DOI : 10.5958/0974-0112.2022.00041.X

Indian J. Hortic. 79(3), September 2022: 305-310

Influence of pre-harvest spray of putrescine on postharvest quality of Indian jujube

Navjot Gupta*, Monika Mahajan and S. K. Jawandha**

Punjab Agricultural University, Regional Research Station, Bathinda - 151001, Punjab, India

ABSTRACT

***Ber* (*Zizyphus mauritiana* Lamk.) is a popular fruit in India because of its higher nutritious value. However, during storage, softening of pulp shortens its shelf-life, which leads to high post-harvest losses. In order to maintain the fruit quality after harvest, an effective technique of pre-harvest putrescine application @ 1, 2 and 3 mM was investigated on *ber* fruits. The sprays were applied at color break stage and the fruit samples were investigated at 0th, 7th, 14th, 21st and 28th day of harvesting for physico-chemical and enzymatic changes. The results suggest that putrescine application @ 3 mM extend the storage life by reducing the weight loss, maintained higher firmness, slowed down the activity of cell wall softening enzymes like pectin methyl esterase and cellulase, and maintained the higher sensory quality of fruits during the cold storage. This spray can be promoted to help the farmers to enhance the marketing period of *ber* fruits.**

Key words: *Zizyphus mauritiana,* putrescine, pre-harvest, storage, enzyme activities.

INTRODUCTION

Indian jujube or *ber* (*Zizyphus mauritiana* Lamk) belongs to family Rhamnaceae is tropical and sub-tropical fruit, that rules over other arid zone fruits (Shobha and Bharti, 13). It is considered as a popular fruit because of its high nutritive value. It is enriched in minerals, vitamins, carotenes, phenolic compounds, flavonoids, anthocyanins and organic acids (Koley *et al*., 6). It is either consumed fresh or can be used to prepare different processed products like jam, jelly, juice, dehydrated products, powders and beverages. Higher moisture content contributes to perishable nature of *ber* fruit that led to its deterioration in a few (2-4) days after harvesting under ambient conditions, thereby results in heavy post-harvest losses. In the cell wall, the degradation of pectin and cellulose caused by fruit softening enzymes like pectin methyl esterases and cellulase is a major factor responsible for ripening of fruits during the storage (Goulao and Oliveira, 2). Hence, modulating cell wall degrading enzymes and delaying the rate of various physio-chemical changes could be helpful in extending the shelf-life and post-harvest quality life of fruits. Polyamines like putrescine, spermine, spermidine etc. are the natural occurring compounds are known to suppress the ethylene production by decreasing 1-amino-cyclopropane-carboxylate (ACC) synthase and oxidase enzyme activities and delay the rate of pulp softening and senescence in fruits (Khan and Singh, 4). Putrescine is one of the predominant polyamines whose role in delaying fruit softening, increased storage life and quality of fruits is known from several studies (Pinero *et al*., 12; Koyuncu *et al*., 7). present study was aimed to investigate the effect of pre-harvest putrescine application in regulating cell wall softening enzymes, fruit firmness and other quality changes in *ber* fruits during low temperature storage.

*Corresponding author: navjotgupta@pau.edu

**Department of Fruit Science, Punjab Agricultural University, Ludhiana 141004, Punjab, India

MATERIALS AND METHODS

The experiment was conducted on 16-year-old *ber* trees (*Z. mauritiana* Lamk.) grown in the horticultural research farm at Regional Research Station, Bathinda during 2019 and 2021. The putrescine @ 1, 2 and 3 mM were sprayed pre-harvest at colour break stage and the control plants were kept untreated (distilled water). Fruits were harvested at optimum maturity and packed in netlon carriers. Packed fruits were kept in corrugated fiber board boxes (CFB) with paper lining having 5% perforations and kept in cold storage at 7-9^{0}C and RH 85-95%. Fruit samples were investigated on 0th, 7th, 14th, 21st, 28th day of storage for various physio-chemical parameters. The physiological loss in weight (PLW) of the fruit was calculated on initial weight basis and expressed in per cent. Fruit firmness was determined by using a penetrometer FT327 (GFFECI, Italy) at each storage interval. Spoilage of *ber* fruits was calculated by manually counting the spoiled fruits at particular storage interval. The TSS, acidity, ascorbic acid content, total sugars, total phenols by AOAC (1) and pectin methyl esterase (PME) and cellulase activity as by

Mahadevan and Sridhar (9) were estimated. The experiment was laid out in Completely Randomized Design with four replicates. The data was pooled and analyzed by one-way analysis of variance (ANOVA) and means 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). The data is presented as mean± standard error. Further, data was subjected to Pearson's correlation analysis to assess the nature and extent of the relationship among various attributes.

RESULTS AND DISCUSSION

An increased fruit weight loss was observed during storage in both treated as well in untreated fruits (Table 1). However, the loss was less in putrescine treated fruits compared to the untreated during storage (p≤0.05). After 28 days of storage, putrecine @ 3 mmol $^{-1}$ and @ 2 mmol $^{-1}$ treated *ber* fruits showed 36 and 32% less weight loss, respectively compared to untreated control. A negative correlation (−0.96, R^2 = 0.91) between PWL and firmness (Tables 2 & 3) was elicited through correlation and linear regression analysis. The physiological weight loss can be reduced by pre-harvest spray of putrescine in pomegranate during storage (Koyuncu *et al*., 7). Fruit firmness substantially declined irrespective of treatments with advancement of storage (Table 1). Rate of decrease in firmness was slower in putrescine treated fruits. At the end of storage, *ber* fruits treated with 2 and 3mM PUT showed 14 and 15% higher firmness compared to untreated control. It has been found from our current study that higher concentration of putrescine was able to maintain better firmness of the fruits during entire storage compared to control and other treatments. It may be due to its role in stabilizing cell walls, and making wall-softening enzymes like polygalacturonase (PG), pectin esterase (PE) less assessable towards it (Valero *et al.,*16). Fruits treated with putrescine @ 1, 2 and 3 mmol L^{-1} showed higher palatability rating up to 28 days of storage and afterward decline was observed, whereas untreated fruits showed palatability rating only up to 21 days of storage, after which decline was observed (Table 1). *Ber* fruits sprayed with putrecine @ 3 mM showed maximum sensory scores among different treatments and the difference was significant (p≤0.05) compared to untreated control. It was observed through analysis that palatability rating has a significant positive correlation (0.95, R^2 = 0.90) with the *ber* fruit firmness (Tables 2 and 3). Hence, in the present the *ber* fruits treated with 2 and 3 mM putrescine were found to be acceptable till 28 days, whereas untreated or control were acceptable till 14th day of cold storage. Our findings are at par with the

Table 1. Variations in physiological weight loss, firmness, palatability rating and spoilage of *ber* fruits under cold storage conditions with respect to different putrescine treatments.

Parameter	Putrescine	Storage Duration (Day)			
		7	14	21	28
PWL (%)	1 mM	1.28±0.05^{b}	2.41±0.07^{a}	2.75±0.05^{a}	4.53±0.07^{b}
	2 mM	1.2±0.05^{b}	2.42±0.06^{a}	2.59±0.05^{b}	4.25±0.07^{c}
	3 mM	1.1±0.06^{b}	2.32±0.06^{b}	2.53±0.05^{b}	4.00±0.06^{c}
	Control	2.1±0.06^{a}	2.46±0.06^{a}	2.74±0.06^{a}	6.25±0.07^{a}
Fruit Firmness (kg cm^{-2})	1 mM	4.25±0.12^{b}	4.0±0.13^{b}	3.0±0.12^{c}	2.0±0.10^{b}
	2 mM	4.4±0.12^{b}	4.15±0.13^{a}	3.3±0.13^{b}	2.9±0.11^{a}
	3 mM	4.7±0.12^{a}	4.35±0.12^{a}	3.65±0.12^{a}	3.1±0.11^{a}
	Control	4.1±0.14^{c}	3.1±0.12^{c}	2.35±0.11^{d}	1.65±0.12^{c}
Palatability rating (1-9)	1 mM	8.82±0.12^{a}	8.65±0.14^{a}	7.35±0.16^{b}	6.4±0.12^{b}
	2 mM	8.9±0.11^{a}	8.8±0.12^{a}	7.6±0.14^{a}	6.8±0.13^{a}
	3 mM	9.03±0.13^{a}	8.95±0.13^{a}	7.85±0.14^{a}	7.1±0.14^{a}
	Control	7.95±0.12^{b}	7.65±0.12^{b}	6.1±0.12^{c}	4.55±0.15^{c}
Spoilage (%)	1 mM	0.00	0.00	1.2±0.05^{b}	1.85±0.05^{b}
	2 mM	0.00	0.00	0.9±0.03^{c}	1.25±0.04^{c}
	3 mM	0.00	0.00	0.00	1.1±0.03^{c}
	Control	0.00	0.00	3±0.08^{a}	8.4±0.15^{a}

Values are mean ± SE (n = 4) and those with same superscript within a column have no significant difference (p≤0.05).

Table 2. Linear regression relationship between various fruit quality attributes.

Combination	Equation	R^2
PWL	Firmness = -1.1887 Firmness+12.02	0.91
PR	Firmness = 0.6694 Firmness-1.7053	0.90
Spoilage	Firmness = -1.2907 Firmness + 11.91	0.85
Total sugars	TSS = 0.879 TSS-2.0327	0.99
Total phenolics	Firmness = 0.0064 Firmness+0.0401	0.79
PME activity	Firmness = 0.0116 Firmness+1.6628	0.002
Cellulase activity	Firmness = -0.1134 Firmness+2.3637	0.146

Table 3. Pearson's correlation coefficient between different fruit quality attributes of *ber* fruits.

Trait	PWL	Firmness	PR	Spoilage	TSS	TA	Total phenols	AA	Total sugars	PME activity	Cellulase activity
PWL	1										
Firmness	-0.96	1									
PR	-0.91	0.95	1								
Spoilage	0.90	-0.98	-0.96	1							
TSS	-0.75	0.81	0.59	-0.76	1						
TA	0.47	-0.32	-0.54	0.31	0.21	1					
Total phenols	-0.97	0.89	0.90	-0.84	0.58	-0.66	1				
AA	-0.91	0.95	0.99	-0.96	0.60	-0.54	0.90	1			
Total sugars	-0.70	0.78	0.55	-0.74	0.99	0.29	0.51	0.56	1		
PME activity	-0.10	-0.01	-0.25	0.05	0.59	0.62	-0.06	-0.24	0.59	1	
Cellulase activity	0.34	-0.38	-0.65	0.46	0.21	0.78	-0.48	-0.64	0.23	0.89	1

results reported by Malik and Singh (10) in mango. The fruit spoilage percentage increased with the progression of storage period. It is clearly exhibited from Table 1 that there was no spoilage in fruits up to 14 days of cold storage. After 21 days of storage there was no spoilage, 0.9 and 1.2% spoilage were noticed in fruits treated with putrescine 3, 2 and 1 mM, respectively whereas 3% spoilage was observed in untreated control. At the end of storage, maximum spoilage (8.40%) was recorded in untreated fruits ($p \leq 0.05$), while the fruits treated with putrescine @ 1, 2 and 3 mM treatments showed the spoilage percentage at the range of 1.85, 1.25 and 1.10%, respectively. A negative correlation (-0.98, $R^2 = 0.97$) was found between the spoilage and fruit firmness (Tables 2 & 3). This suggests that a decrease in fruit firmness has made the fruit more susceptible to spoilage. It could be due to antipathogenic properties of putrescine as reported earlier in straw*ber*ry fruits by Khosroshahi *et al*. (5). The TSS content of *ber* fruits treated with putrescine, increased up to 14 days of storage, after which a decline in TSS was observed by the end of 28 days of storage (Fig. 1A). An increase in TSS with the progression of storage period may be due to the numerous catabolic processes like hydrolysis of starch into sugars in the fruits during ripening and senescence processes (Islam *et al.,* 3). The highest and the lowest TSS of 14.46 and 12.23%, respectively were recorded in untreated fruits after 7 and 28 days of cold storage. The maximum TSS (13.40%) were found in fruits treated with putrescine @ 3 mmol L^{-1}, followed by putrescine @ 2 and 1 mM treatments after 28 days of cold storage. The TSS decline in the putrescine treated fruits, were slower as compared to untreated fruits and the difference was found to be significant ($p \leq 0.05$). The titratable acidity (TA) of the fruits showed a linear declining trend with advancement of storage period (Fig. 1B). A rapid reduction in TA was observed in control fruits as compared to slow reduction in putrescine treated *ber* fruits from 0-28 days of storage at cold temperature. At the end of storage period, the highest TA was recorded in putrescine @ 3 mmol L^{-1} treated fruits (1.55%) and the lowest acidity was recorded in control fruits, *i.e.* 1.25%. This could be because of the decrease in respiration rate in putrescine treated fruits

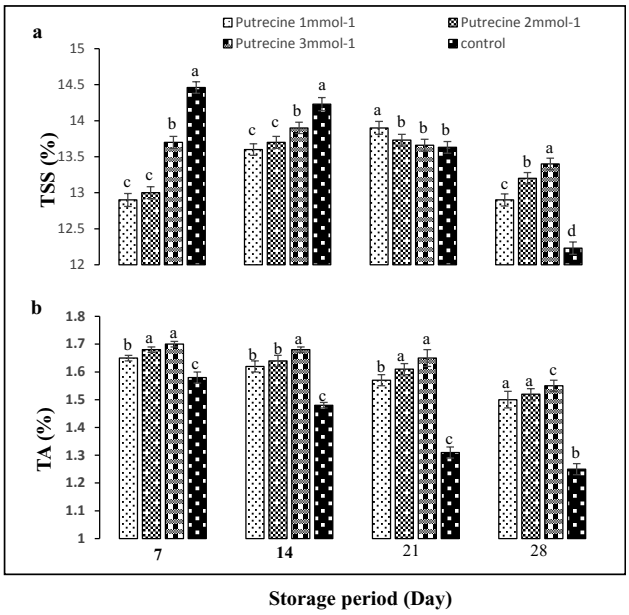

Fig. 1. Total soluble solids, TSS (A), Titratable acidity, TA (B) of *ber* fruits under different treatments. Values are represented as mean ± S.E of 3 replicates. Mean values followed by similar superscript within a column are significantly similar at *p≤0.05.

*Values at zero day: TSS (%): 12.23; TA (%): 1.69

that delays the ripening process. The results were found to be similar to the early reports by Marzouk and Kassem (11) in grapes treated with polyamines. TSS and TA determine the flavour and nutritional status of fruit. After 21 days of storage, the maximum TSS: acid ratio (10.32) was recorded in control fruits as compared to putrescine treated fruits. The results depicted that vitamin C content significantly declined in both treated and control *ber* fruits with increase in storage time. However, this trend was slower in 3 mM putrescine treated fruits (Fig. 2A). A significant (p≤0.05) less reduction of 30, 38 and 39% was observed in 3, 2 and 1 mM PUT treatment compared to 43% reduction in control fruits. It could be because of the inhibitory action of putrescine on the ascorbate peroxidase activity, which ultimately maintains the vitamin C level in the fruits. The total sugars of *ber* fruits showed an increase up to 14 days of storage and thereafter, a significant decline was recorded in both untreated and putrescine @ 1, 2 mmol L^{-1} treated fruits. Whereas, *ber* fruits treated with putrescine @ 3 mmol L^{-1} showed maximum total sugars (10.28%) at 21 days of storage and further decrease thereafter. After 28 days of storage 8.8, 9.74, 9.50% and 9.35% total sugars were observed in control and putrescine @ 3, 2, and 1 mmol L^{-1} treated *ber* fruits, respectively (Fig. 2B). A positive correlation (0.99, R^2 = 0.99) between total sugars and TSS confirms the hypothesis that increased sugars during storage led to increased TSS. The putrescine treatment delayed total sugars losses during storage compared to control, which was significant (p≤0.05), though among treatments there was no significant difference (p<0.05). This delayed sugar losses could be due to the polyamine induced reduced respiration rate as reported by Khan and Singh (4). The total phenolics of both treated and untreated control *ber* fruits showed a decrease with the advancement of storage period (Fig. 2C). This is mainly because most of the polyphenols get oxidized by polyphenol oxidase. however, a decrease was significant ($p < 0.05$) among control and putrescine

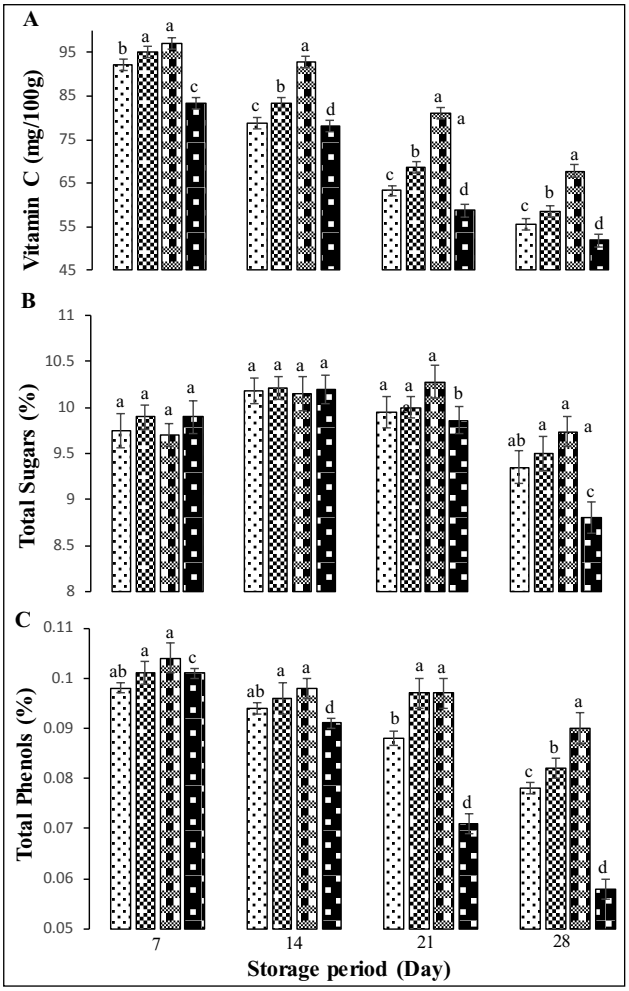

Fig. 2. Vitamin C (A), total sugars (B) and total phenols (C) in *ber* fruits under different treatments. Values are represented as mean ± S.E of 4 replicates. Mean values followed by similar superscript within a column are not significantly different at *p≤0.05.

*Values at zero day: Ascorbic acid (mg/100 g FW): 94.63; total sugars (%): 9.59; total phenols (%) : 0.103.

treated *ber* fruits during storage. About 43, 23, 19 and 11% reduction in total phenolics was found in control and putrescine @ 1, 2 and 3 mmol L^{-1}treated *ber* fruits, respectively. The results showed that putrescine might be responsible for delaying PPO activity due to decreased respiratory activity of *ber* fruits. The role of polyamine in maintaining or increasing the phenolic content in plants was suggested by Kuasno *et al*. (8). A positive correlation between total phenolics and fruit firmness (0.89, R^2 = 0.79) confirms the hypothesis that total phenolics helps in maintaining the firmness by increasing the antioxidant potential of the fruits. The effect of putrescine on maintenance of total phenolics content could be due to its role in delaying the senescence process. Pectin methyl esterase (PME) activity of *ber* fruits increased with progression in storage period irrespective of the treatments (Fig. 3A). After 14 days of storage, maximum PME activity was observed under control, whereas in putrescine treated fruits, it was observed at 21 days of storage and decline afterwards. The results suggested a slow increase in PME activity in putrescine treated *ber* fruits compared to untreated ones. It has been observed that 2 and 3 mM putrescine application retained higher PME activity (8.5 and 11.3%) to that of control. Cellulase activity showed a trend similar to PME activity during 28 days storage (Fig. 3B). Putrescine applications had significantly ($p \leq 0.05$) decrease the cellulase activity of *ber* fruits. Untreated *ber* fruits showed maximum cellulase activity at 14 days, whereas putrescine treated *ber* fruits showed maximum cellulase activity at 21 days of storage followed by decline in all (Fig. 3B). Storage retained 8.59 and 11.36% higher cellulase activity in 2 and 3 mM putrescine treated *ber* fruits compared to untreated ones. Similar trend for PME and cellulase activity was also recorded by Sinha *et al.* (15) in plum fruits. A negative correlation (−0.01, R^2 = 0.002) and (−0.38, R^2 = 0.146) was found between PME activity vs. fruit firmness and cellulase activity vs. fruit firmness, respectively (Tables 2 and 3). The results showed that putrescine treatments @ 2 and 3 mM significantly maintained PME and cellulase activities in *ber* fruits that contributed to its higher firmness (Fig. 3A-B). Polyamines delay the fruit softening by cross linking to negative carboxyl group in the cell wall and reduced PME activity during the storage. In this study, reduced PME activity was found in the *ber* fruits on 14th day of treatment with 2 and 3 mM putrescine compared to untreated fruits. Similar results were reported in pear fruit where putrescine showed inhibitory action on PME activity, thereby maintains the firmness of the fruits (Singh *et al*., 14). Polyamines maintain the fruit firmness and total soluble solid content by regulating the cellulase activity in the cell wall. Therefore, low cellulase activity in putrescine treated *ber* fruits during the storage supported this assumption, thus enhance the shelf life of fruits. The activity of cell wall degrading enzymes like PME and cellulase was reduced by putrescine, thereby slowed the process of fruit softening in *ber* fruits. Therefore, the pre-harvest putrescine spray can be useful tool to extend the shelf-life and enhance the marketing period of *ber* fruits during cold storage conditions (7.5±1°C; RH = 90-95%).

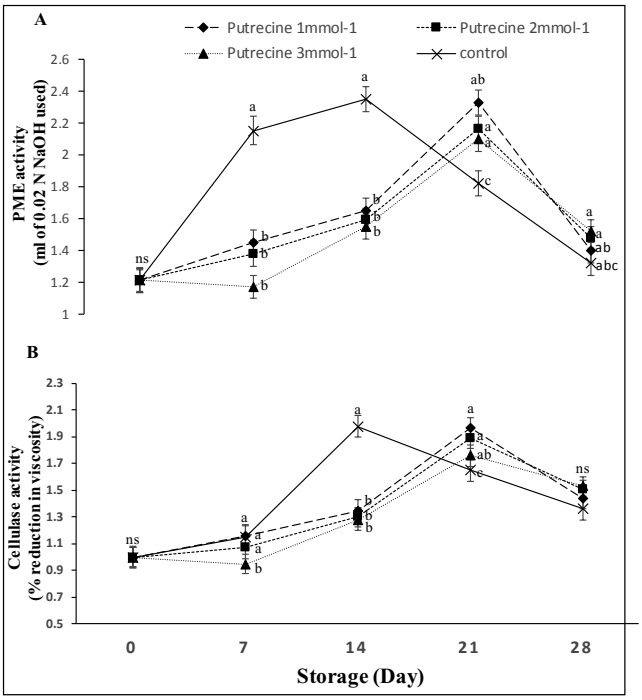

Fig. 3. Pectin methyl esterase (PME) (A) and cellulase (B) in *ber* fruit under different treatments. Vertical bars show mean ± S.E for 3 replicates. Mean values followed by different superscript within a column show that values are significantly different at *p≤0.05.

AUTHORS' CONTRIBUTION

Conceived, designed and performed the experiments (NG); Analyzed and interpreted the data and wrote the paper (MM); Conceived and designed the experiments; contributed reagents and materials (SKJ).

DECLARATION

The authors have declared no conflicts of interest for this article.

ACKNOWLEDGEMENT

The authors would like to thank Department of Fruit Science, Punjab Agricultural University and

Regional Research Station, Bathinda for all the facilities provided to do the research work.

REFERENCES

1. A.O.A.C. 2000. *Official and Tentative Methods of Analysis*, Association of Official Agric Ahemists, Washington, DC, USA.

2. Goulao, L.F. and Oliveira, C.M. 2008. Cell wall modification during fruit ripening: when a fruit is not the fruit. *Trends Food Sci. Technol.***19**: 4-25.

3. Islam, M.K., Khan, M.Z.H., Sarkar, M.A.R., Absar, N. and Sarkar, S.K. 2013. Changes in acidity, TSS and sugar content at different storage periods of the postharvest mango (*Mangifera indica* L.) influenced by Bavistin DF. *Int. J. Food Sci.* **13**: 1-8.

4. Khan, A.S. and Singh, Z. 2010. Pre-harvest application of putrescine influences Japanese plum fruit ripening and quality. *Food Sci. Technol. Int.* **16**: 53-64.

5. Khosroshahi, M.R.Z., Esna-Ashari, M. and Ershadi, A. 2007. Effect of exogenous putrescine on post-harvest life of straw*ber*ry (*Fragaria ananassa* Duch.) fruit cultivar Selva. *Sci. Hortic.* **114**: 27-32.

6. Koley, T.K., Kaur, C., Nagal, S., Walia, S., Jaggi, S. and Sarika. 2011. Antioxidant activity and phenolic content in genotypes of Indian jujube (*Zizyphus mauritiana* Lamk.). *Arabian J. Chem.* **9**: 1044-52

7. Koyuncu, M.A., Erbas, D., Onursal, C.E., Secmen, T., Guneyli, A. and Sevinc Uzumcu, S. 2019. Postharvest treatments of salicylic acid, oxalic acid and putrescine influences bioactive compounds and quality of pomegranate during controlled atmosphere storage. *J. Food Sci. Technol.* **56**: 350-59.

8. Kusano, T., Yamaguchi, K., *Berber*ich, T. and Takahashi, Y. 2007. Advances in polyamine research. *J. Plant Res.* **120**: 345-50.

9. Mahadevan, A., and Sridhar, R. 1982. *Methods on Physiological Plant Pathology*, Madras Sivagami Pub.

10. Malik, A.U. and Singh, Z. 2005. Pre-storage application of polyamines improves shelf-life and fruit quality of mango. *J. Hortic. Sci. Biotechnol.* **80**: 363-69.

11. Marzouk, H.A. and Kassem, H.A. 2011. Improving yield, quality and shelf life of Thompson seedless grapevine by preharvest foliar applications. *Scientia Hortic.* **130**: 425-30.

12. Pinero, M.C., Otalora, G., Collado, J., Lopez-Marin, J., Del Amor, F.M. 2021. Foliar application of putrescine before a short-term heat stress improves the quality of melon fruits (*Cucumis melo* L.). *J. Sci. Food Agric.* **101**: 1428-35.

13. Shobha, D. and Bharati, P. 2007.Value addition to *ber* (*Zyziphus mauritiana* Lamk.) through preparation of pickle. *Karnataka J. Agric. Sci.* **20**: 353-55.

14. Singh, V., Jawandha, S.K., Gill, P.P.S. and Dhillon, W.S. 2019.Preharvest applications of putrescine influences the storage life and quality of pear fruit. *Indian J. Hortic.* **76** : 486-92

15. Sinha, A., Jawandha, S.K., Gill, P.P.S. and Singh H. 2019.Enhancement of storage life and quality maintenance of plum fruits. *Indian J. Hort.* **76**: 493-501

16. Valero, D. and Serrano, M. 2010. Postharvest biology and technology for preserving fruit quality., CRC Press pp. 288.

Received : May, 2022; Revised : September, 2022;
Accepted : September, 2022