Effect of individual vacuum and modified atmosphere packaging on shelf life of guava

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Abstract
The physico-chemical changes occurring continuously in climacteric fruit guava, leads to heavy post harvest losses. Therefore the present study was conducted to evaluate the effect of individual packaging through Vacuum and Modified atmosphere packaging on shelf life of guava. Fruits were individually packed in polythene bags (LDPE) of 200 gauge thickness by Vacuum packaging and modified atmosphere packaging and stored at 7±3°C. Both individual packaging methods were equally effective in reducing the biochemical changes during storage. The decrease in flesh firmness of the fruits was rapid and progressive in unwrapped control fruits but in individually packed fruits, the decrease was slower. Individually wrapped fruits retained higher ascorbic acid and acidity. Vacuum packing and MA though showed minimum PLW and ripening but suffered from the drawback that fruits remained hard and unripened and scored low organoleptically.

Keywords: Vacuum packaging, modified atmosphere packaging, individual packaging, shelf life, Psidium guajava

Introduction
Guava (Psidium guajava L.) is a productive and remunerative crop grown commercially in sub-tropical and tropical regions. It is a delicious and highly perishable fruit known for its delightful taste, flavour, moderate price in market and high nutritional status. In India 1.60 lakh hectare of agricultural land is under cultivation of guava with yield of 1.85 million tones (Panwar, 2008) [1].

Guava fruit is an excellent source of vitamin C, contains about 17% dry matter and 80% moisture. Fruit has an appreciable amount of minerals such as phosphorus, calcium, iron as well as vitamins like niacin, pentothenic acid, thiamin, riboflavin and vitamin A. It excels most other food crops in productivity, hardiness and adaptability of the fruit (Singh et al., 1993) [2]. The physiological and biochemical changes continuously occur after harvest till fruit become unfit for consumption. Like other high moisture fruits, guava too exhibit short shelf life. One way to extend the shelf life of fruits and vegetable is their proper packaging. Now a day’s individual wrapping of fresh fruits and vegetable has gained widespread interest. Individual wrapping of fruits have potential benefit over traditional method of packaging as additional protective covering prevent bruising or any physical damage in the pack during handling. It creates a modified atmosphere around the fruit which promote the additional benefits of individual packaging i.e., reduction in weight loss, maintenance of firmness, reduction in deformation, alleviation of chilling injury, reduction of decay from secondary infection, delay in colour development and senescence (Risse, 2001) [3].

Different types of packaging varying in packaging material and the method of their execution like cling wrap, shrink wrap, vacuum packaging and modified atmosphere packaging are available. Rana et al., (2015) [4] reported guava fruits wrapped in cling film and stored at 7 °C had a shelf life of 21 days as compared to control (unwrapped fruits). Vacuum packaging of fresh commodities involves eliminating the air in package using a suction machine. Vacuum packaging offers an extensive barrier against corrosion, oxidation, moisture, drying out, dirt, electric charge, ultra violet rays, mechanical damages, fungus growth or perishability etc, as product is sealed hermetically in a plastic bag. This method reduces the level of O₂ and N₂ in the package and thus prolongs the shelf life of fruits (Tapia et al., 2003) [5]. Sudha et al., (2007) [6] investigated the influence of packaging treatments on physical and biochemical characteristics of sapota fruits and reported reduced respiratory rate and reduction in PLW.
Modified atmosphere conditions are created inside the package by the commodity itself or active modification. Commodity generated or passive modified atmosphere is evolved as a consequence of commodity’s respiration. The generated modification in atmosphere around the fruits retards the metabolism, slow down the rate of ethylene synthesis, reduces microbial contamination and less moisture loss with better quality retention. Modified atmosphere packaging maintains freshness and extends the shelf life of fresh produce by utilizing basic principles of respiration and permeation simultaneously. Broccoli packaged in PVC films lead to the accumulation of 10% CO₂ within the package, which helped in retention of turgidity and moisture significantly better than non packaged controls.

Keeping in view the importance of individual wrapping of fruits, the present investigation was carried out to study the effect of individual film packaging through vacuum and modified atmosphere packaging of guava fruits on its shelf life.

Material and Method
Harvesting time and methodology used
Plants of guava cv. Hisar Safeda were selected from an orchard at Hisar from the summer season crop. Uniform and healthy fruits were harvested at green mature stage with the help of sharp secateur, leaving a small pedicel on the fruit. Injured and diseased fruits were discarded.

Treatments
Fruits were individually packed in polythene bags (LDPE). For vacuum pack, fruits were packed in sealed polybags of 200 gauge thickness by vacuum packaging machine. In case of modified atmosphere pack, fruits were sealed in polybags individually while control fruits were kept unwrapped. There were 10 fruits per pack and six replicates per treatment. Each pack served as one replicate. The fruits were stored at low temperature (7±3 °C). The sampling was done at 2 days interval up to 21 days of storage.

Physiological Observations
Physiological loss in weight (PLW%), Ripening% and Flesh firmness
The initial and final weight of fruit pack was recorded at the beginning of storage period and on each date of observation respectively. The % loss in weight is expressed in terms of PLW%. Color of fruit was taken as criterion to record ripening. The fruit which turned from green to yellow were regarded as ripened and the intensity of the yellow colour to green was used to determine ripening%. Flesh firmness was measured by pressure tester fitted with a cylindrical plunger of 4mm diameter. Fruit firmness was measured at two sites each on opposite sides. Firmness of three fruits per treatment was measured on each day of observation expressed as kg/cm².

Respiration Rate
Respiration rate of packaged guava fruits was measured with MAP testing machine (M/s Quantek USA, Model 902D). After removal from the pack fruits were kept in respiration jars for 2 hours and O₂ and CO₂ concentrations was measured by injecting the needle of Map testing machine in jars. Results were expressed as μ mole of CO₂ evolved per hour per kilogram of fruit.

Hunter Colour Value
Colour measurement was carried out using a Hunter colorimeter D25 optical sensor (Hunter Associates Laboratory, Trestoa, VA, USA) on the basis of three variables (L, a, b value). The L, a and b value signifies the lightness (100 for white and 0 for black), greenness and redness (-80 for green and 80 for red), blueness to yellowness (-80 for blue and 80 for yellow) respectively. Each sample was replicated thrice and the average values were used in analysis.

Biochemical Parameters
Phenols
Total phenols were estimated by the method of Amorium et al. (1977) [7] using Folin-Ciocalteau reagent. Two g of seed free pulp was extracted with 20 ml of 80% acetone solution. Aliquot obtained was diluted five times with distilled water. To 0.5 ml of aliquot, distilled water (8.5 ml) and folin reagent (0.5 ml) was added. After 3 minutes, 1 ml of 20% Na₂CO₃ was added vigorously and allowed to stand for 30 minutes. The optical density was read for 750 nm against reagent blank. Standard curve was prepared using graded concentration of tannic acid.

Total Soluble Solids (TSS)
The fruit pulp was crushed for juice extraction. The juice of macerated pulp was squeezed by hand through muslin cloth. The juice was immediately utilized for determination of total soluble solids by using Abbe’s hand refractometer of 0-32% range at room temperature. The values were expressed as% total soluble solids of fruit.

Acidity
Acidity was determined by the titrimetric method described by A.O.A.C. (1995) [8]. Five gm of fruit pulp was finely crushed and diluted with 20 ml of distilled water. A suitable aliquot was titrated against 0.5N NaOH using 1% phenolphthalein as indicator. The end point was appearance of pink colour. The result was expressed as g of citric acid per 100gm of fruit pulp.

Ascorbic Acid
The ascorbic acid was determined by the method as described in A.O.A.C. (1995) [8]. 5g of fruit pulp was macerated with pestle in a glass mortar with 5 ml of 3% metaphosphoric acid and filtered through double layer of muslin cloth. Final volume was made to 10 ml with 3% metaphosphoric acid. 5 ml of the aliquot was titrated against 2, 6-dichlorophenol indophenol dye till light pink colour appeared. Results were expressed in mg of ascobic acid per 100g of fresh pulp weight.

Organoleptic Evaluation
Guava samples were evaluated by a panel of 9 semi-trained members for taste, appearance, texture, flavour and overall acceptability on 9 point hedonic scale basis.

Statistical Analysis
The data obtained in the present investigation were subjected to statistical analysis of variance (ANOVA) technique using two factorial completely randomized design, except for parameters PLW & ripening%, where single factorial CRD was used. This critical difference value at 5% levels of significance was calculated and used for making comparison among different treatments during storage.
Results and Discussion

Physiological loss in weight (PLW %)
The physiological loss in weight increased significantly with increased duration of storage. However, the magnitude of losses was much lower due to low temperature. PLW in control fruits was 26.5% and in individually wrapped fruits, it ranged from 3.0-3.2% (Table 1). Progressive increase in PLW during storage may be due to high transpirational and respiratory substrates losses. The higher respiration and evapo-transpiration rates in control fruits because of their direct contact with atmosphere could be the cause of higher PLW in control fruits. Reduction in losses at low temperature may be due to reduced metabolic activities and evapo-transpirational losses. An increase in PLW of guava fruits during storage has also been observed by Kumar et al., (2000) [9].

Individual packaging reduced the PLW significantly. At low temperature, both packaging treatments were equally effective in reducing PLW, as non-significant differences were found among these wrapping methods. Marked reduction in PLW in individually packed fruits may be due to reason that the films used for wrapping adhered to surface of fruit and formed an extra covering, which reduced respiration and prevented evapo-transpirational losses. Remarkable reduction in PLW by individual packaging of fruit in polythene films had also been reported by Sandhu et al., (2000) [10] and Nath et al., (2012) [11] in peach and Kinnow fruits, respectively.

Ripening

The ripening of guava fruits increased progressively with increase in period of storage at lower magnitude. The control fruits attained 30% ripening and in treatments also, ripening was in the range of 10-15% by 21st day of storage at low temperature storage conditions (Data not shown). Progressive increase in ripening with increased period of storage in guava has also been reported by Rana et al., (2015) [4].

Individual wrapping delayed the ripening of fruits significantly. Among packaging treatments, vacuum packed fruits and modified atmosphere packed fruits showed no ripening. However, the control fruits showed only 30% ripening by 21st day of storage. Individual wrapping created the modified atmosphere around the fruit, causing high concentration of CO₂, which interfere the ripening action of ethylene during storage, delayed the chlorophyll degradation and thus retarded ripening of fruits. Remarkable reduction in ripening in individually wrapped gava fruits by LDPE of thickness 150 gauge has also been reported by Sahoo et al., (2015) [12]. Similarly, Hailu et al., (2014) [13] also reported maximum retention of greenness for 30 days in banana fruits packed in 300 gauge LDPE bags at ambient conditions.

Flesh Firmness

The firmness of fruits decreased with increasing period of storage as shown in figure 1. By 21st day of storage, the firmness in control fruits was 6.2 Kg.cm⁻² and in individually wrapped fruits, was 8 Kg.cm⁻². The decrease in firmness of guava during storage was primarily due to the loss of moisture from the surface causing the cells to lose turgidity and breakdown of pectin leading to the degradative changes in cell wall structure and composition. At low temperature, reduced metabolic activities and reduced evapo-transpiration loss of water could be the reason of slower decrease in firmness. Decrease in firmness during storage is also evident due to natural process of ripening taking place in climacteric fruits during storage. Similar decrease in flesh firmness has also been reported in banana stored under ambient, zero energy cool chamber (in BOD) (Narayana et al., 2002) [14].

Individual wrapping reduced significantly the decrease in flesh firmness of fruits during storage (Fig. 1). Vacuum and modified atmosphere packing were equally effective in maintaining the firmness of fruits. Maximum firmness was retained in vacuum packed fruits (8.6 Kg.cm⁻²) followed by modified atmosphere (8.2 Kg.cm⁻²). However, the cling and shrink wrapped fruits showed non-significant differences with control fruits. High firmness in individually wrapped fruits is attributed to the development of atmospheric composition which is inhibitory to the ripening process (Zucolo et al. 2017) [15]. Similar, retention of firmness in shrink wrapped bell pepper in 150 gauge LDPE film stored at 27 °C and 65% RH as compared to control fruits has been reported by Sahoo et al., (2006) [16]. MA packed green banana retained more firmness and greenness during low temperature storage (13±1 °C, 90-95% RH) conditions, as compared to unwrapped, openly kept green banana (Kudachikar et al., 2007) [17].

![Fig 1: Effect of individual Vacuum and Modified atmosphere packaging (MA) on TSS% of guava fruits during storage at low temperature](image-url)
harvested at green mature stage. Decrease in respiration rate of control fruits may be due to post climacteric decline and skin dryness which possibly prevented the exchange of gases. Increase in respiration rate of guava fruit cvs. ‘L-49’ and ‘Allahabad Safeda’ during room temperature storage has also been reported by Patel et al., (1992) [18] and Selvaraj et al., (1998) [19], respectively.

Individual wrapping reduced the respiration rate significantly (Fig. 2). At low temperature, among wrapping methods similar trend of minimum respiration rate in vacuum followed by modified atmosphere packed, was observed. Polythene wrapping prevented the moisture loss, maintained the turgidity and modified the atmosphere around the fruit. Thus O₂ depletion and CO₂ accumulation in pack resulted in reduced respiration rate. Vacuum packed fruits had lowest respiration rate probably because of reduced O₂ concentration and removal of ethylene from the intercellular spaces. Reduced rate of respiration in passion fruits packed in Polythene terepthalate (PET) and stored at ambient (18-23 °C, 58-77% RH) conditions has also been reported by Patel et al., (2009) [20].

Bars followed by different letters are significantly different at P<0.05. Vertical bars represents SE (n=3).

**Fig 2:** Respiration activity in individually Vacuum packed and Modified atmosphere (MA) packed guava fruits stored at low temperature for 21 days

**Hunter Color Lab**

The Hunter color Lab values increased with increasing duration of storage (Fig. 3). The ‘L’ value of control fruit was maximum by 21st day of storage. However, individual wrapped fruits were showing much lower ‘L’ value. Similarly, ‘a’ and ‘b’ values of control fruits and wrapped fruits were maximum by 21st day of storage. However, values were lower than control fruits. The colour changes in guava were indicated by the increase in ‘L’ lightness value, as greenness of fruit decreased gradually. Increase in ‘L’ lightness value during storage period indicated that the lightness of product increased due to ripening. The increase in lightness is mainly due to conversion of dark green colour to light green or yellowish colour during the storage period. The ‘a’ and ‘b’ value showed increased yellowness and decreased greenness indicating ripening of fruit during storage. Thus yellowness index increased. Decrease in the Hunter Lab values of control fruits indicated darkening or decay loss whereas zero value of ‘a’ indicates greyness as the fruit start decaying. The decreased Hunter Lab value thus indicated over-ripening. Similar changes in Hunter Lab values during storage indicating ripening were obtained by Mangraj et al., (2006) [21] for sweet lime, orange and mango.

Individual wrapping slowed down the changes in Hunter Lab values significantly (Fig. 3). Minimum changes in Hunter Lab values were observed in vacuum packed fruits followed by modified atmosphere packed fruits. Changes in Hunter Lab values in both packaging were slow as compared to control. Minimum changes in Hunter Lab values in both wrappings indicated slower changes in greenness of fruit to yellowness, showing delayed ripening. Individual wrapping modified the environment around the fruit. Increased accumulation of CO₂ and reduction in level of O₂ resulted in reduced rate of respiration and thus overall metabolic rate (Porat et al., 2009) [22]. As a result, ripening of fruit increased only in control, while packed fruits showed retarded ripening during storage.

Bars followed by different letters are significantly different at P<0.05. Vertical bars represents SE (n=3).

**Fig 3:** Effect of individual Vacuum and Modified atmosphere packaging (MA) on TSS% of guava fruits during storage at low temperature
Total Soluble Solids
TSS of fruit increased with increasing period of storage. The control fruits had maximum TSS (10.7%) by 18th day of storage (Fig. 4). By 21st day of storage, it decreased to 9.8% in control whereas in individually wrapped fruits it was ranging from 9.9-4%. The general rise in TSS of fruits may be due to hydrolysis of polysaccharides i.e. conversion of starch into sugars by metabolic activities and loss of water from fruit surface. Slow rise in TSS at low temperature could be attributed to reduced metabolic activities and reduced transpirational losses. Similar rise in TSS of fruits with advancement of storage was reported in litchi (Garg et al., 2002) [23] and in peach (Singh et al., 2006) [24] stored at ambient (30-35 °C, 40-60% RH) and cold store (2-4 °C, 80-90% RH) conditions respectively.

Acidity
Acidity of fruits increased with increasing period of storage (Table 1). By 21st day of storage, at low temperature, control fruits had minimum (0.27%) acidity, whereas in individually wrapped fruits it ranged from 0.29-0.35%. The progressive decrease in acidity could be due to conversion of acids into sugars during ripening process. Relatively lower reduction in acidity at low temperature could be attributed to lower rate of ripening. Decrease in acid content with increasing period of storage was also observed in sapota (Jindal et al., 2005) [27] stored at room temperature.

Individual packaging decreased significantly the reduction in acidity of fruits during storage (Table 1). Vacuum packed fruits had highest acidity content followed by modified atmosphere packaging as compared to unwrapped control fruits. In the present investigation the observed effect of individual wrapping on acidity of fruits during storage are matched by the respective changes observed in kiwi fruit. Narayana et al., (2002) [14] observed minimum reduction in acidity of banana packed in 400 gauge unvented polythene LDPE bags stored at ambient and zero energy cool chamber conditions as compared to banana packed in vented polythene LDPE bags and control fruits.

Ascorbic Acid
By 21st day of storage, ascorbic acid content in control fruits was 19.4 mg/100g while in individually wrapped fruits it ranged from 29-31 mg/100g (Table 2). Decrease in ascorbic acid might be due to enzymatic oxidation of L-ascorbic acid to dehydroascorbic acid. Higher ascorbic acid retention at low temperature could be attributed to lower rate of physiological processes. Decrease in ascorbic acid with increasing duration of storage was also reported in bell pepper stored at 27 °C and

Table 1: Effect of individual wrapping on physiological loss in weight (%) and Acidity% at different storage period

<table>
<thead>
<tr>
<th>Period of storage (Days)</th>
<th>Control</th>
<th>Vacuum Pack</th>
<th>Modified atmosphere pack</th>
<th>Control</th>
<th>Vacuum Pack</th>
<th>Modified atmosphere pack</th>
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<td>0.38%</td>
<td>0.40%</td>
<td>0.5%</td>
<td>0.3%</td>
<td>0.49%</td>
</tr>
<tr>
<td>3</td>
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<td>0.3%</td>
<td>0.45%</td>
<td>0.5%</td>
<td>0.3%</td>
<td>0.49%</td>
</tr>
<tr>
<td>6</td>
<td>0.9%</td>
<td>0.7%</td>
<td>0.4%</td>
<td>0.5%</td>
<td>0.3%</td>
<td>0.49%</td>
</tr>
<tr>
<td>9</td>
<td>1.4%</td>
<td>1.3%</td>
<td>0.4%</td>
<td>0.5%</td>
<td>0.3%</td>
<td>0.49%</td>
</tr>
<tr>
<td>12</td>
<td>1.7%</td>
<td>1.8%</td>
<td>0.37%</td>
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<td>0.3%</td>
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<td>2.4%</td>
<td>0.5%</td>
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<tr>
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<td>2.8%</td>
<td>0.24%</td>
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</tr>
<tr>
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<td>3%</td>
<td>3.2%</td>
<td>0.21%</td>
<td>0.5%</td>
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<td>0.33%</td>
</tr>
</tbody>
</table>

Means with different superscripts (a, b) in column differ significantly (p<0.05) from each other

Fig 4: Effect of individual packaging methods on TSS% of guava fruits during storage at low temperature

Bars followed by different letters are significantly different at P<0.05 Vertical bars represents SE (n=3).
65% RH (Sahoo et al., 2006) [16] and waxed apples at 27 °C and 10 °C. Individual wrapping of fruits helped in retention of higher ascorbic acid in fruits during storage (Table 2). Maximum ascorbic acid was retained in vacuum (36.4 mg/100g) followed by modified atmosphere (35.4 mg/100g) packed fruits. Individual wrapping created the modified atmosphere around the fruits leading to reduced O2 concentration, which slowed down the oxidation of ascorbic acid and thus retained higher ascorbic acid content. The result obtained in the present investigation are in accordance with the previously reported results by Sudha et al. (2007) [6], where maximum retention of ascorbic acid has been reported in 150 gauge polythene vacuum packed sapota as compared to unwrapped control fruits stored at room temperature.

Table 2: Effect of individual wrapping on Total Phenol content (mg/100g) and Ascorbic acid content (mg/100g) at different storage period

<table>
<thead>
<tr>
<th>Period of storage (Days)</th>
<th>Control</th>
<th>Vacuum Pack</th>
<th>Modified atmosphere pack</th>
<th>Control</th>
<th>Vacuum Pack</th>
<th>Modified atmosphere pack</th>
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<td>0</td>
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<td>62.8a</td>
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<td>41.3a</td>
<td>41.3a</td>
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<td>61.6b</td>
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<td>49.1b</td>
<td>52.7c</td>
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<tr>
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<td>43.6b</td>
<td>47.3c</td>
<td>28.3f</td>
<td>35.2b</td>
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<tr>
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<td>23.4a</td>
<td>38.2b</td>
<td>40.9c</td>
<td>24.7g</td>
<td>34.1b</td>
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<tr>
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<td>17.7a</td>
<td>34.5b</td>
<td>32.5b</td>
<td>21.8h</td>
<td>30.8b</td>
<td>31.6i</td>
</tr>
<tr>
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<td>13.6a</td>
<td>29.1b</td>
<td>30.9b</td>
<td>19.4i</td>
<td>30.6b</td>
<td>29.9j</td>
</tr>
</tbody>
</table>

Means with different superscript (a, b) in column are different from each other

Total Phenols
Decrease in phenolic content with the advancement of storage was observed at low temperature also (Table 2). By 21th day of storage at low temperature, phenolic content in control fruits was 12.8 mg/100g while in individually wrapped fruits it ranged from 30-31 mg/100g. Decrease in phenolic content could be related to ripening during storage. During ripening, there was increased activity of PPO, the enzyme responsible for oxidation of phenols and this increased activity may be responsible for the decreased phenolic content during storage. Slower decrease in phenol at low temperature might be due to low rate of physiological processes. Similar decrease in tannin content of guava cvs. ‘Allahabad Safeda’ and ‘Lucknow – 49’ stored at room temperature has been reported by Tandon et al., (1982) [26].

Individual wrapping of fruits reduced the decrease in phenolic content during storage. At low temperature, among all wrapping methods, vacuum (47.3mg/100g) showed maximum retention of phenolics followed by modified atmosphere (48.3 mg/100g) packed fruits (Table 2). However, differences were found to be non-significant among vacuum and modified atmosphere wrapping methods. In the present investigation, the individual wrapping helped in retention of phenolic content during storage by reducing the respiratory rates and by delaying ripening. Similarly, lower decrease in tannin content as compared to control fruits in 150 gauge LDPE film shrink wrapped bell pepper and mangoes in stored at 27 °C and 65% RH has been reported by Sahoo et al., (2015) [12].

Overall Acceptability
At low temperature storage conditions, the overall acceptability of control fruits improved up to 3rd day only and turned unacceptable by 15th day (Fig. 5). Among wrapped fruits MA and vacuum packed fruits showed increase in acceptability up to 6th day and was 7.4 and 7.2 which then decreased to 4.9 and 4.8 by 21st day and thus showing unacceptable overall organoleptic quality of the fruits.

Among individually wrapped fruits, vacuum and MA packed fruits had lower score and were unacceptable by the end of storage period (Fig. 5). Organoleptic score of all packed fruits at later storage period remained low despite of the fact that there was delay in ripening, retention of green colour and flesh firmness, lower PLW and maintenance of sufficient TSS. The development of off flavour and accumulation of ethanol and acetic acid were the main factor responsible for the incipient taste and reduced organoleptic score of MA and vacuum packed fruit. The hardening of fruits in MA and vacuum pack could be possible due to gelling behaviour of water soluble pectins, which reconstitute to form protopectin and lead to flesh hardening on prolonged storage. Rana et al., (2015) [4] also reported highest mean score for all the attributes in cling wrapped guava in LDPE film stored at 10°C, followed by shrink wrapped.

Fig 5: Effect of individual packaging methods on overall acceptability of guava fruits during storage at low temperature

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Conclusion
In conclusion, individual wrapping reduced the magnitude of physico-chemical changes during storage and helped in retention of quality for longer time, while reduced ripening rate and hard texture, strongly altered the overall acceptability of fruits. However, both treatments were equally effective in maintaining desirable characteristics and overall acceptability by minimizing the physico-chemical changes. Vacuum packing and MA though showed minimum PLW and ripening but suffered from the drawback that fruits remained hard and unripened and scored low organoleptically.

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References