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Predicting the optimum harvesting dates for different exotic apple varieties grown under North Western Himalayan regions through acoustic and machine vision techniques

Nazrana Rafique Wani^a, Syed Zameer Hussain^{a,*}, Gopinath Bej^b, Bazila Naseer^{a,*}, Mushtaq Beigh^a, Ufaq Fayaz^a, Tamal Dey^b, Abhra Pal^b, Amitava Akuli^b, Alokesh Ghosh^b, B.S. Dhekale^c, Fehim J. Wani^d

^a Division of Food Science and Technology, Sher-e-Kashmir University of Agriculture Sciences and Technology of Kashmir, Shalimar 190025, India ^b Centre for Development of Advanced Computing (C-DAC), Kolkata, India

^c Division of Agricultural Statistics, Sher-e-Kashmir University of Agricultural Sciences and Technology (SKUAST) Kashmir, Shalimar 190025, India

^d Division of Agricultural Economics & Statistics, Faculty of Agriculture, Wadura, SKUAST-Kashmir, J&K, India

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Keywords: Non-destructive Correlation Harvest dates Varieties Consumption	Acoustic system and machine vision were used to evaluate the effects of different harvest dates on the quality and sensory attributes of exotic apple varieties of North Western Himalayan. <i>Gala Redlum</i> (V1) was harvested at 110 (H1), 120 (H2) and 130 (H3) Days from Full Bloom (DFFB); <i>Red Velox</i> (V2) and <i>Super Chief</i> (V3) were harvested at 130 (H1), 140 (H2) and 150 (H3) DFFB. Highest acoustic coefficient (21.13) and firmness (20.72 lbs) recorded at first harvest date (H1) decreased significantly ($p \le 0.05$) (19.86 to 17.90 lbs) at second harvest (H2) and (17.77 to 16.80 lbs) at third harvest date. Highest starch iodine rating (3.72); anthocyanin content (24.81 mg/100 g); total soluble solids (12.10 %); total sugars (8.75 %) were recorded at H3 in all the varieties. For <i>Gala Redlum</i> (V1) 130 DFFB and for <i>Red Velox</i> (V2) and <i>Super Chief</i> (V3) 150 DFFB were predicted as suitable harvesting dates for table consumption.

Introduction

Apple (*Malus domestica*) is one of the largest fruit produced in the world. The annual production of apples was recorded as 81.60 million tonnes during 2021–22 (USDA, 2022). India with a production of 2057 MT stands fifth in the global apple production. In India, apple is mostly grown in Jammu and Kashmir (J&K), Himachal Pradesh, Uttarakhand, Arunachal Pradesh and Nagaland. However, J&K is the leading apple producer and accounts for about 77% of total apple production of the country. Many indigenous apple varieties like *Ambri, Kesari, Hazratbali* and *Maharaji* are grown in J&K and studies related to their maturity indices and quality are well documented (Doryanizadeh et al., 2017). From the last few years several exotic varieties have been introduced in J&K to boost the apple industry of India. The purpose of introducing exotic varieties in the region is to boost the apple production, productivity and to have maximum high-grade apples for higher economic returns. The traditional apple cultivation is being replaced by exotic/

high density apple orchards in a systematic manner. However, the optimum date of apple maturity for these exotic varieties under the climate conditions of J&K is yet to be standardized. Since altitude, climate and soil conditions are the critical factors which influence the physicochemical, textural and sensory quality of apples (Kviklys et al., 2022). Therefore, there is an urgent need to standardize the harvesting time for various exotic varieties of apple for fresh consumption and storage using non-destructive and destructive methods before popularizing these exotic apple varieties in Jammu and Kashmir. The prerequisite for maximizing the economic returns from apples is their optimum harvest date, as early or late harvesting leads to deleterious effects on its quality. If the fruit is harvested too early, it is more prone to bruise damage during storage and is more susceptible to physiological disorders resulting in smaller fruit weight, poor eating qualities, inferior surface color, less sugar content and poor flavor due to poor production of aroma generating compounds (Jemrić et al., 2016). In contrast, if the fruit is harvested too late, it results in minimum storage life and high

* Corresponding authors. E-mail addresses: zameerskuastj@rediffmail.com (S.Z. Hussain), sheikhbazila@gmail.com (B. Naseer).

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susceptibility to rot, mechanical damage, CO_2 injury and senescence. Such fruits are less crispy and juicy with high sugar / acidity ratio resulting in poor sensory appeal.

Various destructive and non-destructive methods are used to assess the maturity indices and overall quality of apple. However, destructive methods employed for the purpose are tedious and damages the samples. Non-destructive methods are quick, less tedious and perform the quality analysis without damaging the fruit. However, accuracy and efficiency of these methods depends upon the reliability and relevance of the technique employed (Ali and Hashim, 2022). Therefore, it is imperative to understand the mechanism which could allow the operator to monitor and decide on non-destructive technique required to assure the fruit quality. Texture is a well-known quality attribute of apples and is closely related to the structural integrity of fruit tissues, soluble solids and fruit firmness. Textural attributes like crispness, hardness and crunchiness have the strong positive influence on overall quality and thus govern the consumer acceptability of fruits (Kim et al., 2022). Texture of fruits is usually measured by texture analyser, which is a destructive technique. During texture analysis, the sound produced by compression probe is recorded while fruit is destroyed. Acoustic technique quantifies the elastic waves generated within a material as a result of internal cracking without destroying the sample (Zdunek et al., 2010). Therefore, acoustic systems can be an ideal non-destructive technique to be explored for detection of apple firmness.

Colour is another important maturity index which governs the quality of apples. However, colour is usually measured through Hunter Lab colorimeter, which is time consuming. Moreover, the accuracy of the method depends upon the calibration of the setup and the conditions under which colour analysis is done. Machine vision is an engineering technology that combines mechanics, optical instrumentation, electromagnetic sensing, digital video and image processing technology. As an integrated mechanical-optical-electronic-software system, machine vision has been widely used for examining, monitoring, and controlling a very broad range of applications like sorting and grading of fruits based on colour and surface defects; determining shape, size, volume, textural feature and surface area (Gururaj et al., 2022) of fruits. Blasco, Aleixos, and Moltó, (2003) used machine vision system to determine the size, colour, stem location and detection of external blemishes in case of oranges, peaches and apples. The colours of the fruits estimated by the system were well correlated with the colorimetric index values that are currently used as standards.

Therefore, it has a good scope to emerge as a quick and nondestructive method for determining the colour. Non-destructive techniques using acoustic characteristics have been applied for measuring the firmness of several fruits such as apples, pears, tomatoes, and mandarins (Nishani, Deshpande and Nagajjanavar, 2022). Fathizadeh et al. (2020) measured the firmness of Royal Gala apples using acoustic vibration response method under two storage conditions. Nondestructive techniques using machine vision has been used for detecting ripeness level in Red Delicious apple and also, for detection of defect, contour and size of Red Fuji apples (Wang et al., 2022).

However, very limited literature is available regarding the correlation of destructive and non-destructive techniques. Such studies are needed to create a data base for interpreting the quality through nondestructive approach. Therefore, the present study was planned with an aim to evaluate different quality attributes of exotic apple varieties at different harvesting dates grown under Northern highland Himalayan regions of Kashmir through both destructive and non-destructive approaches. Furthermore, a correlation analysis of results obtained through destructive and non-destructive methods was also done. Such information will be very useful for future researchers working in nondestructive field.

Materials and method

Materials

Three exotic apple varieties namely, *Gala Redlum* (V1), *Red Velox* (V2) and *Super Chief* (V3) were manually harvested by two labourers in morning time at three different dates from high density apple orchard of Sher-e- Kashmir University of Agricultural Sciences and Technology, Kashmir (SKUAST-K), J&K, India. *Gala Redlum* was harvested at 110 (H1), 120 (H2) and 130 (H3) Days from Full Bloom (DFFB) while as *Red Velox* and *Super Chief* were harvested at 130 (H1), 140 (H2) and 150 (H3) DFFB as per the commercial maturity chart. The apples were cleaned to remove some dust and were kept in separate high density polyethylene crates (HDPE) under ambient conditions (Temperature = 30.18 ± 4.12 ; RH = 77.08 ± 11.53) for evaluation. Approximately twenty (20 ± 5) apples pertaining to each variety and each harvest date were taken for experimental measurement of various physico-chemical and quality parameters. Chemical reagents used in the research work were procured from Sigma Aldrich (USA).

Acoustic set-up for non-destructive quality evaluation of apples

Firmness of apples was estimated through non-destructive way using acoustic setup (Fig. 1a). Digital signal processing and analysis techniques were applied on the acoustic signals generated through acoustic setup. A sample holding arrangement with rubber padded caps on the top and bottom sides was used to hold the apples, while hammered with a low-mass Teflon bar. An electronic controller and a solenoid valve were used to lock and release the hammer electronically (Fig. 1b). The electronically controlled Teflon bar generated the acoustic signal while hitting the apples. The generated signals were recorded by the microphone placed inside the setup and were stored in the computer in a.wav file on the desktop within a duration of 2 s. The signal was read from the. wav file into a one-dimensional array after clipping from right and left sides to remove undesirable sounds (sound of the solenoid valve). A moving average technique was also applied to reduce the noise from the signal. A 25% overlapping moving mask window was considered in the time domain while signal processing to ensure less information loss. A frequency domain analysis was applied to the masked array. The features extracted in the frequency domain were fundamental frequency and power spectrum. The fundamental frequency has a positive correlation with firmness, therefore high-frequency sound indicated good quality fruits. The data generated through acoustic setup as well as the manual firmness data were used to create a mathematical correlation model for the estimation of firmness. Based on the mathematical correlation model, the firmness index of apples was predicted.

Determination of physico-chemical properties of apples using destructive methods

Firmness

Penetrometers are widely used to measure apple firmness and to estimate textural attributes such as crispness. Apples were held steady on a firm surface and the probe of Effegi hand-held penetrometer (FT 327) was pushed into the fruit to a depth of 8 mm, corresponding to a mark inscribed on the shaft of the probe. The fruit firmness was measured on the scale provided in the penetrometer in lbs.

Starch iodine rating

Starch iodine test was carried out to determine conversion pattern of starch into sugars. Potassium iodine solution was prepared by mixing 25 g of potassium iodide into 150 ml of distilled water and then 10 g of iodine crystals were added to it and left overnight. Solution was filled up to one liter with distilled water. Fruits were cut at right angles to core, approximately halfway from stem to calyx end. Fruit was dipped into the iodine solution. The staining of fruit was rated on a scale ranging from 1



a)



b)



Fig. 1. (a) Integrated setup of acoustic and machine vision (b) Non-destructive acoustic system showing different components (c) Machine vision system showing different components.

(100% starch) to 6 (0% starch) (Brookfield et al., 1997).

Anthocyanins

Anthocyanin content was estimated from 1 g of fruit peel with a known volume of methanolic HCl. 95% methanol and 1.5 molL^{-1} HCl in the ratio of 85:15, (v/v) were poured in a beaker contained 1gm of apple peel and stored overnight under ambient conditions. The mixture was filtered and a small aliquot was diluted with methanolic HCl. Optical density (OD) was checked at 530 nm using UV–VIS Double Beam Fixed Bandwidth Spectrometer (IG-27DS) (Iglesias et al., 2012). Anthocyanin content was calculated using Equation (1):

Anthocyanin content (mg/100g) =
$$\frac{totaloptical density/100}{98.20}$$
 (1)

Pectin

For extraction of pectin, protocol reported by Virk and Sogi (2004) was followed. 50 g of whole apples were boiled in 0.01 molL⁻¹ HCl (300 ml) for around 30 min and then filtered. The residue was washed with hot water and to the residue, 0.05 molL^{-1} (100 ml) HCl was added. The residue was boiled for 20 min and then washed again with hot water. The collected residue was again added to 0.03 molL^{-1} HCl (100 ml) and boiled for 10 min. The filtrate was pooled and volume was made up to 500 ml. Aliquot (100–200 ml) was taken and 250 ml of water was added and then the sample was neutralized with NaOH. Afterwards an excess of 10 ml of 1 molL⁻¹ NaOH was added to it and the mixture was left to stand overnight. Then 50 ml of 1 molL⁻¹ acetic acid was added and after 5 min 25 ml of 1 molL⁻¹ CaCl₂ was also added to it. After 1 h, the mixture was boiled for 2 min and filtered through Whatman No.1 and allowed to dry. Pectin in the form of calcium pectate was measured using Equation (2):

Calcium pectate % =
$$\frac{wt. of calcium pectate \times 500}{ml of filtrate taken for estimation \times wt.of sample \times 100}$$
 (2)

Determination of juice yield, total soluble solids, titratable acidity and sugars

Apples (approximately 500 g) were crushed in a lab scale juicer (Bajaj Neo JX4, India) and the juice was extracted to measure the juice yield, total soluble solids (TSS), titratable acidity and sugars.

Juice yield

Juice yield percentage from known weight of apples was calculated using Equation (3):

$$Juice \ yield = \frac{weight \ of \ juice \ extracted}{weight \ of \ sample} \ \times \ 100$$
(3)

Total soluble solids

Total soluble solids (TSS) of apple juice were measured by using a hand/Abbe refractometer of range 0-32° brix (Perma, Japan) and readings were expressed as degree brix at 20° C using reference table (AOAC 2012). The calibration was done with distilled water. Two drops of fresh apple juice were placed on the refractometer lens and TSS was measured (Ghinea et al., 2022).

Titratable acidity

The extracted juice was used to estimate the acidity of fruit by titrating the fruit pulp extract with 0.1 N NaOH using phenolphthalein as indicator (Ranganna, 1986) and acidity was expressed in terms of percentage malic acid.

Estimation of sugars

Lane and Eynon method was followed for estimation of total,

reducing and non-reducing sugars. Apple juice (10 ml) was neutralized with NaOH using phenolphthalein indicator. Lead acetate (2 ml) was added to the neutralized sample and kept undisturbed for 10 min. Potassium oxalate (2 ml) was added and the final volume was made up to 250 ml using distilled water. Then filter it using Whatman no.1and titrated it against Fehling's A & Fehling's B solution (5 ml each and distilled water 25 ml) using methylene blue as an indicator. Total and reducing sugars were estimated using Equation (4):

Total and Reducing sugars (%) =
$$\frac{Ff \times Vd}{Tv \times Ws} \times 100$$
 (4)

where Ff = Fehling's factor; Vd = volume made up; Tv = titre value; Ws = weight of sample taken.

Non-reducing sugars were estimated by subtracting reducing sugars from total sugars (Ningegowda et al., 2022).

Sensory evaluation

Sensory evaluation of apple varieties harvested at different dates was performed by a team of 35 panellists selected from the scientific staff of Division of Food Science & Technology, SKUAST-K, J&K, India. Quality of apples in terms of various sensory attributes like crispness, tenderness, juiciness, mealiness, and appearance were rated on a continuous 9point hedonic scale with the lowest intensity 0 indicating "dislike extremely" and highest intensity 9 indicating "like extremely". Texture descriptors for sensory analysis were selected as per the recommendations of Mehinagic et al. (2003) for apples. Apples were washed and cut into eight slices. Two slices of unpeeled apples from each variety and harvest date were presented to each panelist under good illumination at room temperature. The sensory assessors were instructed to rinse the mouth with potable water and consume bland crackers between sample analyses. The samples were previously randomized to avoid position bias, and coded with random three-digit numbers. The panelists rated the individual samples for different sensory attributes i.e., crispness, tenderness, juiciness, mealiness, and appearance and overall acceptability score was calculated as the average of these sensory attributes.

Evaluation of principle dimensions and color coordinates by machine vision system and conventional method.

Experimental setup of machine vision

The imaging system used for quality analysis of apple fruit in this study mainly consists of an image acquisition system (lighting arrangements and camera setup), frame grabber, image processing and analysis software and computer hardware (Fig. 1c). At first, the image of the sample was captured and circular cropping of the image was performed. Background segmentation was applied to remove anything other than the sample from the image. Afterwards, connected component labeling was applied and noise was removed based on the labeled area. Then, shape, size, and color-based features were extracted. These features were correlated with the quality parameters of apple.

Measurement of length, width and thickness

Linear dimensions of length, "L"; width, "W" and thickness, "T" were measured using manual method also. A DVC (digital vernier caliper: model no CD-6['] CSX, Mitutoyo-Japan) having least count of 0.01 mm was used for measurement for L, W, and T.

Measurement of colour attributes

Colour of apples was determined according to standard method of AOAC (2012). Hunter Lab colour flex model A60-1012–312 (Hunter Associates laboratory, Reston, VA) calibrated each time with a white and black standard was used for measurement. Results were expressed as Hunter colour values of L*, a* and b*, where L* denotes lightness and darkness, a* redness and greenness and b* yellowness and blueness.

Streif index

The true quality of apple cannot be explained by individual parameter. Streif index takes into account different quality attributes such as firmness, soluble solid content (SSC) and starch content in determining the harvesting date (Musacchi and Serra, 2018). Streif index was calculated using equation (5):

Streif index = Firmness $(kg/cm^2)/$ [soluble solid content (Brix) × starch (1–6 scale)] (5)

Firmness values were calculated using penetrometer as described under 2.3.1.

Statistical analysis

Triplicate data obtained from destructive and non-destructive techniques was analysed using one-way analysis of variance (ANOVA) followed by Duncan's multiple range test (DMRT) to determine the statistical difference at $p \leq 0.05$ of measured parameters at different harvest dates within the exotic apple varieties. Student's *t*-test was employed to determine the significance of analyzed data for principal dimensions and colour coordinates measured using machine vision system and conventional methods. Standard statistical software, SPSS (version 16.0, USA) was used for data analysis. Pearson's correlation coefficient was also computed to determine the relationships between acoustic coefficient and various destructive parameters. In order to understand more about the relationship between the variables, principal component analysis (PCA) was performed on the physico-chemical properties analyzed at different harvest dates using Acoustic and destructive techniques by XLSTAT trial version (2019).

Results and discussions

Acoustic coefficient of exotic apple varieties at different harvest dates

The acoustic impulse response technique is a fast non-destructive measurement normally applied to measure the firmness and elastic behaviour of spherical fruits. Acoustic coefficient measured in this study was used to determine the firmness of apples harvested at different dates and the results showed that acoustic coefficient decrease significantly (p \leq 0.01) irrespective of variety (Fig. 2). *Gala Redlum* (V1), *Red Velox* (V2) and *Super Chief* (V3) recorded highest acoustic coefficient values of 21.13, 20.10 and 20.62, respectively, when harvested at H1. Softening of apples causes a loss in turgor during ripening and was manifested in

terms of low acoustic index produced by apples harvested at 3rd harvest dates. Acoustic signal is generated by the elastic behavior of the cell wall in case of fruit tissue and this signal decreases proportionally with the ripening of fruit (Toivonen and Brummell, 2008). Loosening of the glycan-cellulose backbone leads to swelling of cell wall and its weakening. This is accompanied by increase in porous structure, which in turn decreases the amplitude of acoustic signal within the fruit (Toivonen and Brummell, 2008). Since elastic waves are reflected on the border of the solid shaped fruits, increase in the intercellular spaces causes attenuation of waves with prolonged harvesting and a consequent decrease in the amount of acoustic signal produced. Zdunek et al. (2010) also reported a decrease in the number of acoustic signals from first to 10th day of shelf-life.

Effect of different harvesting dates on physico-chemical properties of apple varieties measured using destructive techniques.

The physico-chemical quality characteristics listed in (Table 1) showed significant ($p \le 0.05$) differences with respect to each harvest date. Starch iodine test, anthocyanin content, juice yield, TSS, total sugars, reducing sugars and non-reducing sugars showed increasing trend while as firmness, acidity and pectin content of different apple varieties showed decreasing trends with respect to prolonged harvesting dates.

Effect on firmness

Firmness is one of the texture attributes that indicates the internal quality associated with other attributes such as mealiness, crispness and hardness (Fatihzadeh et al., 2021). Firmness of apples harvested at different dates decreased significantly ($p \le 0.01$) irrespective of the variety. Highest firmness values of 20.72 lbs was recorded in variety V1 (*Gala Redlum*) followed by variety V2 (*Red Velox*) with a firmness value of 18.70 lbs and variety V3 (*Super Chief*) with a value of 17.17 lbs during the early harvest date. Cells loose water and cell walls become thinner with the advancement of ripening which leads to loss of firmness and is simultaneously accompanied by a large amount of degradation of the cell wall material and pectin. Yousef et al. (2016) reported that the fruit firmness decreased significantly during the harvesting period until the last harvest date in plum fruit.

Gala variety (V1) harvested at 110 DFFB; *Red Velox* (V2) and *Super Chief* (V3) harvested at 130 DFFB were having harder texture and did not develop the desired quality attributes. While, *Gala Redlum* harvested at 130 DFFB; *Red Velox* and *Super Chief* harvested at 150 DFFB were found



Fig. 2. Non-destructive acoustic coefficient of tested apple varieties with respect to harvest dates.

Variety	Firmness (.	lbs)		Starch io	dine rating		Anthocyan	in (mg/100 g	0	Pectin (%)			Juice Yield	(%)		Total solub	le solids (°Br	ix)
	H1	H2	H3	H1	H2	Н3	H1	H2	H3	H1	H2	H3	Η1	H2	H3	H1	H2	H3
۲Л	20.72^{aA}	17.90^{bA}	16.80^{cA}	1.52^{cA}	2.44 ^{bA}	3.72^{aA}	19.65 ^{cC}	21.22^{bC}	24.81^{aC}	0.50^{aA}	0.48^{aA}	$0.35^{\rm bA}$	63.58 ^{bB}	68.98 ^{aB}	65.75 ^{cB}	10.09^{aC}	11.21^{bB}	12.10^{cA}
	\pm 0.067	\pm 0.049	± 0.058	++	± 0.053	± 0.082	\pm 0.458	± 0.062	\pm 0.270	± 0.053	± 0.017	± 0.020	\pm 0.061	± 0.058	\pm 0.047	\pm 0.026	± 0.194	± 0.082
				0.082														
V2	18.70^{aB}	17.89^{bA}	16.38^{cA}	1.64^{cA}	$2.48^{\rm bA}$	3.72^{aA}	48.82^{cA}	50.30^{aA}	52.10^{aA}	0.50^{aA}	0.43^{bB}	0.30^{cA}	64.35^{aA}	69.34 ^{bA}	67.45^{aA}	11.34 ^{bA}	12.36^{aA}	12.52^{aA}
	± 0.053	± 0.053	± 0.051	+1	± 0.082	± 0.071	± 0.036	± 0.156	± 0.250	± 0.082	± 0.031	± 0.022	\pm 0.073	± 1.075	± 0.053	± 0.042	± 0.194	± 0.022
				0.053														
V3	17.17^{aC}	16.65^{bB}	15.92^{cB}	1.58^{cA}	2.50^{bA}	3.66^{aA}	21.80^{cB}	24.60^{B}	26.70^{aB}	0.46^{aA}	0.42^{bB}	0.29^{bA}	61.35^{cB}	68.25 ^{bA}	65.54^{aB}	10.48^{cB}	11.20^{bB}	11.72^{aB}
	± 0.031	± 0.040	± 0.053	++	± 0.082	\pm 0.011	± 0.077	± 0.072	± 0.310	± 0.042	\pm 0.041	± 0.036	± 0.042	± 1.072	± 0.081	± 0.051	± 0.152	± 0.082
				0.082														
r	0.723^{**}			-0.922*			-0.430^{*}			0.510^{*}			-0.496^{*}			-0.933^{**}		

Table 1

different uppercase supe significant at p < 0.05; ** significant at p < 0.01rows and value

1.0

suitable for table consumption. Apple firmness continuously decreased during the tested period, reaching a total firmness loss of 18.87% in V1, 12.35% in V2 and 7.33% in V3, respectively. The correlation coefficient of firmness with non-destructive acoustic coefficient was found to be 0.723 with a *p* value of \leq 0.01 indicating that firmness of fruit was positively correlated with acoustic coefficient. Both acoustic coefficient and firmness (measured by penetrometer) decreased in apples with delay in harvesting, thus acoustic index was an effective index to predict the firmness of apples harvested at different dates. Due to enzymatic activity, solubilization of pectin takes place which leads to disassembly of cell wall structure. Thus, with the advancement in harvesting time, mechanical strength of cell wall gets reduced leading to a simultaneous reduction in fruit firmness and acoustic co-efficient. Shmulevich et al. (2003) reported positive correlation between the acoustic fruit response and the firmness or softening of several fruits such as mango, melon, avocado, apple and tomato.

Effect on starch iodine rating

Starch iodine index records the degree of starch disappearance in the flesh, as starch concentration decreases with the progress in maturation and ripening (Farcuh, 2021). Starch index values for Gala (V1) increased significantly from 1.52 to around 3.72 when harvested at 110 DFFB and at 130 DFFB, respectively (Table 1). Likewise, for Red Velox (V2) and Super Chief (V3), starch iodine rating increased from 1.64 to 3.72 and from 1.58 to 3.66 with the advancement in harvesting date from 130 DFFB to 150 DFFB, respectively (Table 1). Therefore, delayed harvesting reduced the starch content of the apples, which is reflected by higher starch iodine rating. The decrease in starch content may be attributed to the conversion of starch to soluble sugars with the progress of time. Subedi et al. (2018) reported that higher starch index of 4.90 was recorded in Royal Delicious apples immediately after harvest. Nondestructive acoustic coefficient showed highly significant ($p \leq 0.01$) negative correlation with starch index (r = -0.922). Starch content decreases in apples towards delayed harvesting by breakdown of starch into simple sugars. This increases the starch iodine rating, which is also evident by significant positive correlation between starch index rating and total sugars (r = 0.636, $p \le 0.01$, Table S1). Total sugars also recorded a significant negative correlation with acoustic coefficient (r = $-0.520p \le 0.01$, Table S1).

Effect on anthocyanins

Anthocyanins are the pigments that impart the dark color to the apple peels. Anthocyanin content and colour development, continuously and significantly increased during fruit maturation reaching its maximum at the 3rd harvest i.e., 130 DFFB for V1 and 150 DFFB for V2 & V3 (Table 1). Apple varieties harvested earlier at H1 showed less colour development than the ones harvested latter. Anthocyanin content was recorded highest in Red Velox (V2) (52.10 mg/100 g) followed by Super Chief (V3) (26.70 mg/100 g) and Gala (V1) (24.81 mg/100 g) at the last harvesting date (H3) (Table 1) in each variety. The reason behind the highest anthocyanin content in late harvested apples might be due to full color development by associated enzyme phenylalanine ammonia-lyase (PAL) and the low anthocyanin content in early harvested apple might be due to more chlorophyll content than anthocyanins (Dar et al., 2019). Yousef et al. (2016) reported significant differences in anthocyanins content of the plum fruit at the different harvest dates. The correlation coefficient of anthocyanins with nondestructive acoustic coefficient was found to be -0.430 with a p value of \leq 0.04 indicating that anthocyanins were negatively correlated with acoustic coefficient.

Effect on pectin content

Softening of apples has been highly associated with the modification of cell wall materials. Pectin content decreased in all the tested varieties with delay in harvesting time, however, the decrease recorded in V1 from H1 to H2 and V3 from H2 to H3 showed non-significant variation

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(Table 1). However, pectin content of V2 showed a significant (p < 0.05) variation for all the three harvest dates. Highest pectin content was recorded at the first harvest date (H1) in all the varieties in the order of Gala > Red Velox > Super Chief. Since pectin development is directly related to firmness in apples, Gala Redlum with highest firmness values also recorded highest pectin content of 0.50% (Table 1). As the firmness of fruits was found to decrease with harvest dates, the pectin content also showed a concomitant decrease. Robert et al. (2008) reported a slight dip in the pectin content of chicory roots from first to mid harvesting dates. It has been postulated that pectin is stabilized due to presence of inter and intra molecular bonds between arabinogalactan side chains. The hydrolysis of these inter-polymeric cross chains affects the cell wall integrity and pectin yield. Pectin content was found to be positively correlated with acoustic coefficient (r = 0.510 and p \leq 0.05), because fruits high in pectin content showed higher firmness values (r = 0.856 and p < 0.01), and recorded higher acoustic coefficient as well.

Effect on juice yield

Different harvesting stages showed a significant (p \leq 0.05) effect on the percent juice content of tested apple varieties. The maximum juice content was recorded in Red Velox (V2) (69.34 %) followed by Gala Redlum (V1) (68.98%) and Super Chief (V3) (68.25%), when harvested at second harvest date (H2) specific to each variety. It was observed that juice yield increased significantly (p \leq 0.5) from H1 to H2 and then showed a significant decline from H2 to H3 in all the tested varieties (Table 2). The endogenous fruit enzymes cause the breakdown of insoluble polysaccharides into more soluble form which leads to higher juice yield (Toy et al., 2022). In the present study, the juice content was lower in initial observation as compared to the later dates which was also reported by Chalise and Giri (2019) in case of Royal Delicious Apple. The authors reported that total increment in the juice content of fruit from first harvest to the final harvest was 9.25%. The correlation coefficient of juice yield with non-destructive acoustic coefficient was found to be -0.496 with a *p* value of ≤ 0.05 indicating that anthocyanins were negatively correlated with acoustic coefficient (Table 2).

Effect on total soluble solids

Total soluble solids (TSS) of apples and other fruits is an important quality factor which determines the fruit taste. TSS of apple is a major quality parameter which is correlated with texture and composition. TSS content of apple fruit was affected by different harvest dates (Table 2). Results depicted in (Table 2) revealed that TSS increased significantly ($p \le 0.05$) with the advancement in harvest dates in case of V1 and V3. Variety 2 (*Red Velox*) showed highest TSS followed by variety 1 (*Gala Redlum*) and variety 3 (*Super Chief*) when harvested at last date i.e., 130 DFFB for V1 & 150 DFFB for V2 & V3. The increase in TSS could be

attributed to the breakdown of starch and complex polysaccharides into simple sugars (Kviklienė and Valiuškaitė, 2009). Increase in total sugars observed from H1 to H3, in this study is likely a possible reason for the increase in TSS of apple varieties from initial to last harvest date. This is also supported by the significant positive correlation of Total soluble solids recorded with total sugars (r = 0.925, $p \le 0.01$, Table S1). Since, TSS percentage is a function of total dissolved solids and moisture content of the fruit, the increase in TSS could be also due to accumulation of soluble solids and moisture loss. These results are in accordance with the finding of Crisosto, (1994) who reported that soluble solids content (SSC) increases fruit maturation and ripening in plum fruit. Bhat et al. (2012) also reported that TSS content of the fruits increased with the delayed harvesting. The correlation coefficient of total soluble solids with non-destructive acoustic coefficient was found to be -0.933 with a p value of < 0.01 which indicated highly significant negative correlation between the two. Since TSS showed a negative relationship with firmness (r = -0.600, p \leq 0.01, Table S1), higher TSS of fruits harvested at last date produced less acoustic signal as these fruits were less firm.

Effect on titratable acidity

Fruit acidity plays a significant role in consumer acceptance and end use. There was significant ($p \le 0.01$) difference in titratable acidity of apple varieties harvested at different dates (Table 2). The highest content of titratable acidity was found in H1 followed by H2 and H3 irrespective of the variety. The acidity was found highest in V2 (*Red Velox*) followed by V1 (*Gala Redlum*) and V3 (*Super Chief*). Acidity of fruits decreased significantly ($p \le 0.01$) with delayed harvesting. Fruits of third harvest recorded significantly lower acid content as compared to fruits of first harvest which is also supported by the findings of Bhat et al. (2012) in case of pear. Ghafir. (2009) reported that fruit acidity was significantly affected by rate of respiration which consumed organic acid and thus decline acidity. The correlation coefficient of titratable acidity with non-destructive acoustic coefficient was found to be non-significant (Table 2).

Effect on total, reducing and non-reducing sugars

Starch accumulates at very early stages of fruit development in apples and with the advancement of maturity, the accumulated starch is hydrolyzed into sugars. The results depicted in Table 2 showed that total and reducing sugars increased significantly ($p \leq 0.05$), with the advancement in harvest dates from H1 to H3 reaching a maximum concentration at last harvest date (H3). However, non-reducing sugars showed a decreasing trend for all the harvest dates irrespective of the varieties. Total and reducing sugars were found to be highest in variety 2, whereas non-reducing sugars were high in variety 1 (Table 2). Total, and reducing increased from 9.42 to 9.72 % and 8.43 to 9.13% in *Red*

Table 2

Effect of different harvest dates on acidity, total sugars, reducing sugars, non-reducing sugars and overall acceptability of apple varieties and its correlation with acoustic coefficient.

Variety	Acidity (% malic acid)			Total sug	Total sugars (%)			Reducing sugars (%)			Non-reducing sugars (%)			Overall acceptability		
	H1	H2	H3	H1	H2	Н 3	H1	H2	H3	H1	H2	H3	H1	H2	H3	
V1	$0.25^{ m aA}$ \pm	0.24^{bA} \pm	$0.22^{ m cA}$	$8.50^{ m cC}$ \pm	$8.62^{ m bC}$	8.75 ^{aC} ±	7.42 ^{cC} ±	7.77 ^{bC} ±	$8.21^{ m aC}$	$1.13^{ m cA}$	$0.85^{ m bA}$ \pm	0.54^{aA}	7.42 ^{cB} 土	$8.30^{ m bA}$ \pm	8.72^{aA} \pm	
V2	0.004 0.26 ^{aA}	$0.004 \\ 0.25^{bA}$	0.004 0.23 ^{cA}	0.022 9.42 ^{cA}	0.025 9.57 ^{bA}	0.028 9.72 ^{aA}	0.058 8.43 ^{cA}	$0.042 \\ 8.78^{bA}$	0.038 9.13 ^{aA}	0.024 0.99 ^{cB}	0.018 0.79 ^{bA}	0.032 0.59 ^{aA}	0.192 7.24 ^{cA}	$0.288 \\ 8.09^{\mathrm{bB}}$	0.020 8.44 ^{aB}	
V3	$^{\pm}$ 0.004 0.22 ^{aB}	± 0.004 0.21 ^{aB}	$^{\pm}$ 0.008 0.19 ^{aB}	$^{\pm}$ 0.024 8.92 ^{cB}	$^{\pm}$ 0.021 9.05 $^{ m bB}$	± 0.032 9.32 ^{aB}	± 0.062 7.97 ^{cB}	$^{\pm}$ 0.024 8.30 ^{bB}	± 0.022 8.77 ^{aB}	± 0.051 0.95 ^{cB}	$^{\pm}$ 0.017 0.75 ^{bA}	$^{\pm}$ 0.029 0.55 ^{aA}	$^{\pm}$ 0.100 7.21 ^{bA}	$^{\pm}$ 0.067 8.35 ^{aA}	± 0.033 8.40 ^{aB}	
	± 0.004	± 0.004	± 0.008	± 0.042	± 0.313	± 0.035	± 0.040	\pm 0.011	± 0.042	± 0.042	± 0.016	± 0.029	± 0.082	± 0.181	± 0.223	
r	0.266 ^{NS}			-0.520*	-0.520*			-0.769**			-0.895**			-0.808**		

V1: *Gala Redlum*, V2: *Red Velox* V3: *Super Chief*; H1: First harvest date, H2: Second harvest date, H3: Third harvest date, r = Correlation coefficient of acoustic index, Values with different lower case superscripts (a, b, c) within rows and value with different uppercase superscripts (A,B,C) withing columns differ significantly at 5 % level of significance.

NS: non-significant, *significant at p < 0.05; ** significant at p < 0.01.

Velox (V2) from H1 to H3. Non-reducing sugars decreased from 1.13 to 0.54 % in *Gala Redlum* (V1) from H1 to H3 (Table 2). Bhat et al. (2012) showed the increase in total and reducing sugars with increasing harvest dates which may be attributed to the hydrolysis of polysaccharides to monosaccharides and concentration of juice as a result of moisture loss through transpiration. The correlation coefficient of total sugar, reducing sugar and non-reducing sugar with non-destructive acoustic coefficient was found to be -0.520 ($p \le 0.05$), -0.769 ($p \le 0.01$) and -0.895 ($p \le 0.01$), respectively indicating the negative correlation between these parameters.

Effect on sensory evaluation

Apple varieties were evaluated for sensory attributes immediately after harvest in order to perceive the consumer acceptance among the samples harvested at different dates. Overall acceptability (OA) increased significantly in Gala Redlum (V1) and Red Velox (V2), however, overall acceptability of Super Chief (V3) showed a non-significant increase with the advancement in harvest dates (Table 2). Highest overall acceptability was recorded at the last harvest date in all of the tested varieties. OA scores of 8.72, 8.44 and 8.40 were received by V1, V2 and V3 at H3 respectively (Table 2). Since the total soluble solids and total sugars were recorded highest at last harvest date, which might have enhanced the sensory score of the varieties harvested later than the once harvested earlier. Acoustic coefficient showed a highly significant (p <0.01) negative correlation with overall acceptability of apple fruits harvested at different dates (r = -0.808, Table 2). This implied that apples with lowest acoustic firmness received high scores for sensory descriptors of crispness, tenderness, juiciness, mealiness, and appearance and vice-versa for fruits that recorded higher acoustic coefficient. Acidity in apples harvested at last date was lower as compared to apples harvested earlier, which was in relation to the high overall acceptability score and lower acoustic coefficient recorded by such fruits. Likewise, high OA in apples harvested later might be accorded to the higher sugar accumulation in apples towards delayed harvesting, which is also evident by positive correlation of total sugars with overall acceptability (r = 0.472, p < 0.01, Table S1).

Evaluation of principle dimensions and color coordinates by machine vision and conventional method

Length, width and thickness of apples measured using digital vernier caliper and machine vision

The physical properties of fruit are the fundamental and basic parameters required during the designing of graders, peeler, de-seeder, pricking, cutting, packaging machines, and so on (Singh et al., 2019). Fig. 3 (a-c) displays the statistics of the outer diameters (length, width and thickness) of apples measured manually using the vernier calipers as well as by machine vision system. It is evident that the statistics of outer diameters of fruits of all the cultivars measured by former method showed non-significant difference in comparison to the latter one. The length, width and thickness of *Super Chief* (V3) was found to be more followed by *Red Velox* (V2) and *Gala Redlum* (V1). This result is in good agreement with that of Chalidabhongse et al. (2006) who reported that mango lengths and widths measured using the image processing method are higher than those obtained manually using a Vernier caliper. Thus, the machine vision-based measurement method could be reliable and accurate for measuring the fruit size with high speed and convenience.

Colour attributes (L*, a*, b*) of apples measured using hunter color lab and machine vision

The changes in skin colour of apples are usually caused by chlorophyll degradation. During fruit development and maturation, both chlorophyll breakdown and synthesis are observed, but following maturation and ripening processes, decrease in the chlorophyll content is noticed, which results in decreasing intensity of green coloration (Rutkowski et al., 2008). The results depicted in Fig. 4 (a-c) indicated

that L* and b* values were highest in harvest 1, whereas, a* was highest in harvest 3. The a* coordinate reflects the change from green to red coloration as a consequence of the disintegration of chlorophyll and the unmasking of xanthophyll. Therefore, measurement of the a* coordinate can provide a basis for determining the harvest date. Higher redness (a*) observed at last harvest date in all the tested varieties was also accorded to the highest anthocyanin content recorded at the last harvest date (Table 1). The difference between colour coordinates measured using Hunter Colorimeter and Machine Vision system were non-significant, indicating the reliability and robustness of machine vision over conventional colorimeter based method. Generally, the red colour is preferred, even if the dark red is becoming less popular. New alternative colours are recently catching consumer attention in many fruits, therefore, breeding interventions in this direction are also demanded (Telias et al., 2011). In terms of consumer acceptability, apples with higher redness are considered more appealing. Delwiche and Baumgardner (1983) recognized changes in the a* coordinate to be a very good indicator of harvest maturity in peaches.

Standardization of optimum harvest date

Streif index (SI) is considered a reliable parameter to determine the harvesting date (Musacchi and Serra, 2018) as it takes into account fruit firmness, total soluble solid and starch rating. Various researchers (Guz et al., 2017, Overbeck et al., 2013) have determined the optimum harvesting dates for different apple varieties based on the SI and reported that with the advancement in maturity SI decreases. Table S2 showed that with the increase in harvesting date from H1 to H3 SI decreased from 0.047 to 0.026 in Gala Redlum (V1), 0.070 to 0.024 in Red Velox (V2) and 0.072 to 0.026 in Super Chief (V3). Based on the different quality attributes SI range of 0.046 to 0.026; 0.041 to 0.024 and 0.041 to 0.026 were presumed to be optimal ranges for harvesting Gala Redlum (V1), Red Velox and Super Chief apples grown under temperate conditions of Kashmir in North Western Himalayas when intended for fresh consumption. Icka and Damo (2014) also reported that Streif index decreased from 0.35 to 0.04 when harvesting was prolonged from first to third harvesting date in apples. Icka and Damo (2014) recommended SI range of 0.075 to 0.10 for optimum harvesting of Red Delicious apples in Korca region.

TSS/ Acid ratio is also considered as a reliable index for determining the optimum harvesting date of apples as it indicates the trend of fruit ripening (Icka and Damo 2014). Results depicted in (Table S2) showed that TSS/ acid ratio increased with the delay in harvest dates. Highest TSS/acid ratio was recorded in the fruits harvested at third harvest date. (Table S2) showed that the TSS/ Acid ratio ranged from 41.00 to 54.00 for *Gala Redlum* (V1), 44.29 to 53.50 for *Red Velox* (V2) and from 48.51 to 59.19 for *Super Chief* (V3) with advancement in harvest dates from H1 to H3, respectively. Icka and Damo (2014) recommended TSS/acid ratio range between 40 and –50 for Red Delicious in Korça region. Bhat et al. (2012) also reported increment in total soluble solids/acid ratio of Pear with delayed harvesting. TSS/acid ratio recorded for exotic apple varieties in this study are comparable with the findings of Icka and Damo (2014) for Red Delicious apple.

Ripeness at harvest is one of the primary factors affecting apple quality as high-quality apples are those that will ensure consumer satisfaction according to organoleptic preferences. Highest overall acceptability scores of 8.72, 8.44 and 8.40 were recorded in *Gala Redlum* (V1), *Red Velox* (V2) and *Super Chief* (V3), respectively at third harvest date.

Therefore, on the basis of lowest SI, optimum TSS/acid ratio, highest overall acceptability, third harvest date i.e., 130 DFFB for *Gala Redlum* (V1), 150 DFFB for *Red Velox* (V2) and *Super Chief* (V3) were identified as optimal harvest dates for fresh/table consumption of exotic apple varieties tested in this study.







b)



Fig. 3. (a-c) Length, width and thickness of apple varieties measured using vernier callipers (VC) and machine vision (MV) harvested at different dates (H1, H2 and H3).







b)



Fig. 4. (a-c) Colour coordinates (L*, a* and b*) of apple varieties using Hunter colour meter and machine vision (MV) harvested at different dates (H1, H2 and H3).

Principal component analysis model

PCA was performed to evaluate the maturity characteristics of exotic apple varieties at different harvesting dates to provide an easy visualization of the complete data set in a reduced dimension. Among the maturity and quality characteristics, acoustic coefficient, firmness, starch iodine rating, anthocyanin, pectin, juice yield, total soluble solids, acidity, total sugars, reducing, non-reducing sugars, principal dimensions (L, W and T), color coordinates (L*, a* and b*) were considered to characterize the tested varieties harvested at three different harvest dates (Fig. S1). Principal components 1 (PC1) and 2 (PC2), accounted for 78.5% and 18.7% of total variability in Gala Redlum (V1) (Fig. S1a); 84.6% and 12.8% in Red Velox (V2) (Fig. S1b) and 78.7% and 16.1% in Super Chief (V3) (Fig. S1c), respectively. Exotic apple varieties showed a sequential distribution over PC1, leftwards from the first (H1) to the last (H3) harvest. First/early harvesting date (H1) was clearly separated from late harvesting date (H3): the first one was located to the left of the PC1 axis and the last one to the right of the graph, however the mid harvesting date (H2) was coinciding exactly on the axis of PC1. Although the distribution pattern was gradual, the mid harvest date showed more similar behaviour, in terms of the different quality parameters studied.

Acoustic coefficient, firmness, TSS, total sugars, reducing sugars and overall acceptability contributed 5.03%, 5.13%, 5.27%, 5.19%, 5.09% and 5.02%, respectively to dimension 1 whereas juice yield contributed 20.41% variation to dimension 2 in *Gala Redlum* (V1) (Table S3). The parameters such as acoustic coefficient, firmness, starch iodine, anthocyanin, reducing sugars and non-reducing sugars contributed 4.90%, 4.15%, 4.33%, 4.60%, 4.87% and 4.57%, respectively variation to dimension 1 whereas TSS, juice yield, total sugars, pectin and overall acceptability accounted for 8.03%, 9.03%, 5.70%, 5.25% and 5.89%, respectively variation to dimension 2 in *Red Velox* (V2) (Table S3). However, in *Super Chief* (V3) parameters such as acoustic coefficient, TSS, anthocyanin, non-reducing sugars and overall acceptability contributed 5.07%, 5.05%, 5.14%, 5.14% and 4.83% to dimension 1, respectively, however, juice yield and total sugar contributed 12.08% and 5.15% variability to dimension 2, respectively (Table S3).

Apple varieties harvested latter at last harvest date (H3) correlated positively with starch iodine rating, anthocyanin content, total soluble solids, total sugars, reducing, non-reducing sugars, length, width, thickness, b* (yellowness) and overall acceptability and negatively with acoustic coefficient, firmness, pectin, acidity, L* (brightness) and a* (redness). Better sensory score of late harvested varieties could be attributed to sweetness in taste and better colour that is reported to influence the consumer apple purchasing patterns. In contrast, varieties harvested earlier (H1) presented higher firmness, acidity, and acoustic coefficient values and lower overall acceptability score, which decreased subsequently with advancing ripeness.

Conclusion

In the present work, non-destructive techniques i.e. acoustic and machine vision were explored to predict optimum harvesting dates for different exotic apple varieties (*Gala Redlum, Red Velox and Super Cheif*) grown under temperate conditions of North West Himalayas. Acoustic coefficient irrespective of variety showed a decreasing trend with the increase in harvesting date. Firmness and acidity of all the three varieties decreased, whereas, starch iodine rating, anthocyanin content, total soluble solids (TSS), total sugar and reducing sugars showed increasing trends with the increase in harvesting dates. Overall acceptability also increased significantly with the increase in harvesting date. Based on the acoustic results and correlation analysis it was presumed that acoustic technique could be a reliable indicator for determination of optimum harvesting dates. Colour coordinates and dimensional parameters measured through machine vision were at par with the values obtained through hunter colorimeter and vernier caliper. Therefore, machine vision can be successfully used in place of hunter colorimeter and Vernier caliper to measure colour and dimensional properties quickly and accurately. Overall inference drawn from destructive and non-destructive quality evaluation results was that, for fresh consumption 130 DFFB is the suitable harvesting date for *Gala Redlum* and 150 DFFB is the suitable harvesting date for *Red Velox* and *Super Chief*.

CRediT authorship contribution statement

Nazrana Rafique Wani: Investigation, Writing – original draft. Syed Zameer Hussain: Conceptualization, Supervision. Gopinath Bej: Software, Methodology. Bazila Naseer: Writing – review & editing. Mushtaq Beigh: Resources. Ufaq Fayaz: Methodology. Tamal Dey: Software. Abhra Pal: Software. Amitava Akuli: Software. Alokesh Ghosh: Validation. B.S. Dhekale: Formal analysis. Fehim J. Wani: Data curation.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.fochx.2023.100754.

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