Annals of Plant and Soil Research 26(1):105-115 (2024)
https://doi.org/10.47815/apsr.2024.10339

Metabolic alterations in pear cultivars during storage at ambient conditions

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Received: November: 2023; Revised accepted: January: 2024

ABSTRACT

'Patharnakh' (PN) (Pyrus pyrifolia Burm. Nakai) and 'Punjab Beauty' (PB) [Pyrus communis L. × Pyrus pyrifolia Burm. Nakai] are the leading low-chill pear cultivars of the subtropics of India. Diurnal temperature and relative humidity during the fruit harvest period are high, which considerably affects the shelf life of fruits. Fruits of 'PN' and 'PB' pear harvested at physiological maturity were stored for 16 days at ambient temperature, and the effects of the storage day interval on physical and qualitative parameters were studied. Both cultivars showed reductions in fruit firmness, physiological weight loss, total soluble solids, and juice acid content during storage. Activities of fruit softening enzymes such as polygalacturonase (PG), pectin methylesterase (PME), and cellulase were enhanced, whereas those of superoxide dismutase (SOD) were reduced during storage. Fruit firmness was negatively correlated with polygalacturonase, pectin methylesterase, PPO, and cellulase enzymes in both cultivars. In both cultivars, the increased browning of the fruit during storage was negatively correlated with total phenolic content and positively correlated with polyphenol oxidase activity during storage. 'Patharnakh' and 'Punjab Beauty' fruits maintain desirable quality parameters up to 6–12 days and 4–8 days, respectively, during storage at ambient conditions.

Keywords: storage, polyphenol oxidase, fruits, firmness, acidity

INTRODUCTION

Pears have great importance in the global market for pome fruits. Pear ranks second next to the apple fruit crop in the world in terms of area, production, and productivity among temperate fruits. In India, it is grown in Uttarakhand, Himachal Pradesh, Jammu & Kashmir, Punjab, and some areas of Assam and Nilgiris hills. The emergence of new pear varieties that can be developed into a commercial crop with novel and interesting traits is a great opportunity for the improvement of the fruit market. In Punjab, pear cultivation is dominated by the low-chill hard pear cultivar 'Patharnakh' belonging to the Oriental pear group (*Pyrus pyrifolia* Burm. Nakai) and the semi-soft pear cultivar 'Punjab Beauty', a hybrid between *Pyrus communis* L. and P*yrus pyrifolia* Burm. Nakai, and fruits are harvested at physiological maturity in the half month of July. The pear harvest coincides with the hot, humid season, resulting in a rapid deterioration of the fruit's shelf life. It is documented that pear fruits retain their post-harvest shelf life for up to 10 days at room temperature (25–30 °C) after 10–15 days. Quality-related parameters get reduced rapidly. After harvest, its marketability depends on its delicate flesh, peel texture, rich juice, good taste, and excellent aroma. The changeability in physico-chemical and sensory parameters and cell wall-degrading enzyme activity can be used to understand the ripening behaviour of pear cultivars. Fruit quality decreased after harvest due to rapid changes in respiration, the activity of cell wall degrading enzymes, and the infestation of pathogens during transportation and storage.

Loss of firmness is the most important characteristic indicating the deterioration of pear fruits; these changes directly influence the quality of the fruits as well as their storage life, transportability, and marketing. The quantification of organic acids and soluble sugars is associated with the production of quality fruits. Sugar content in pear fruit improves during the early storage period and further declines with the advancement of storage at room temperature due to fermentation into alcoholic content (Kaur and Dhillon 2015). Fruit softening is generally observed during the ripening of various types of fruits (Murayama *et al.,* 2006). Loss of turgor pressure and degradation of the cell wall in climacteric fruits, such as pears, contribute to the decrease in firmness and fruit quality (Khin *et al.,* 2007, Zhou *et al.*, 2008). It has been seen that pectin dissolves when things soften, which is caused by

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the action of enzymes polygalacturonase (PGU) and cellulase (Payasi *et al.,* 2009). Reports of changes in cell wall pectic enzymes in guava (Abu-Bakr *et al.,* 2003) and pear (Zhou *et al.,* 2011) supported the theory. However, not much is known about how these enzymes change the cell membrane and cell wall or how important related fruit-softening enzymes are in hard and semi-soft pear cultivars. So, the purpose of this exploratory study was to look at how the activity of the cell wall-degrading enzymes polygalacturonase, pectin methylestrase and cellulase changes as pear fruits are stored at room temperature and how these changes affect fruit ripening.

MATERIALS AND METHODS

The experiments were performed over two consecutive years, 2019 and 2020, during the summer-rainy periods. There were two types of pears grown each year: Patharnakh and Punjab Beauty. They were picked at full maturity (145 ± 5 g, 70 ± 2.5 N firmness, and 12 ± 0.25 °Brix soluble solid content) in the last week of July from the Fruit Research Farm of Punjab Agricultural University in Ludhiana (30.90 °N, 75.86 °E), Punjab, India. The fruits were selected on the basis of their uniform size, colour, and absence of bruises and diseases. The fruits were randomly picked in the morning during the two years of study. Plastic crates were used for collecting the fruits and were immediately transferred to the post-harvest laboratory within 2 hours. The fruits were washed with a sodium hypochlorite solution (2.5 mL) and allowed to dry at room temperature (32 ± 2°C). For storage studies, 80 uniform fruits were grouped randomly into 4 sets of 20 fruits for each replication under each cultivar. After thoroughly washing, fruits were air-dried, packed in the corrugated fibreboard boxes (CFBs), and stored at 0-1°C and 90–95% RH. The fruits were analysed at 0, 4, 8, 12, and 16 days of cold storage. On the day of harvesting, fresh fruit quality was also analysed.

The fruits of two cultivars (five in number) were randomly selected, weighed, and then placed in individual 1.45-L glass jars. A tiny hole was made through the lids of the respective jars, and high-strength adhesive tape was used to cover the hole in the lid. Afterward, these jars containing pears were maintained at 10°C. A gas analyser (Systech Instruments: Model GS3/P, UK) was used to determine the respiration rate of the pears by analysing the gas and CO_2 concentration through the hole in the lid of every jar. This measurement was repeated 4 times for each storage interval. The respiration rates of both Patharnakh and Punjab Beauty pears were expressed in terms of mg CO_2 per kilogram per hour. The respiration rate was calculated using the following formula that considers CO_2 emission and fruit mass:

$$R \text{ (mg CO /kg/hr)} = \frac{\Delta \text{ carbon dioxide}}{1000} \; \frac{\text{Fresh mass (g)}^{*} \; \Delta t}{60}$$

Fruit firmness was determined by using a penetrometer (model no FT-327, USA). Two readings were taken from the opposite sides of each fruit after the peel was removed. The values were expressed as Newton (N) force.

PPO was assayed according to the method of Zauberman *et al.* (1991) with little modification. Flesh from five fruits (0.2 g) was mixed together in 2 ml of 0.02 M phosphate buffer (pH 6.8) that contained polyvinyl pyrrolidone that did not dissolve. The mixture was mixed together and spun at 10,000 × g for 30 minutes at 4°C. The clear liquid that came out was used to get enzyme extract, and 4-methyl catechol was used as a substrate. A cuvette was filled with 0.5 ml of enzyme extract, 0.1 ml of phosphate buffer (pH 6.8), and 0.1 ml of 4-methyl catechol. This was done to measure the activity of PPO. Then, the increase in absorbance at 410 nm was recorded for 3 minutes in a spectrophotometer. One unit of enzyme activity was defined as the amount that increases the absorbance per minute by 0.01 and was expressed as units per minute per gram of fresh weight (FW). For enzyme extraction, fruit tissue (0.1 g) was homogenate in a prechilled pestle and mortar, followed by the addition of 2 ml of cold 0.1 M potassium phosphate buffer [1 mM ethylenediaminetetraacetic acid (EDTA), 1% polyvinyl pyrrolidone, and 10 mM β mercaptoethanol] at 7.5 pH. The homogenate was centrifuged at 10,000 × g at 4°C for 30 min. The supernatant was used for the enzyme assay method of Marklund and Marklund (2005). In a spectrophotometric cuvette, 0.1 M Tris-HCl buffer at pH 8.2 (1.4 ml), 6 mM EDTA (0.5 ml), 6 mM pyrogallol (1 ml), and enzyme extract (0.1 ml) were added, and the mixture was measured using a spectrophotometer. The change in absorbance was recorded at 420 nm for up to 3 minutes at an interval of 1 minute. Superoxide dismutase activity was expressed as units per minute per gram of FW. One unit (U) of SOD activity was defined as the amount of enzyme that causes 50% inhibition of pyrogallol and was expressed as units per minute per gram of FW. The Folin-Ciocalteu reagent method and a spectrophotometer (Thermo Scientific SPECTRONIC 20 D+, USA) set to 760 nm absorbance were used to find out the total phenolic content (TPC) of fruits. The results were calculated using a gallic acid standard curve and expressed as micrograms of gallic acid equivalent per kg of fresh fruit weight. 25 To find out how much AsA was in the fruit pulp (10 g), a metaphosphoric acid solution was added and the mixture was titrated against the dye (2,6-dichlorophenol-indophenol) until the pink colour showed up. Ranganna (2000) described a method for recording and calculating the titre value, which she expressed as milligrams per kilogram of fruit.

For the extraction of enzymes, pre-chilled pear pulps (0.01 kg for each treatment) were grounded in a pestle and mortar and homogenized at 4°C for 30 min. Tris-HCl buffer at pH 8.0 (10 mL) at a rate of 0.05 mol L-1 consisting of 0.001 mol L-1 ethylene diamine tetraacetic acid (EDTA), 5% insoluble polyvinyl pyrrolidone, and 2 M NaCl (1 mL) were used for the extraction of pectin methylesterase. Phosphate buffers (0.01 mol L-1) at pH 7.0 (10 mL) consisting of 1 mM EDTA and 5% insoluble polyvinyl pyrrolidone (w/v) were used for the extraction of polygalacturonase, and 0.05 mol L-1 NaCl was used for the extraction of cellulase activity (Lohani, Trivedi, & Nath, 2004). The homogenate was then subjected to centrifugation (10,000× g) for 20 min at 4°C. Afterwards, the enzyme activity was determined by the lucid supernatant. By the method of Abu-Goukh and Bashir (2003), pectin methylesterase (PME) activity was determined, which was expressed as mmol kg-1 min-1 methyl ester. By the method described by Lohani *et al.* (2004), polygalacturonase activity was determined, which was expressed as mmol kg-1 min-1 D-galactose. By measuring the reducing groups released from carboxymethyl cellulose, cellulase activity was determined and expressed as mmol kg-1 min-1 D-galacturonic acid. (Chin *et al.,*1999). The calculated mean data for the 2020-21 years were analysed with two-way analysis of variance using SAS statistical analysis software 9.3 (Institute Inc., Cary, NC, USA) at ($p \leq 0.05$). Correlation analysis was performed with XL Stat-Pro 7.5.3 to determine the effect of storage days on the analysed parameters of pear.

RESULTS AND DISCUSSION

Firmness & Weight loss %

The effect of storage on the firmness of two pear cultivars is presented in Fig. 1a. The softening of fruit during storage and transportation is a limitation that compromises the quality and commercialization of fruits. For this reason, the firmness of Patharnakh and Punjab Beauty pear cultivars showed varying degrees of reduction over time. By day 16, the rate of softening of Patharnakh fruits was lower than that of Punjab Beauty, and values were higher between 8 and 16 days in both cultivars. During storage, firmness reduced from 76 N to a range of 41 N in Patharnakh and 68 N to 30 N in the Punjab Beauty cultivar. The gradual decrease in fruit firmness is due to the frangible structure of the cell wall, degradation of starch, and disintegration of the cell membrane (Adhikary *et al.,* 2022). Nevertheless, the fruits of the Patharnkah cultivar displayed significantly higher mean firmness in contrast to the Punjab Beauty at different intervals of storage. At 0-day storage, fruits exhibited a maximum firmness of 76.14 N in Patharnakh, which is up to 10% higher than the fruits of Punjab. Fruit firmness is considered an important index of the texture and storage life of pears. The destructive effect of storage on fruit firmness may be due to the increased activities of cell wall-degrading enzymes, notably cellulose, pectin methylestrase, and polygalacturonase (Maftoonazad and Ramaswamy 2005).

Quantitative losses were particularly excessive after 8 days until the end of storage, thus rendering the fruit commercially unacceptable (Fig. 1b). As shown in Fig 1b, the physiological loss in weight of Patharnakh and Punjab Beauty pear cultivars increased during different storage intervals, and a maximum rate of up to 3.87 to 8.01 % was observed in Punjab Beauty between 4 and 12 days, compared to 2.81 to 5.54 % in Patharnakh at ambient storage conditions. Irrespective of 0 days of storage, the weight loss in fruits steadily increased in both cultivars up to 16 days of storage. Storage interval days reduce weight loss in fruits and retain freshness up to 0–2 days of storage as compared to 4–16 days of storage. The maximum economic weight loss was exhibited in Punjab Beauty at 10.77% at the 16th day of storage, indicating low acceptability in the market at this stage. Up to the 8th day of storage, the fruits of the Patharnakh cultivar demonstrated the lowest weight loss, although from the 12th day onwards, the weight loss increased till the end of storage. On the 16th day, both cultivars exhibited maximum weight loss.

Physiological loss consists of metabolic activities, respiration, transpiration, and the water pressure gradient between fruit tissues, environment, stage of ripening, and storage temperature (Hafez *et al.,* 2019). It acts as a detrimental factor to aggravate the fruit freshness, which might be associated with a loss of moisture from the tissue (Barman *et al.,* 2014).

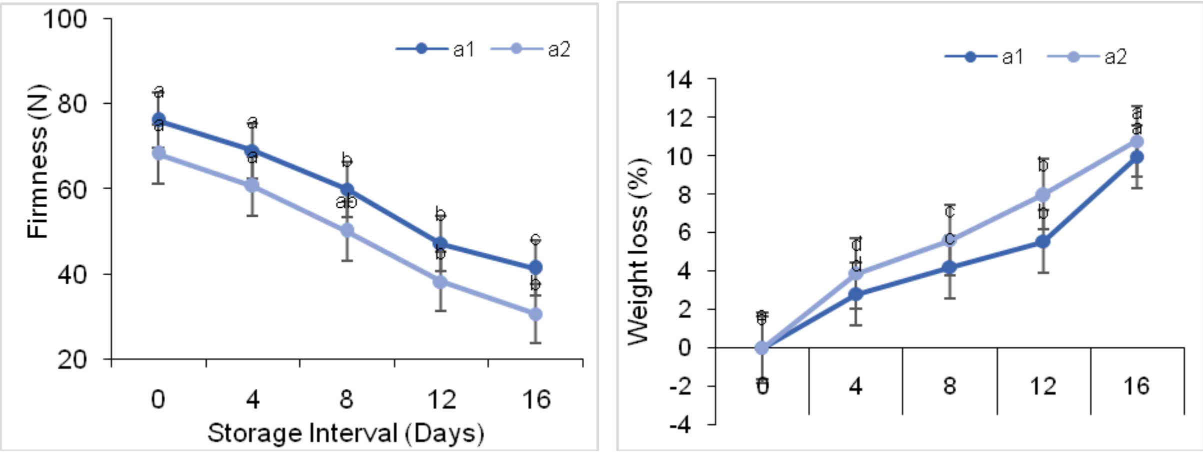

Fig. 1: Changes in fruit firmness (A), physiological loss in weight (B) of pear fruits during storage at ambient conditions. Vertical bars represent ± SE of means for 4 replicates. Different letters indicate the significant differences among storage periods (P≤0.05) and a1-Patharnakh, a2-Punjab Beauty

Browning Index and respiration rate

The browning index varied among both cultivars of pear during the days of storage (Fig. 2a) and was significantly lower in the Patharnakh cultivar. The initial storage days showed a delay in browning incidence of fruits as compared to the 8 to 16 days of storage in both cultivars. This might be attributed to the inhibitory role of storage days in modifying the internal atmosphere of fruits. Nevertheless, the incidence of internal browning was maximum at the 16th day in Punjab Beauty, and it was 36.82% more than the Patharnakh cultivars. The maximum increase was exhibited from 8–16 days in both cultivars, whereas at 0–4 days, no browning incidence was depicted in both cultivars. The gaseous gradients in the brown tissues of pear pulp may be attributed to an imbalance between an oxidative and reductive reaction, which leads to the accumulation of reactive oxygen species (Tripathi and Oelmuller 2012). It may induce a loss of membrane integrity that is visible macroscopically. The enzymatic oxidation, specifically due to the PPO activity in phenolic compounds, causes the formation of brown-colored polymers, which increase steadily during storage (Fig. 2a). This disorder evident in the postharvest storage of pears may be due to an alternation in the internal gaseous atmosphere of the fruit (Saquet *et al.,* 2003).

During storage at ambient conditions (Fig. 2b), the respiration rate of the fruits is lowered proportionally for both cultivars. From the 4th day of storage, fruits of Punjab Beauty showed a 47% increment in respiratory activity until the last day of storage, while in Patharnakh pears, there was an approximate 62% increase in respiration rate from the 4th day of storage until the 16th day of storage. The maximum increase in respiration rate was depicted in Punjab Beauty, and it was 15% more than the fruits of Patharnakh. The findings of Makino (2013) showed that the living tissues of fruits undergo continuous change, even after harvesting. This means that metabolic reactions are active in cells, and the rate of product deterioration is generally proportional to the respiration rate. As the respiration rate increased, the shelf life of fruits deteriorated drastically.

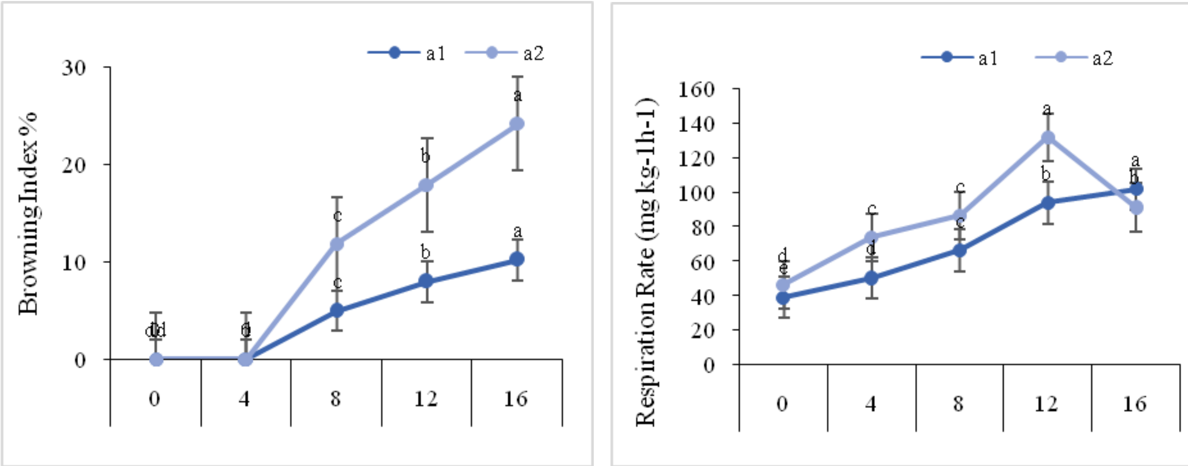

Fig. 2: Changes in Browning Index % (A) and Respiration rate (B) in pear fruits during storage at ambient conditions. Vertical bars represent ± SE of means for 4 replicates. Different letters indicate the significant differences among storage periods (P≤0.05) and a1-Patharnakh, a2-Punjab Beauty

TSS, pH, and TA, AsA

TSS content increased up to 4 DAS in Patharnakh and 8 DAS in Punjab Beauty and then declined during the advanced storage period (Fig. 3a). The effect of storage days on the TSS content of pears is presented in Fig. 3a. In the Punjab Beauty cultivar, the TSS content increased with storage and reached its maximum level on the 8th day, then declined on the 12th day of the storage period. However, in Patharnakh cultivar fruits, the TSS content was reduced after the 4th day of storage. The rise in TSS content during the initial period of storage advised that the ripening process was still in continuation or respiration rate had been enhanced in stored ones, and eventually, after a particular stage, TSS declined due to the lack of available substrate. Of course, changes in TSS from the 8th day onwards in Punjab Beauty and the 4th day onwards in Patharnakh may be due to the higher rate of respiration during storage (Porat *et al.,* 2005). The conversion of sugars to ethanol under stored conditions was due to the fermentation of overripe fruits and resulted in an adverse effect on the fruit quality. The decreasing drift of TSS during the storage of fruits is possibly due to the decline in carbohydrates and pectin content. Besides, partial hydrolysis of proteins as well as decomposition of glycosides into small units in the respiration process may also be the reason (Latorres *et al.,* 2018). At the end of the storage period, the lower TSS values in both cultivars may be attributable to a higher respiration rate.

Juice pH in fruit consistency increased with the progression of storage duration and reached its maximum value on the 16th day of storage (Fig. 3b). In the Punjab Beauty cultivar, the increase in juice pH was at a higher rate from the 4th day onwards, and this rate of increase was at its minimum at the 0th day of storage. However, in Patharnakh fruits, the juice pH increased up to the 8th day of storage; afterwards, a decline was noticed, and this may be due to alcohol and aldehyde formation within the fruit tissues. The increase in juice pH over storage time is attributed to the degradation of organic acid, which is consumed as a respiratory substrate. Our present results are in close conformity with the findings of Wani *et al.* (2014), who attributed higher fruit pH with storage to the reduction of total acidity.

The acidity % in pears declined with the advancement of storage (Fig. 3c). Throughout the storage studies, fruits of the Punjab Beauty cultivar showed ~56% loss in TA, while fruit of Patharnakh showed an approximate loss of 51% TA from 0 DAS until the end day of storage. The reduction in TA during the storage of fruit points out the transformation of organic acids into sugars during the respiration process (Sharma & Rao 2015).

In our study, AsA content measured at the 16th day of storage was approximately 2 times lower than at the 0th day of storage in both cultivars (Fig. 3d). As an antioxidant, AsA might be involved in oxidative reduction reactions within the fruit's internal atmosphere, which was modified by the storage. Fruits of the Patharnakh cultivar displayed a comparatively slower rate of ascorbic acid reduction during progressive storage days, and it was 9% lower as compared to the Punjab Beauty cultivar. The results of Nasrin *et al.* (2020) also revealed that higher ascorbic acid levels in lemons at harvesting time were reduced continually with the progress of storage duration.

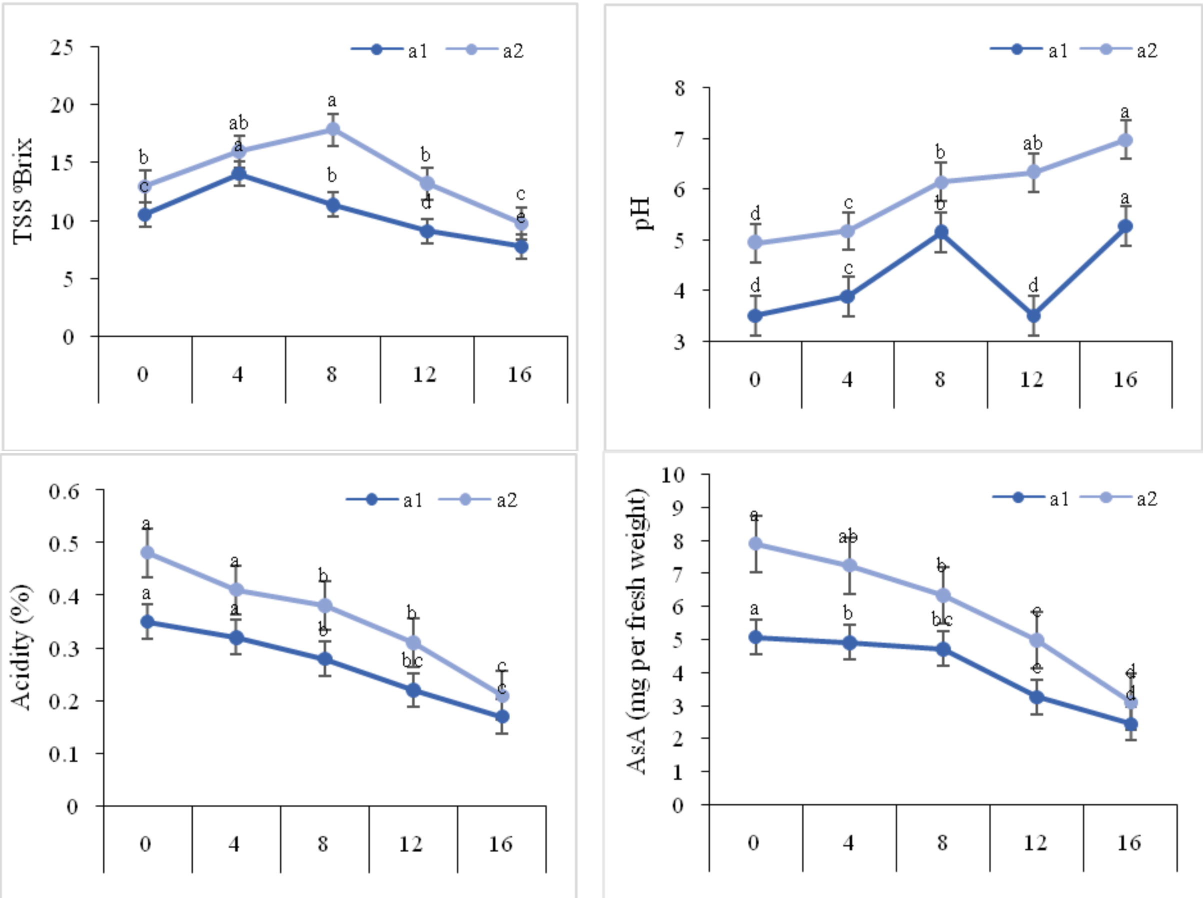

Fig. 3: Changes in Total Soluble Solids (A), pH (B), Acidity % (C) and Ascorbic acid (D) of pear fruits during storage at ambient conditions. Vertical bars represent ± SE of means for 4 replicates. Different letters indicate the significant differences among storage periods (P≤0.05) and a1-Patharnakh, a2-Punjab Beauty

Total phenolic content and Polyphenol oxidase enzyme

The change in TPC and PPO for both pear cultivars during storage is illustrated in Fig. 4a and 4b. On the initial day of storage, the TPC in fruit was at its maximum, while the PPO activity was at its minimum. As the storage period progressed, TPC declined with a concomitant increase in PPO activity until the 16th day of storage. During the study period, there was a 54% and 59% reduction in Punjab Beauty and Patharnakh, respectively. However, the PPO activity on all the storage days increased, peaked, and then decreased on the last day of storage. The maximum activity of PPO was observed in the Punjab Beauty cultivar at the 12th day of storage, and it was approximately 4% more as compared to the Patharnakh cultivar. The decline in TPC of pears during storage in our studies is in conformity with the findings of Shen *et al.* (2013) in citrus fruits. A decrease in total phenolic contents during storage is directly proportional to the oxidation of the PPO enzyme, where phenol is converted into quinone compounds (Wong and Leong 2005). In the pear fruit, the enzymatic browning leads to deterioration in quality.

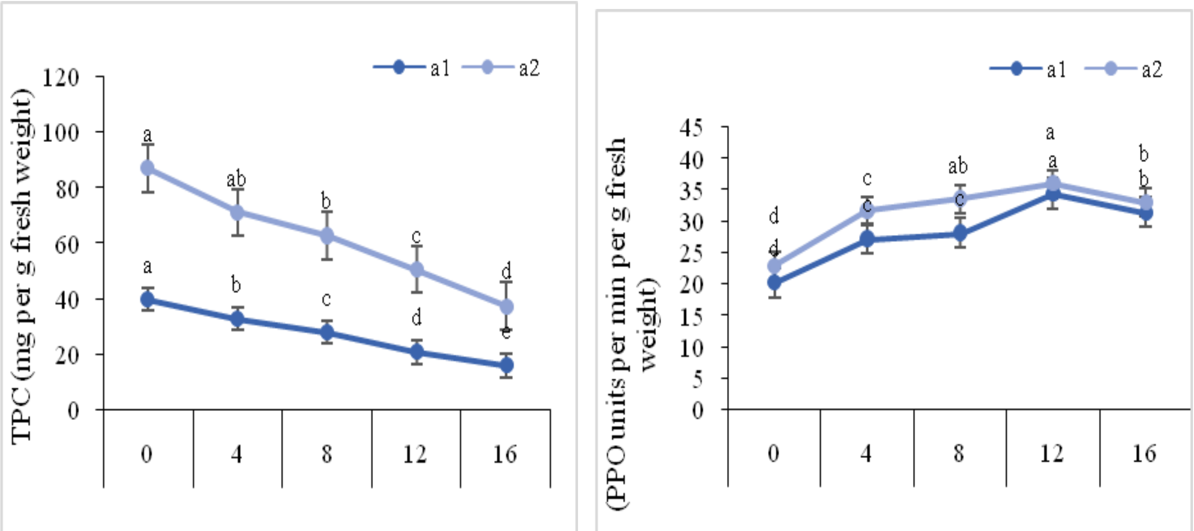

Fig. 4: Changes in Total Phenolic Content (A), Polyphenol oxidase (B) of pear fruits during storage at ambient conditions. Vertical bars represent ± SE of means for 4 replicates. Different letters indicate the significant differences among storage periods ($P \leq 0.05$) and a1-Patharnakh, a2-Punjab Beauty

Pectin methyl esterase, Polygalacturonase, Cellulase and Superoxide dismutase activity

The softening enzyme activities, namely pectin methyl esterase, polygalacturonase, and cellulase, exhibited a comparative trend depending upon the storage interval (Fig. 5 (a-c). The enzymatic activity in pears increased steadily with the expansion of the storage period up to the 12th day. However, as compared to Punjab Beauty, the fruits of the Patharnakh cultivar showed slowed activity of fruit softening enzymes during storage. Fig. 4a illustrates that pectin methyl esterase (PME) activity was at its peak in the Punjab Beauty cultivar on the 8th day of storage; afterwards, it decreased till the end day of storage. Similarly, in the Patharnakh cultivar, the activity of pectin methyl esterase was at its peak on the 8th day of storage, but it showed a lesser increase than in the Punjab Beauty cultivar.

Likewise, the polygalacturonase activity in both cultivar fruits illustrated an increasing trend over time, but the highest polygalacturonase activity was observed in Punjab Beauty on the 12th day of storage (Fig. 5b). Interestingly, the activity of polygalacturonase in Punjab Beauty and Patharnakh cultivars escalated up to the 12th day of storage and subsequently decreased. Both cultivars maintained lower cellulase activity (Fig. 5c) up to the 4th day of storage; afterwards, a sharp increment in cellulase activity was noticed on the 12th day of storage and subsequently decreased at the end of storage. After the 12th day of storage, the activity decreased due to the consumption of cell substrate, but the enzyme activity was much higher in fruits stored up to the 12th day than the 16th day because of the faster rate of substrate consumption. Likewise, the activity of pectin methyl esterase (PME) was inhibited in fruit at 0 and 4 days of storage, and afterwards it increased until 12 days of storage. Pectin dimethyl esterification, catalysed by pectin methyl esterase, not only acts as a substrate for polygalacturonase (PG) but also changes the pH level as well as the cation exchange mechanism of the cell wall, leading to fruit softening (Micheli 2001). In the present study, all the softening enzymes increased steadily over time, although the activity of cellulase and Polygalacturonase was quite lower in Patharnakh than Punjab Beauty. Softening of fruit tissues is influenced by the hydrolysis process of peptic substances and by varied cell wall deteriorating enzyme activity (Payasi *et al.,* 2009). Mechanistically, disassembling of the cell wall and changes in the pectic substances cause fruit softening during the ripening process (Rodriguez-Marin *et al.,* 2002). The results of this study supported the assumption that the higher levels of softening enzyme activities in pear during storage favoured the retention of firmness of the fruit flesh.

Superoxide dismutase **(**SOD) activity gradually decreased with storage, but it exhibited maximum activity in Punjab Beauty as compared to those recorded in the fruits of Patharnakh

(Fig. 5d). However, the superoxide dismutase (SOD) activity of both cultivars decreased after the 4th day of storage. On the 16th day of storage, the fruits of Punjab Beauty showed 63% higher SOD activity than the fruits of Patharnakh. Antioxidant enzymes play an important role in suppressing oxidative stress. An antioxidant such as SOD protects cells against oxidative damage by scavenging ROS. In this study, fruits at 0 and 4 days of storage retained higher SOD activity during storage as compared to the rest of the storage. In this study, stored conditions encourage the oxidative stress caused by ROS overproduction in pears and also enhance the peroxidation of membrane lipids, postponing loss of membrane function and alleviating the oxidative stress of postharvest pears. Low levels of SOD activity contributed to the development of major postharvest disorders in pear, as observed by Saba and Moradi (2016).

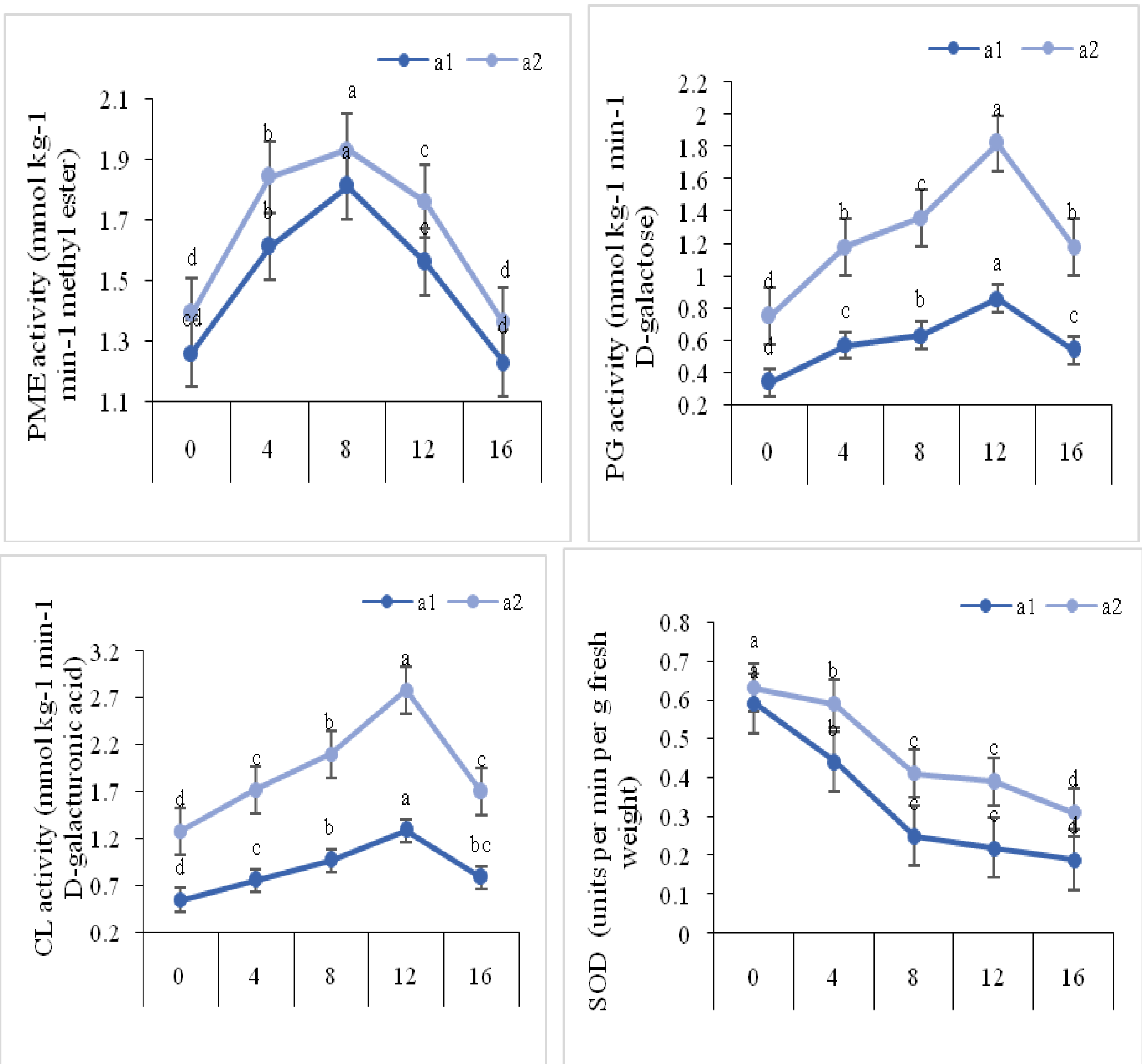

Fig. 5: Changes in activities of enzymes as Pectin methyl estrase (A), Polygalacturonase (B), Cellulase (C) and Superoxide dismutase (D) in pear fruits during storage at ambient conditions. Vertical bars represent ± SE of means for 4 replicates. Different letters indicate the significant differences among storage periods (P≤0.05) and a1-Patharnakh, a2-Punjab Beauty

Table 1: Pearson correlation analysis of different physio-biochemical attributes of Patharnakh

Variables	TPC	FM	RR	PME	CL	PG	PPO	SOD	BI	WL	TSS	TA	pH	AsA
TPC	1													
FM	0.968**	1												
RR	-0.757	-0.791	1											
PME	-0.322	-0.304	0.522*	1										
CL	-0.742	-0.772	0.968**	0.567**	1									
PG	-0.699**	-0.739	0.954**	0.690**	0.963**	1								
PPO	-0.865**	-0.860**	0.787	0.446	0.737**	0.763**	1							
SOD	0.874**	0.844**	-0.694	-0.247	-0.617	-0.588*	-0.854	1						
BI	-0.866	-0.864	0.585*	-0.033	0.511*	0.468	0.785**	-0.868	1					
WL	-0.937**	-0.904**	0.715	0.476	0.690**	0.683**	0.886**	-0.916**	0.825**	1				
TSS	-0.754	-0.717	0.696	0.778	0.707**	0.753**	0.764**	-0.700**	0.469	0.878**	1			
T	0.912**	0.934**	-0.726	-0.226	-0.732	-0.685*	-0.792	0.746**	-0.837	-0.822	-0.617**	1		
pH	-0.601**	-0.685	0.402*	0.104	0.489	0.451	0.403	-0.274	0.486	0.445	0.323	-0.793**	1	
AsA	0.799**	0.763**	-0.608	-0.290	-0.501*	-0.518*	-0.843	0.947**	-0.801	-0.903	-0.734**	0.640**	-0.141	1

**Significant at 5%, **Significant at 1%, TPC – Total phenolic content, FM- Firmness, RR- Respiratory Rate, PME- Pectin methyl estrase, CL- Cellulase, PG- Polygalacturonase, PPO- Polyphenol oxidase, SOD- Superoxide dismutase, BI- Browning Index, WL- Weight loss, TSS- Total Soluble Solids, TA- Total acidity %, AsA- Ascorbic acid*

Correlations Analysis

The correlation among fruit quality parameters that had supremacy on the postharvest storage life of pears was examined by Pearson's coefficient of correlation and was computed by a linear association between parameters. The statistically significant associated combinations of variables were evaluated with the correlation coefficient. The results further declared that an increase in weight loss led to a reduction in fruit firmness during storage at ambient conditions in both cultivars (Table 1 & 2). Fruit firmness was negatively correlated with weight loss throughout storage in Patharnakh and Punjab Beauty (-9.04 and -0.928), respectively. Likewise, firmness elucidated a negative relationship with cellulase, polygalacturonase (PPO), and pectin methylestrase (PME) enzymes in Patharnakh and Punjab Beauty cultivars. The increased activity of cellulase and polygalacturonase enzymes in both cultivars causes degradation of cell wall polysaccharides. The relationship revealed that the cell wall polysaccharides in pear were associated with fruit softening. Similar results were also reported in previous studies conducted with guava (Abu-Bakr and Elbashir 2003) and pear (Adhikary *et al.,* 2022). In both cultivars, browning of the fruit during storage was negatively correlated with TPC (-0.866 and -0.882) and positively correlated with PPO activity (0.785 and 0.843), respectively. This decrease in TPC in fruit during storage is directly related to the oxidation of the PPO enzymes, where phenol turns into a quinone compound (Capotorto *et al.,* 2017).

Table 2: Pearson correlation analysis of different physio-biochemical attributes of Punjab Beauty

Variables	TPC	FM	RR	PME	CL	PG	PPO	SOD	BI	WL	TSS	TA	pH	AsA
TPC	1													
FM	0.973**	1												
RR	-0.675	-0.722**	1											
PME	-0.257	-0.265	0.567*	1										
CL	-0.637	-0.677	0.985	0.510*	1									
PG	-0.659	-0.714	0.978**	0.677*	0.955**	1								
PPO	-0.905**	-0.956	0.797	0.333	0.760**	0.803**	1							
SOD	0.939**	0.942**	-0.637	-0.114	-0.619**	-0.606	-0.873	1						
BI	-0.882	-0.923	0.495*	-0.015	0.440	0.480	0.843**	-0.923	1					
WL	-0.940**	-0.928	0.609	0.389	0.579	0.642**	0.852**	-0.908**	0.826**	1				
TSS	-0.688	-0.655	0.652**	0.787*	0.636	0.730**	0.642**	-0.585	0.393	0.825**	1			
T	0.924	0.941**	-0.698	-0.164	-0.667**	-0.670	-0.851	0.957**	-0.926	-0.882	-0.572*	1		
pH	-0.933	-0.954	0.530*	0.062	0.489	0.523*	0.869**	-0.958	0.984**	0.906**	0.522*	-0.946**	1	
AsA	0.960**	0.956**	-0.627	-0.311	-0.599**	-0.643	-0.881	0.944**	-0.874	-0.993**	-0.768	0.919**	-0.942**	1

**Significant at 5%, **Significant at 1%, TPC – Total phenolic content, FM- Firmness, RR- Respiratory Rate, PME- Pectin methyl estrase, CL- Cellulase, PG- Polygalacturonase, PPO- Polyphenol oxidase, SOD- Superoxide dismutase, BI- Browning Index, WL- Weight loss, TSS- Total Soluble Solids, TA- Total acidity %, AsA- Ascorbic acid*

This study represents the storage competency of fruits of pear cultivars 'Patharnakh' and 'Punjab Beauty' under ambient conditions. The results showed loss in weight, firmness and sugar content in fruits of both the cultivars. The activities of cellulase, PG and PME showed the positive effect on fruit softening; hence spoilage occurred during storage of fruits. It can be summarized from the results that reduction in sugar content and fastening of activities of cell wall degrading enzymes between 65 days after storage in 'Patharnakh' and 32 days in 'Punjab Beauty' fruits makes them less desirable for further storage under ambient temperature conditions.

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