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# Influence of hexanal concentration and exposure time on quality of cold stored apples (*Malus domestica*)

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ABSTRACT

The present study was carried out during 2019 at ICAR-IARI to evaluate the effectiveness of hexanal, a volatile aldehyde in prolonging the storage life of apple cv. Royal Delicious while maintaining its quality. Application of different concentrations of hexanal as a solution @ 0.01%, 0.02% and 0.03% v/v for 2, 3 and 4 min was tested. Fruits given no hexanal treatment were taken as control. The treated and control apple fruits were stored under 1-2°C and 80-90% RH for a period of 90 days and evaluated for physical and biochemical changes at regular interval of 15 days. The study demonstrated that exogenous application of 0.01% hexanal applied as dip treatment for 3 minute soaking time given to freshly harvested Royal Delicious apple fruits reduced physiological loss in weight, retained better pulp firmness and maintained the overall quality of the fruits up to 3 months storage under 1-2°C. Thus, hexanal as a dip treatment holds promise in prolonging postharvest storage life and preserving quality of apple cv. Royal Delicious.

**Keywords**: Apple, Anthocyanin, Colour, Hexanal, Storage, Quality

Apple fruits are relished for their distinct taste, crunchiness and nutritive value. Since, consumer preference for taste, flavor and other quality attributes of apple vary according to geographical region, more than 7500 different cultivars are grown across continents. World apple production is estimated to be 89.33 million MT of which 23.27 lakh tonnes is cultivated in India (Anonymous 2017-18). For marketing purpose, the fruits are transported from temperate hilly terrain of cultivation to far flung warmer areas where they may be stored for periods of up to 6 months before marketing. Improper harvesting, handling, storage, transportation and marketing practices tend to bring about losses in their quality. Postharvest losses of fruits and vegetables in India are estimated between 30–35% (Sekhar *et al*. 2013). In case of apple, nearly 10–25% losses are attributed to fungal diseases such as of grey mold and blue mold (Van Zeebroeck *et al*. 2007). Prevalence of high temperature in the supply chain makes the fruits more liable to rapid senescence and decay. Hence, to control and combat these postharvest changes, there is a need for a cost effective and eco-friendly technology. For apple, methods to mitigate fruit spoilage can be best applied during the storage period in the marketing channel. Hexanal, a cost effective antimicrobial volatile aldehyde, can be applied as a postharvest dip treatment before the fruits are put in cold stores. It has been approved as a food additive by the U.S.F. D.A with an oral-mammalian $LD_{50}$ value of 3700 mg/kg (EAFUS 2016). Hexanal is effective in inhibiting the enzyme phospholipase D that is responsible for membrane degrading processes. Hexanal in solution has shown prolonged storage life and maintenance of quality in tomatoes, bananas and papaya (Cheema *et al.* 2014, Venkatachalam *et al*. 2018, Hutchinson *et al.* 2018). But, scanty literature is available for its use during postharvest storage of apple for maintaining quality and extending their storage life.

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## MATERIALS AND METHODS

Apple cv. Royal Delicious was harvested at Seobagh in Himachal Pradesh at optimum maturity stage, sorted, graded and transported within 24 h in cartons (20 kg capacity) to Division of Food Science and Postharvest Technology, ICAR-Indian Agricultural Research Institute, New Delhi during 2019. The fruits were kept in cold store (1-2°C) prior to treatment.

*Hexanal treatment*: Hexanal solutions of 0.01%, 0.02%, 0.03% (v/v) were prepared in water. Apple fruits were dipped in the solution for 2, 3 and 4 min. Water dipped fruits were taken as control. Fruits were then air dried. Post each treatment, fruits were stored under 1-2°C at 80-90% RH. Both treated and control fruits were analyzed for different quality attributes at interval of 15 days till 90 days of storage period.

*Physiological loss in weight (PLW)*: Every single fruit was weighed at regular intervals with the help of an electronic balance (Citizen make) for the measurement of weight loss during storage. PLW was calculated as given below and data was expressed in percentage.

$$\text{PLW (\%)} = \frac{\text{Initial weight - Weight after storage interval}}{\text{Initial weigt of fruits}} \times 100$$

*Fruit colour (hue)*: The colour of apple peel was determined by using colorTec PCM machine in L*, a* and b* coordinates. L* indicates the lightness co-efficient while a* indicates a hue of red purple (-a*) to bluish green (+a*) and b* indicates yellow (+b*) to blue (-b*). Peel colour was measured at 3 random places on each fruit surface. Hue angle of fruits was determined by the equation given by Velickova *et al.* (2013).

$$\text{Hue angle} = \tan^{-1} (b/a)$$

*Pulp firmness*: Firmness of apple fruit pulp was determined using a puncture probe of texture analyzer with a 2mm/s pre-test speed, 0.5 mm/s test speed till a distance of 5 mm and 3 mm/s post-test speed. Maximum force during the puncture of the apple pulp, expressed in Newton (N) was used to denote the pulp firmness.

*Titratable acidity (TA) and total sugars*: Titratable acidity of the control and treated apples was determined by the method described by Ranganna (2007) while total sugar content was estimated as described in AOAC (2000).

*Total anthocyanin content*: The total anthocyanin content in apple peel was determined by using the pH-differential method (Nayak *et al*. 2019). The pigment content was calculated and expressed as cyanidin 3, 5-glucoside per kg fresh weight, based on total volume of extract and weight of sample.

*Statistical analysis*: The treatments were examined statistically using factorial CRD with 50 fruits taken in each treatment with 3 replications. The data obtained was analyzed using analysis of variance (ANOVA) by calculating C.D. (Panse and Sukhatme 1984) using MS Excel.

## RESULTS AND DISCUSSION

*Physiological loss in weight (PLW)*: Hexanal treatment significantly affected the loss in weight of apple fruits during 90 day storage period under cold conditions. The weight loss of apple fruits augmented with increase in storage days irrespective of treatment applied. Apple fruits did not reach the 10% PLW level that is usually considered unfit for marketing of apple fruits. Nevertheless, control fruits demonstrated highest PLW (7.71%) till end of 90 days storage. Dipping of fruits in hexanal solution resulted in lower loss in weight that differed significantly with treatment dose and time. Least values of PLW (6.59%) were observed for fruits dipped in 0.01% hexanal solution for 3 min. The incremental PLW during storage maybe due to the loss of moisture and increased respiration rate during storage. Previously, Jan *et al.* (2012) reported an increase in weight loss of apples during 150 days of storage. Since, the exogenous application of hexanal inhibits the phospholipase D (PLD) and lipooxygenase enzymes present in the fruit peel, it assists in delayed ripening process and hence a lower PLW (Thakur *et al.* 2017).

*Pulp firmness*: During the course of the experiment, there was a gradual decline in pulp firmness with progressive increase in storage period (Table 1). Hexanal treated apple fruits exhibited comparatively higher pulp firmness than untreated (control) ones. At the end of storage period, untreated apple fruits showed maximum reduction in pulp firmness (~40%). With regard to hexanal treatment, lower dose application was most favourable to retain pulp firmness. Fruits subjected to hexanal dip @ 0.01% for 3 min resulted in about 73% retention in firmness of the pulp. Dipping apple in higher dose of aqueous hexanal (0.03%) retained 61-65% pulp firmness (Table 1). Hexanal application on apple fruits influenced the firmness positively for a longer

Table 1 Influence of hexanal treatments on pulp firmness (N) of Royal Delicious apples during cold storage

| Treatment | Storage period (days) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 0 | 15 | 30 | 45 | 60 | 75 | 90 | Mean |
| Control (untreated) | 2.74 | 2.59 | 2.45 | 2.23 | 1.97 | 1.83 | 2.74 | 2.20 |
| Hexanal dip | 2.77 | 2.60 | 2.49 | 2.28 | 1.95 | 1.82 | 2.77 | 2.23 |
| 0.01%, 2 min | 2.65 | 2.62 | 2.52 | 2.32 | 2.22 | 2.10 | 2.65 | 2.35 |
| 0.01%, 3 min | 2.70 | 2.61 | 2.46 | 2.30 | 2.20 | 2.04 | 2.70 | 2.30 |
| 0.01%, 4 min | 2.70 | 2.58 | 2.40 | 2.29 | 2.15 | 2.00 | 2.70 | 2.27 |
| 0.02%, 2 min | 2.67 | 2.52 | 2.37 | 2.25 | 2.07 | 1.87 | 2.67 | 2.22 |
| 0.02%, 3 min | 2.70 | 2.58 | 2.33 | 2.22 | 1.96 | 1.80 | 2.70 | 2.19 |
| 0.02%, 4 min | 2.68 | 2.56 | 2.30 | 2.16 | 1.97 | 1.79 | 2.68 | 2.17 |
| 0.03%, 2 min | 2.72 | 2.56 | 2.29 | 2.14 | 1.94 | 1.77 | 2.72 | 2.15 |
| 0.03%, 3 min | 2.70 | 2.56 | 2.29 | 2.14 | 1.95 | 1.74 | 2.70 | 2.15 |
| 0.03%, 4 min | 2.74 | 2.59 | 2.45 | 2.23 | 1.97 | 1.83 | 2.74 | 2.20 |
| Mean | 2.70 | 2.58 | 2.39 | 2.23 | 2.04 | 1.88 | 1.75 | |
| CD ($P<0.05$) | Treatment = 2.17; Storage days= 1.81; Treatment × storage days =3.76 | | | | | | | |

Table 2 Influence of hexanal treatments on hue (°) of Royal Delicious apples during cold storage

| Treatment | Storage period (days) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 0 | 15 | 30 | 45 | 60 | 75 | 90 | Mean |
| Control (untreated) | 81.11 | 76.00 | 71.12 | 63.36 | 58.83 | 50.77 | 46.19 | 63.90 |
| *Hexanal dip* | | | | | | | | |
| 0.01%, 2 min | 81.45 | 78.91 | 73.60 | 66.85 | 62.12 | 55.82 | 51.99 | 67.25 |
| 0.01%, 3 min | 81.78 | 79.67 | 76.13 | 71.33 | 70.13 | 68.06 | 60.84 | 72.56 |
| 0.01%, 4 min | 82.45 | 79.10 | 74.22 | 71.85 | 66.79 | 64.81 | 59.09 | 71.19 |
| 0.02%, 2 min | 82.45 | 77.76 | 72.92 | 69.09 | 68.11 | 64.32 | 55.85 | 70.07 |
| 0.02%, 3 min | 80.92 | 76.66 | 73.39 | 66.66 | 61.14 | 55.63 | 52.04 | 66.63 |
| 0.02%, 4 min | 82.26 | 77.76 | 73.43 | 66.27 | 59.84 | 53.33 | 51.90 | 66.40 |
| 0.03%, 2 min | 81.30 | 76.24 | 66.85 | 66.16 | 58.41 | 53.81 | 49.87 | 64.66 |
| 0.03%, 3 min | 82.45 | 76.24 | 68.81 | 64.54 | 59.22 | 54.66 | 50.93 | 65.26 |
| 0.03%, 4 min | 82.45 | 76.23 | 70.95 | 67.02 | 58.60 | 54.09 | 48.12 | 65.35 |
| Mean | 81.86 | 77.46 | 72.14 | 67.31 | 62.32 | 57.53 | 52.68 | |
| CD (P<0.05) | Treatment = 2.17; Storage days= 1.81; Treatment × storage days =3.76 | | | | | | | |

period of time, primarily because hexanal is a potent inhibitor of enzymes that disturb the membrane integrity. Hexanal application at low concentration as dip maintained the best firmness, primarily because of damaging effect at higher concentration. Furthermore, hexanal has been reported to down regulate the genes responsible for enzymes involved in ethylene biosynthesis (Tiwari and Paliyath 2011), resulting in better pulp firmness (Paliyath and Subramanian 2008). Further, as the days of storage progressed, the pulp firmness decreased which might be due to continued ripening of fruits during storage, wherein, insoluble pectic substances of the cell wall are degraded into soluble pectin, resulting in pulp softening (Solomos and Laties 1973).

*Fruit colour (Hue)*: With the advancement in storage, a reduction in hue was observed irrespective of treatment (Table 2). Control fruits recorded the highest reduction in hue (61%) at end of 90 day storage under cold conditions. Hexanal dip @ 0.01% for 3 min resulted in least reduction (~44%) in hue in comparison to all other treatments. In fruits where both red and yellow intensity change, as in case of Royal Delicious apple fruits, the decrease in hue angle is a reliable parameter to express the increase in red colour intensity. The hue angle (60.84) was the highest on the 90 day in hexanal (0.01%) dip treated fruits while the untreated apples showed lower hue 46.19 (Table 2). The slower colour development in hexanal treatment apple may be because of low rate of ripening, reduction in ethylene evolution, respiration rate and lower anthocyanin pigment development in the fruits as also reported earlier by Paliyath and Subramanium (2008) in apples sprayed with hexanal prior to harvest.

*Titratable acidity (TA)*: A gradual rise in the titratable acidity was observed with the advancement of storage followed by a decline irrespective of the treatment (data not shown). However, treatments did not show any significant ($P \leq 0.05$) variation of TA during the entire storage period of 90 days under 1-2°C. The untreated apple fruits reached maximum acidity values on 45 days of storage (0.274%) followed by a decline to a final value of 0.233% after 90 days of cold storage. Hexanal dipped apple fruits showed a rise in acidity till 60 days irrespective of dose and time. This might be as a result of retardation of metabolic and anabolic processes in treated fruits. Hexanal dip @ 0.01% for 3 min given to apples recorded the highest TA (0.275%) compared to other hexanal treatments on 90$^{th}$ day of storage. A decline in the TA of the untreated fruits is due to utilization of organic acid during ripening process and onset of senescence. In contrast, hexanal treated fruits maintained an increasing trend for TA till end of storage, possibly due to a reduction in the respiration rate and delayed ripening. Similar results have been reported by Hutchinson *et al*. (2018) and Yumbya *et al.* (2018) with EFF in papaya and banana fruits, respectively. Cheema *et al*. (2014) also recorded similar pattern in TA in tomatoes given a dip treatment of 2.5 min in 0.02% hexanal solution and 0.02% EFF.

*Total sugars*: Data (Fig 1) depicts the changes in total sugar content in untreated and hexanal treated fruits of apple cv. Royal Delicious. Irrespective of treatment, all samples showed an initial increase followed by a decline in total sugar levels. The total sugar content varied significantly ($P \leq 0.05$) with treatment and storage days. For the untreated fruits, the increase of total sugars was observed till 45 days of storage that declined thereafter. The 0.01% hexanal dip for 3 minute yielded maximum total sugar content till 90 days of storage (11.74%). The initial rise in total sugars during storage is because of conversion of starch to sugars during ripening (Magein and Leurquin 2000, Rivera-Espinoza *et al.* 2005). Owing to curbing of respiratory metabolic activities, hexanal treated fruits showed a rise in sugars for prolonged storage period. Interaction between treatment and storage period was also found to be significant.

*Total anthocyanin content*: A significant increase in the peel anthocynain content across all treatments was observed

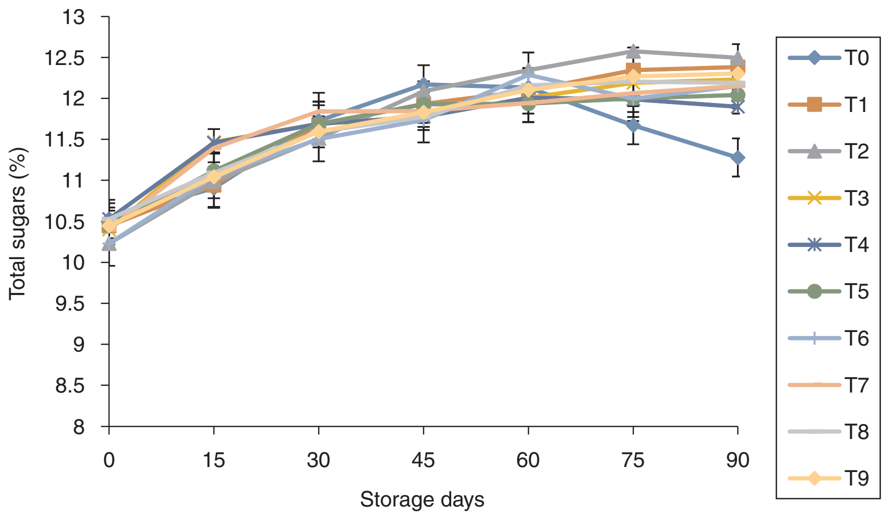


Fig 1 Influence of hexanal dip on total sugars (%) of Royal Delicious apple fruits during cold storage.

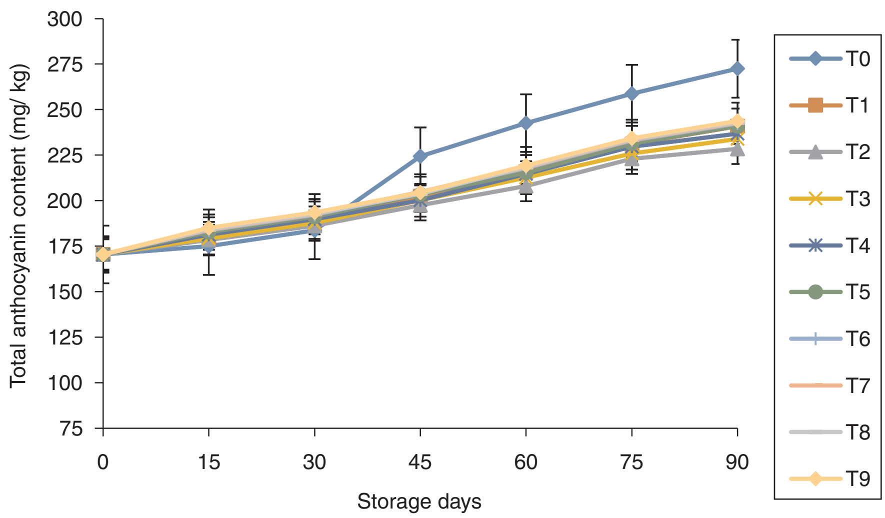


Fig 2 Influence of hexanal dip on total anthocyanin content of Royal Delicious apple fruits during cold storage.

with the advancement in storage period (Fig 2). However, untreated fruit demonstrated the highest rise (~60%) in anthocyanin content from 170.43 mg/kg to 272.43 mg/kg FW. In hexanal treated apple fruit, the rise in anthocyanin content varied from ~34-43%. Least increase in peel anthocyanin (from 170 to 228.47 mg/kg FW) was recorded in fruits treated with 0.01% aqueous hexanal for 3 min. Hexanal treated apple demonstrated lower total anthocyanin content in comparison to untreated fruit. Such phenomena may primarily be because of suppressed ripening in these fruits with simultaneous slow pigment synthesis as also reported by Sharma *et al*. (2010) in cherries. Anthocyanin content increased with the advancement in storage interval due to gradual ripening of the fruit (Fig 2), yet hexanal treated fruit demonstrated a suppressed rise in total anthocyanin content in comparison to untreated fruit.

The current study demonstrated that exogenous application of hexanal applied as dip treatment given to freshly harvested Royal Delicious apple fruits reduces physiological loss in weight, retains pulp firmness and improves the overall quality of the fruits up to 3 month storage under 1-2°C. Dipping of freshly harvested fruits in 0.01% hexanal for 3 min was found to be the best to maintain overall fruit quality. Hexanal is an economical and commercially viable alternative to preserve apple fruit quality during prolonged storage. Hence, postharvest hexanal application of aqueous hexanal holds promise in prolonging postharvest storage life and preserving quality of apple cv. Royal Delicious in a cost effective manner.

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