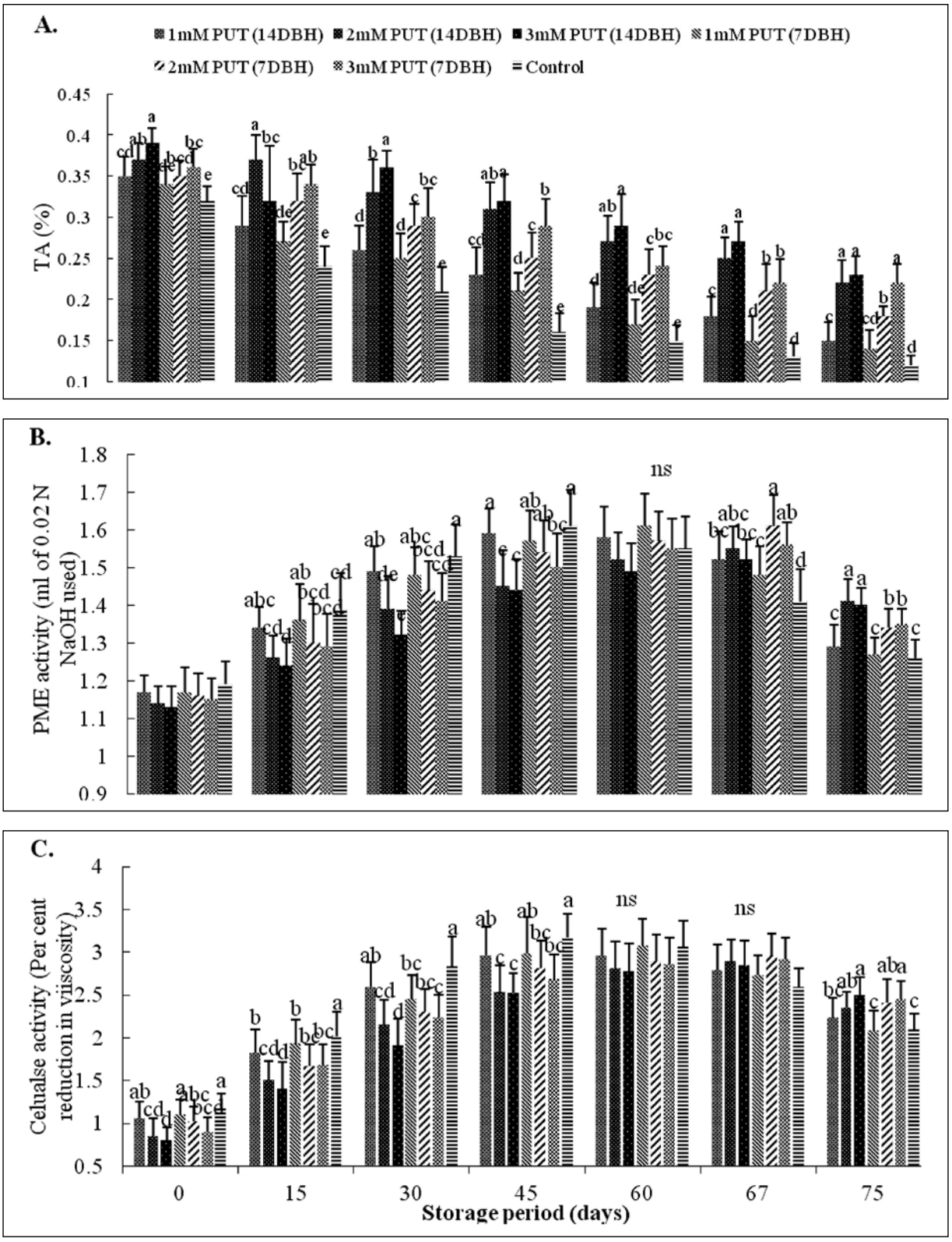

Fig. 2. Variation in SSC (A), PME (B) and cellulase activity (c) of pear fruit during cold storage in relation to different pre-harvest PUT applications. Vertical bars represent ± S.D. of means for 4 replicates. Mean values followed by same letters within a column are not significantly different at *$p \leq 0.05$.

2mM PUT (14DBH) application. However, the lowest TA was recorded in control. Similarly, Razzaq *et al.* (15) reported that TA decreased during storage of mango; however, PUT treated fruit maintain 1.95 fold higher TA compared with control after 28 days of cold storage. Slowing down of fruit respiration and hindering the ethylene production with PUT treatment retains the higher fruit acidity during storage (Valero *et al.*, 17)

Our study showed that fruit colour changed from greenish to yellowish during the storage period (Fig. 1B). At the time of storage, there was no statistical difference in colour value *b** in all the applications. Afterwards, an increment in the colour value *b** was noted in all the treatments with the progression of the storage period. The lowest *b** value was registered in 3mM PUT (14DBH) application throughout the storage study, while highest in the control. Exogenous application of polyamines retards the hydrolytic activities of chloroplast thylakoid membranes, which reduced the loss of chlorophyll content (Popovic, 14) and delayed the changes in peel colour during ripening. Similar results were reported by Hosseini *et al* (7) in pear *cv*. Spadona.

At the time of storage, PME activity did not vary significantly in all the treatments (Fig. 2b). On the 15th day of storage, PME activity was increased; however, lowest PME activity (1.24 ml of 0.02 N NaOH used) was recorded in 3mM PUT (14DBH) application. PME activity was lower in 2mM PUT (14DBH) and 3mM PUT (14DBH) up to 67th day of storage than in control, while it increased up to 45th day in control and afterwards declined. On 60th day of storage, no-significant difference in PME activity was observed. At the end of storage, PME activity reduced in all the treatments; however, 3mM PUT (14DBH) application registered 10% higher PME activity in comparison to the untreated fruit. Similarly, Hosseini *et al.* (8) reported the lowest PME activity in banana fruit treated by foliar application of 2mM PUT. Barman *et al.* (4) reported that polyamines and pectin binding block the access of pectin methyl esterase, pectin esterase and poly-galacturonse and reduce the rate of fruit softening.

An increase in cellulase activity was recorded during the initial period of storage and at the time of storage, 31.62% lower cellulase activity was estimated in 3mM PUT (14DBH) application in comparison to the control (fig 2C). Cellulase activity increased slowly in 2mM PUT (7 & 14 DBH) treatments and 3mM PUT (7 & 14DBH) treatments up to 67th day of storage, while in control it rapidly increased up to 45th day and then declined. On 60th and 67th day of storage, cellulase activity did not vary significantly ($p \leq 0.05$) in all the treatments. At the end of storage, highest cellulase activity (2.50% reduction in viscosity) was recorded in 3mM PUT (14DBH) treatment, while the lowest (2.10% reduction in viscosity) in the control. Polyamine applications affect the firmness as well as soluble solids content during the storage of fruit due to ethylene regulated ripening enzyme like cellulase (Koehler *et al.,* 11). The higher cellulase activity at the end of storage might be due to high cellulose molecule retained in the fruit.

CONCLUSION

It can be concluded that as compared to control pre-harvest treatment of 3mM PUT (14 DBH) was most effective to extend the storage life of 'Punjab Beauty' pear fruit by 7 days under cold storage conditions.

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