

The combined effect of lactic acid and natural plant extracts from guava leaves and pomegranate peel on the shelf life of fresh-cut apple slices during cold storage

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Abstract

Freshly-cut or minimally processed fruits have a shorter shelf life than unprocessed ones. The present study was conducted to investigate the effect of natural plant extracts from guava leaves (GE 0.1%), pomegranate peel (PE 0.1%), and lactic acid (LA 1%) on the shelf life of fresh-cut apple slices (FAS) during storage. Microbiological quality, physicochemical properties, and sensory characteristics of FAS samples were monitored during cold storage (4±1°C). Samples treated with natural antimicrobials showed lower psychrotrophic bacterial and yeasts and moulds count compared to the control at day 0 and during the storage. All treated samples were free of Enterobacteriaceae up to the end of storage periods. During storage, moisture content and titratable acidity decreased while pH and soluble solids increased in all samples. Generally, the changes in the physicochemical properties of treated samples were lower compared to control. Sensory scores of all samples decreased throughout the storage, treated samples retained higher scores compared to the control. The treatments of LA 1% + PE 0.1% and LA 1% + GE 0.1% were effective in keeping the quality and prolonging the shelf life of FAS. Such treatments showed better microbial, physiochemical, and sensorial quality over a storage period of 18 days under refrigerated conditions. Our findings suggested the potential use of these natural plant extracts to retain the quality and prolong the shelf life of a fresh-cut apple.

1. Introduction

Increasing demand for fresh, convenient, available, nutritious, and ready-to-eat food products was observed in the last few years (Huang *et al.*, 2021). Minimally processed fruits (MPF) (also called ready-to-eat, easy-to-use, fresh-cut, or pre-cut produce) are products that have been subjected to some processing techniques including cleaning, washing, peeling, cutting, and packaging without further treatments, and kept under refrigeration conditions to maintain the quality and freshness of the whole fruit (De Corato, 2020).

However, MPF is highly susceptible to quality changes during processing and storage and has a shorter shelf life than unprocessed ones, which is mainly related to processing operations i.e. peeling, cutting, slicing, and shredding (Ma *et al.*, 2017). Quality changes of MPF include enzymatic browning, tissue softening, loss of nutrients, formation of off-flavour, and microbiological

spoilage. Tissue damage and the release of nutrients due to processing operations may increase microbial contamination (Ma *et al.*, 2017). Moreover, MPF may be contaminated with some pathogenic microorganisms during production, harvesting, processing, transportation, handling, retailing and storage (Stephan *et al.*, 2015). Apple, one of the most consumed fruits in the world, is a rich source of dietary fibres, minerals, and polyphenols. Fresh-cut apple, like most MPF, is a perishable product due to the damages that occurred by minimal processing operations.

The shelf life and safety of MPF may be improved through the application of good agricultural practices (GAP), good manufacturing practices (GMP), and maintaining the cold chain (Legnani and Leoni, 2004). In fact, the control of food spoilage and pathogenic bacteria is mainly achieved by chemical control particularly washing with sanitizing solutions. However, the use of

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synthetic chemicals is not recommended due to the undesirable aspects including carcinogenicity, toxicity, teratogenicity, and slow degradation periods. In the last few years, plant extracts have been examined for their effectiveness to be used as promise and safe food preservatives. Several studies have also shown the good potential of natural antimicrobials to increase the safety and shelf life of MPF (Delgado *et al.*, 2004; Verma *et al.*, 2012; Khalifa *et al.*, 2016; Gull *et al.*, 2021).

Guava leaves extract has antioxidant, antibacterial, and anticancer activity due to the presence of phenolic compounds (Nantitanon and Okonogi, 2012; Correa *et al.*, 2016; Seddiek *et al.*, 2020). Likewise, pomegranate peel, a by-product of pomegranate juice extraction, is an excellent source of nutrients and phenolic compounds (Kumar *et al.*, 2021). Previous studies reported that pomegranate peel has antioxidant and antimicrobial effects (Gull *et al.*, 2021). Lactic acid, generally recognized as safe (GRAS), has a strong antibacterial effect against a wide range of different food spoilage and pathogenic bacteria (Wemmenhove *et al.*, 2018; Chen *et al.*, 2019). Therefore, the current study aimed to use natural antimicrobials (guava leaves and pomegranate peel extracts) along with lactic acid at low concentrations to enhance safety, maintain quality, and extend the shelf life of fresh-cut apple slices during cold storage.

2. Materials and methods

2.1 Raw materials

Fresh apple (*Malus domestica*) fruits, fresh guava (*Psidium guajava*) leaves, and fresh pomegranate (*Punica granatum*) peel were procured from a private farm in Beheira governorate, Egypt. Chemicals, reagents, and microbial media were purchased from El-Gomhouria company for chemicals and drugs, Alexandria, Egypt. Storage packages of low-density polyethylene (LDPE) were procured from the local market.

2.2 Preparation and evaluation of the plant extracts

Plant extracts of guava leaves and pomegranate peel were prepared and evaluated in terms of total phenolics, total flavonoids, antioxidant activity, and antimicrobial activity as shown in the study by Seddiek *et al.* (2020).

2.3 Preparation of apple fruits

Fruits of similar size and maturity stage and free of physical damage, microbial infection, and insect infection were selected. Then, apple fruits were washed with tap water, drained, and sliced into a crescent shape using an apple slicer with the removal of undesirable parts (Siroli *et al.*, 2015).

2.4 Disinfection treatments

According to our latest study (Seddiek *et al.*, 2020) and the preliminary experiments, the best concentrations of the plant extracts were 0.1% of guava leaves extract (GE) and 0.1% of pomegranate peel extract (PE). Regarding lactic acid (LA), previous studies reported that 1% of LA solution was the best treatment for extending the shelf life without affecting the quality of minimally processed vegetables (Ali, 2009). Accordingly, fresh-cut apple slices (FAS) were disinfected by spraying with the following solutions: PE 0.1%, GE 0.1%, LA 1%, LA 1% + PE 0.1%, LA 1% + GE 0.1%, and the control. Then, treated samples and the control were dried on a sanitized table using a fan for moisture removal. All samples were kept in a refrigerator for three hours to complete the dryness. Finally, samples were packaged in LDPE bags, sealed, and stored in a refrigerator at 5±1°C. For further analyses, FAS samples were withdrawn at 0, 4, 8, 12, 16, and 18 days of storage.

2.5 Storage studies

2.5.1 Microbial analysis

Total psychrotrophic bacterial counts were determined using plate count agar medium (PCA Oxoid C.M 325) according to Martin-Diana *et al.* (2006). *Enterobacteriaceae* group counts were determined using violet red bile glucose agar medium (VRBG Oxoid C.M 485) by following the method described by Klaiber *et al.* (2005). Yeasts and moulds counts were determined using Rose Bengal chloramphenicol medium (Oxoid C.M 549) according to Voon *et al.* (2006). The microbial counts were expressed as the logarithm of colony-forming units per gram of fresh weight of the samples (log CFU/g).

2.5.2 Moisture content

The moisture content of FAS samples was determined by the direct oven drying method (AOAC, 2006). Ten grams of FAS samples were dried in the oven at 105°C until constant weight. Moisture content was calculated as follows:

$$\text{Moisture content (\%)} = \frac{\text{Weight of fresh sample} - \text{weight of dried sample}}{\text{Weight of fresh sample}} \times 100$$

2.5.3 pH

The pH of FAS samples was measured using a digital pH-meter (Denver Instruments Co., USA) at room temperature according to Martin-Diana *et al.* (2006). The device was previously calibrated using buffer solutions. Ten grams of FAS samples were blended for 2 mins in 20 mL of distilled water. Then, the pH-meter electrode was put in the juice and the readings were taken instantly.

2.5.4 Titratable acidity

The titratable acidity (TA) was estimated by following the method described by Nourian *et al.* (2003). Approximately 10 g of FAS samples were added to 90 mL of distilled water and mixed in a blender. Then, the mixture was filtered and titrated with 0.1 N sodium hydroxide to an endpoint (using phenolphthalein indicator). TA was expressed as the percentage of malic acid per 100 g of FAS samples as follows:

$$\text{TA (\%)} = \frac{\text{Ml of NaOH} \times 0.1 \text{ N} \times 0.067}{\text{Weight of sample}} \times 100$$

2.5.5 Total soluble solids (TSS)

The TSS of FAS samples were estimated using a hand-held refractometer (ATAGO N1, Brix 0-32%, Japan) according to the method of AOAC, (2006). A small piece of FAS samples was put in a lemon press to squeeze out the juice. A drop of the juice was placed on the refractometer prism, the cover plate was lowered, and the readings were taken.

2.5.6 Sensory evaluation

Organoleptic characteristics of FAS samples were evaluated using the nine-point hedonic scale (from like extremely = 9 to dislike extremely = 1) according to the method described by Kumar *et al.* (2021). The samples were coded and evaluated by 10 experienced panellists in

terms of colour, odour, texture, taste, and overall acceptability.

2.6 Statistical analysis

For all samples, determinations were carried out in triplicates and the results were statistically analyzed using ANOVA. Duncan's multiple range test was applied to determine the significant ($P < 0.05$) differences between means using SPSS software (SPSS Inc., Chicago, USA) (Alqahtani *et al.*, 2021).

3. Results and discussion

3.1 Microbial analysis

Microbial spoilage is the major factor responsible for limiting the shelf life of fresh-cut fruits and vegetables. Therefore, microbial growth was investigated during the storage period. The effects of disinfection treatments on the total psychrotrophic (TP), *Enterobacteriaceae*, and yeasts and moulds count of fresh-cut apple slices (FAS) during cold storage are shown in Table 1. The freshly treated samples with natural antimicrobials were almost free of TP growth, whereas the control contained 2.27 log CFU/g. Moreover, the TP count of the control increased significantly ($P < 0.05$) with prolonged storage; it reached 4.01 log CFU/g after 8 days of storage. On the contrary, the treated samples were more stable during storage with an insignificant ($P > 0.05$) increase in the TP

Table 1. Effect of disinfection treatments on the total psychrotrophic, *Enterobacteriaceae*, and yeasts and moulds count (log CFU/g) of fresh-cut apple slices stored at 5±1°C

Microbial counts (log CFU/g)	Storage days	Treatments					
		Control	PE 0.1%	GE 0.1%	LA 1%	LA 1% + PE 0.1%	LA 1% + GE 0.1%
Total psychrotrophic	0	2.27±0.15 ^{Ca}	-	-	-	-	-
	4	3.39±0.19 ^{Ba}	1.89±0.20 ^{Bb}	1.94±0.19 ^{Bb}	-	-	-
	8	4.01±0.23 ^{Aa}	2.01±0.24 ^{ABb}	2.07±0.26 ^{ABb}	1.46±0.21 ^{Ac}	-	-
	12	ND	2.22±0.25 ^{Aa}	2.30±0.27 ^{Aa}	1.59±0.24 ^{Ab}	1.32±0.24 ^{Ab}	1.31±0.21 ^{Bb}
	16	ND	ND	ND	1.73±0.27 ^{Aa}	1.45±0.28 ^{Aa}	1.42±0.17 ^{ABa}
	18	ND	ND	ND	ND	1.68±0.32 ^{Aa}	1.66±0.24 ^{Aa}
<i>Enterobacteriaceae</i>	0	-	-	-	-	-	-
	4	-	-	-	-	-	-
	8	0.36±0.22 ^{Aa}	-	-	-	-	-
	12	ND	-	-	-	-	-
	16	ND	ND	ND	-	-	-
	18	ND	ND	ND	ND	-	-
Yeasts and molds	0	-	-	-	-	-	-
	4	0.59±0.19 ^{Aa}	-	-	-	-	-
	8	0.71±0.26 ^{Aa}	0.58±0.25 ^{Aa}	0.61±0.26 ^{Aa}	-	-	-
	12	ND	0.66±0.28 ^{Aa}	0.68±0.29 ^{Aa}	0.62±0.17 ^{Aa}	-	-
	16	ND	ND	ND	0.68±0.22 ^{Aa}	0.62±0.27 ^{Aa}	0.61±0.28 ^{Aa}
	18	ND	ND	ND	ND	0.68±0.31 ^{Aa}	0.65±0.26 ^{Aa}

Values are presented as mean±standard deviation, n = 3. Values with different lowercase superscripts within the same row and values with different uppercase superscripts within the same column are significantly different ($P < 0.05$). PE: pomegranate peel extract, GE: guava leaves extract, LA: lactic acid, ND: not determined due to sensorial rejection or spoilage.

count. The shelf lives were 12 days for the samples treated with PE 0.1% and GE 0.1%, 16 days for the samples treated with LA 1%, and 18 days for the samples treated with LA 1% + PE 0.1% and LA 1% + GE 0.1%. According to Moreira *et al.* (2015), the TP count of all treated samples remained below the acceptable limit prescribed for fresh-cut products (6.0 log CFU/g) throughout the storage period. The samples treated with LA 1% + PE 0.1% and LA 1% + GE 0.1% showed the longest shelf life (18 days), which could be due to the combined effect of natural plant extracts from guava leaves and pomegranate peel with lactic acid. Same trends were observed for yeasts and moulds count during storage i.e. the treated samples were more stable during storage with an insignificant ($P>0.05$) increase in the yeasts and moulds count compared to the control. By the end of storage, the highest fungal count (0.71 log CFU/g) was recorded by the control after 8 days of storage, whereas the lowest fungal count (0.65 log CFU/g) was recorded by the sample treated with LA 1% + GE 0.1% after 18 days. Regarding *Enterobacteriaceae*, the control recorded 0.36 log CFU/g after 8 days, whereas all treated samples were free of *Enterobacteriaceae* growth up to the end of storage. Previous studies reported that LA, GE, and PE have antimicrobial effects against a wide range of food spoilage and pathogenic microbes (Ali, 2009; Mostafa *et al.*, 2018; Hemeg *et al.*, 2020; Seddiek *et al.*, 2020).

3.2 Moisture content

The loss of water content of fresh-cut products is an indicator of quality decrease. Accordingly, it should be determined intervalley during the storage (Ali, 2009). The changes in the moisture content during storage of FAS treated with different natural antimicrobials are shown in Table 2. The moisture content of the control and treated samples decreased significantly ($P<0.05$) throughout the storage period. Generally, the decrease in the moisture content of the control was higher than that of the treated samples. In addition, no significant differences were found among all treatments during the shelf-life period, as they exhibited the same trend. In fact, the water loss of FAS may be related to the natural

catabolic processes during storage as a result of respiration, senescence, and other metabolic processes, such changes are motivated by enzymes and accelerated by the operations of minimal processing (Martin-Diana *et al.*, 2006). Generally, fresh-cut products may be rejected by the consumers if the water loss was more than 5% (Gross *et al.*, 2016). In the present study, the maximum decrease in the moisture content of FAS samples at the end of storage was approximately 2%, which did not exceed the prescribed limits. Therefore, the disinfection treatments did not cause a perceptible decrease in the freshness of FAS in terms of water loss. Our findings are in harmony with the results reported by Martin-Diana *et al.* (2006) and Tappi *et al.* (2017) who observed a slight decrease in the moisture content of minimally processed carrot, lettuce, and apple slices; they also observed only a few differences among samples during storage.

3.3 pH

The changes in the pH of FAS treated with different natural antimicrobials were monitored throughout the storage period (Table 3). The pH values of FAS samples ranged from 3.38–3.56 to 3.68–4.11 at day 0 and the end of storage, respectively. Significant ($P<0.05$) differences were observed between freshly treated samples, which could be related to the effect of LA i.e., the samples treated with LA showed lower pH values. The pH values increased significantly ($P<0.05$) for all the samples during storage. This increase in pH values could be due to the growth of gram-negative bacteria during the storage, which plays a principal role in the spoilage of fresh-cut products (Gomez-Lopez *et al.*, 2005). Additionally, Limbo and Piergiovanni (2007) reported that the plant cells may produce anaerobic metabolites in terms of volatile compounds, which in turn may be responsible for the increase in pH values during storage. In general, the increase in the pH of treated samples was lower than that of the control. Moreover, the samples treated with LA 1% + PE 0.1% and LA 1% + GE 0.1% retained lower pH values throughout the storage, which could be due to the inhibitory effect of the natural plant extracts from guava leaves and pomegranate peel with

Table 2. Effect of disinfection treatments on the moisture content (%) of fresh-cut apple slices stored at $5\pm 1^\circ\text{C}$

Storage days	Treatments					
	Control	PE 0.1%	GE 0.1%	LA 1%	LA 1% + PE 0.1%	LA 1% + GE 0.1%
0	89.22±0.15 ^{Aa}	89.22±0.24 ^{Aa}	89.22±0.22 ^{Aa}	89.22±0.18 ^{Aa}	89.22±0.14 ^{Aa}	89.22±0.19 ^{Aa}
4	88.15±0.21 ^{Bb}	88.19±0.25 ^{Bb}	88.16±0.26 ^{Bb}	88.63±0.22 ^{Ba}	88.81±0.19 ^{ABa}	88.83±0.26 ^{ABa}
8	87.37±0.14 ^{Cc}	87.79±0.13 ^{Cb}	87.68±0.17 ^{Cb}	88.31±0.17 ^{Ca}	88.52±0.20 ^{BCa}	88.58±0.12 ^{BCa}
12	ND	87.41±0.14 ^{Db}	87.26±0.27 ^{Db}	87.90±0.20 ^{Da}	88.11±0.27 ^{Ca}	88.19±0.27 ^{Ca}
16	ND	ND	ND	87.31±0.11 ^{Eb}	87.63±0.31 ^{Da}	87.68±0.26 ^{Da}
18	ND	ND	ND	ND	87.30±0.37 ^{Da}	87.33±0.24 ^{Da}

Values are presented as mean±standard deviation, n = 3. Values with different lowercase superscripts within the same row and values with different uppercase superscripts within the same column are significantly different ($P<0.05$). PE: pomegranate peel extract, GE: guava leaves extract, LA: lactic acid, ND: not determined due to sensorial rejection or spoilage.

Table 3. Effect of disinfection treatments on the pH of fresh-cut apple slices stored at 5±1°C

Storage days	Treatments					
	Control	PE 0.1%	GE 0.1%	LA 1%	LA 1% + PE 0.1%	LA 1% + GE 0.1%
0	3.56±0.02 ^{Ca}	3.51±0.02 ^{Cb}	3.54±0.01 ^{Dab}	3.38±0.02 ^{Dd}	3.42±0.02 ^{Ec}	3.44±0.01 ^{Ec}
4	3.79±0.05 ^{Ba}	3.62±0.05 ^{Bb}	3.66±0.04 ^{Cb}	3.47±0.06 ^{Cc}	3.50±0.05 ^{DEc}	3.53±0.03 ^{DEc}
8	4.11±0.07 ^{Aa}	3.71±0.09 ^{Abc}	3.74±0.08 ^{Bb}	3.57±0.03 ^{Bd}	3.58±0.07 ^{CDd}	3.59±0.05 ^{CDcd}
12	ND	3.79±0.06 ^{Aa}	3.85±0.03 ^{Aa}	3.61±0.05 ^{ABb}	3.65±0.06 ^{BCb}	3.69±0.07 ^{BCb}
16	ND	ND	ND	3.68±0.07 ^{Aa}	3.70±0.01 ^{Ba}	3.74±0.08 ^{Ba}
18	ND	ND	ND	ND	3.81±0.08 ^{Ab}	3.91±0.09 ^{Aa}

Values are presented as mean±standard deviation, n = 3. Values with different lowercase superscripts within the same row and values with different uppercase superscripts within the same column are significantly different ($P<0.05$). PE: pomegranate peel extract, GE: guava leaves extract, LA: lactic acid, ND: not determined due to sensorial rejection or spoilage.

lactic acid on the microbial growth. The trends obtained for the changes in pH values in the present study are consistent with the results reported by Rico *et al.* (2008).

3.4 Titratable acidity (TA)

Titrate acidity was determined to monitor the changes in the organic acid content of the product during storage. The effects of disinfection treatments on the TA of FAS during storage are presented in Table 4. The TA of the samples varied between 0.62–0.64 to 0.44–0.50 at day 0 and the end of storage, respectively. Generally, TA values decreased significantly ($P<0.05$) in all samples during storage. The decrease in TA could be due to the utilization of organic acids as substrates for enzymatic reactions of the respiratory metabolism (Valero and Serrano, 2010). No significant ($P>0.05$) differences were observed between all treatments throughout the storage period, as they revealed the same trend. However, disinfection treatments significantly ($P<0.05$) inhibited this decrease in TA values during the first 8 days of storage. The control showed the lowest TA value (0.44%) at the end of storage. Among all treatments, FAS samples treated with LA 1% + PE 0.1% and LA 1% + GE 0.1% effectively maintained higher TA values (0.50%) at the end of storage compared to the PE 0.1%, GE 0.1%, and LA 1% treated samples, which exhibited TA values of 0.46, 0.47 and 0.48%, respectively. Garcia *et al.* (1998) reported that the disinfection treatments of fruits and vegetables may decrease the respiration rate and accordingly, delay the degradation of organic acids. Our findings agreed with a recent study conducted by

Gull *et al.* (2021) who observed that TA values decreased gradually during the storage of apricot fruits under refrigeration conditions.

3.5 Total soluble solids (TSS)

The TSS of fresh-cut fruits and vegetables is an important parameter affecting the consumer's acceptance. The changes in the TSS of FAS treated with different natural antimicrobials throughout the storage period are shown in Table 5. Generally, the TSS of all samples increased significantly ($P<0.05$) with the extended storage. Abebe *et al.* (2017) reported that the increase in TSS could be due to the hydrolytic transformations and degradation of polysaccharides into simple sugars, the decomposition of pectic substances into simple components, and the concentration of juice due to moisture loss. Additionally, the sugars and acids of fresh-cut fruits and vegetables are utilized as substrates in the respiratory metabolism, which in turn may cause changes in the TSS with the advancement of storage (Zheng *et al.*, 2007). In the current investigation, the increase in TSS of the control was higher than that of the treated samples. Among all samples, the control revealed the highest TSS value (14.00%) at the end of storage. Moreover, FAS samples treated with LA 1% + PE 0.1% and LA 1% + GE 0.1% revealed better storage stability with TSS values of 12.53 and 12.59%, respectively at the end of storage, whereas samples treated with PE 0.1%, GE 0.1%, and LA 1% exhibited TSS values of 13.84%, 13.86% and 13.90%, respectively. The disinfection treatments could prevent

Table 4. Effect of disinfection treatments on the total titratable acidity (% of malic acid) of fresh-cut apple slices stored at 5±1°C

Storage days	Treatments					
	Control	PE 0.1%	GE 0.1%	LA 1%	LA 1% + PE 0.1%	LA 1% + GE 0.1%
0	0.62±0.03 ^{Aa}	0.64±0.02 ^{Aa}	0.63±0.03 ^{Aa}	0.64±0.02 ^{Aa}	0.63±0.04 ^{Aa}	0.63±0.06 ^{Aa}
4	0.58±0.02 ^{Ba}	0.60±0.04 ^{ABa}	0.59±0.04 ^{ABa}	0.60±0.06 ^{ABa}	0.59±0.02 ^{ABa}	0.59±0.05 ^{ABa}
8	0.44±0.04 ^{Cb}	0.56±0.03 ^{Ba}	0.55±0.02 ^{Ba}	0.56±0.05 ^{BCa}	0.55±0.03 ^{BCa}	0.55±0.02 ^{BCa}
12	ND	0.46±0.02 ^{Cb}	0.47±0.04 ^{Cb}	0.53±0.03 ^{CDa}	0.52±0.03 ^{CDa}	0.52±0.02 ^{CDa}
16	ND	ND	ND	0.48±0.01 ^{Da}	0.51±0.01 ^{CDa}	0.52±0.03 ^{CDa}
18	ND	ND	ND	ND	0.50±0.02 ^{Da}	0.50±0.02 ^{Da}

Values are presented as mean±standard deviation, n = 3. Values with different lowercase superscripts within the same row and values with different uppercase superscripts within the same column are significantly different ($P<0.05$). PE: pomegranate peel extract, GE: guava leaves extract, LA: lactic acid, ND: not determined due to sensorial rejection or spoilage.

Table 5. Effect of disinfection treatments on the total soluble solids (%) of fresh-cut apple slices stored at 5±1°C*

Storage days	Treatments					
	Control	PE 0.1%	GE 0.1%	LA 1%	LA 1% + PE 0.1%	LA 1% + GE 0.1%
0	11.85±0.23 ^{Ca}	11.85±0.29 ^{Da}	11.85±0.24 ^{Da}	11.85±0.18 ^{Da}	11.85±0.23 ^{Ca}	11.85±0.12 ^{Ca}
4	12.69±0.26 ^{Ba}	12.21±0.17 ^{Cb}	12.23±0.26 ^{Cb}	12.02±0.22 ^{Cdb}	11.94±0.26 ^{BCb}	11.92±0.23 ^{BCb}
8	14.00±0.24 ^{Aa}	12.71±0.16 ^{Bb}	12.79±0.17 ^{Bb}	12.30±0.24 ^{Cc}	12.22±0.17 ^{ABCc}	12.20±0.28 ^{ABCc}
12	ND	13.84±0.26 ^{Aa}	13.86±0.29 ^{Aa}	12.77±0.27 ^{Bb}	12.41±0.32 ^{ABb}	12.37±0.19 ^{ABb}
16	ND	ND	ND	13.90±0.17 ^{Aa}	12.50±0.31 ^{Ab}	12.48±0.27 ^{Ab}
18	ND	ND	ND	ND	12.53±0.28 ^{Aa}	12.59±0.31 ^{Aa}

Values are presented as mean±standard deviation, n = 3. Values with different lowercase superscripts within the same row and values with different uppercase superscripts within the same column are significantly different ($P<0.05$). PE: pomegranate peel extract, GE: guava leaves extract, LA: lactic acid, ND: not determined due to sensorial rejection or spoilage.

Table 6. Effect of different antimicrobial treatments on the organoleptic properties of fresh-cut apple slices stored at 5±1° C during different periods

Quality attributes	Storage days	Treatments					
		Control	PE 0.1%	GE 0.1%	LA 1%	LA 1% + PE 0.1%	LA 1% + GE 0.1%
Colour	0	8.44±0.28 ^{Aa}	8.45±0.18 ^{Aa}	8.43±0.15 ^{Aa}	8.53±0.24 ^{Aa}	8.61±0.19 ^{Aa}	8.63±0.19 ^{Aa}
	4	7.40±0.19 ^{Bb}	8.18±0.27 ^{ABa}	7.99±0.25 ^{Ba}	8.31±0.25 ^{ABa}	8.41±0.29 ^{ABa}	8.44±0.25 ^{ABa}
	8	5.88±0.22 ^{Cb}	7.94±0.18 ^{BCa}	7.79±0.29 ^{BCa}	8.06±0.19 ^{Ba}	8.15±0.36 ^{ABa}	8.17±0.19 ^{BCa}
	12	ND	7.75±0.29 ^{Cab}	7.50±0.37 ^{Cb}	7.95±0.29 ^{Bab}	8.04±0.25 ^{Ba}	8.08±0.33 ^{BCa}
	16	ND	ND	ND	7.89±0.26 ^{Ba}	7.96±0.34 ^{Ba}	7.99±0.28 ^{BCa}
	18	ND	ND	ND	ND	7.88±0.29 ^{Ba}	7.91±0.15 ^{Ca}
Odour	0	8.48±0.12 ^{Aa}	8.51±0.25 ^{Aa}	8.46±0.25 ^{Aa}	8.64±0.15 ^{Aa}	8.68±0.24 ^{Aa}	8.70±0.28 ^{Aa}
	4	7.58±0.24 ^{Bb}	8.25±0.24 ^{ABa}	8.21±0.14 ^{ABa}	8.42±0.23 ^{ABa}	8.49±0.13 ^{ABa}	8.51±0.14 ^{ABa}
	8	6.12±0.19 ^{Cb}	8.01±0.13 ^{Ba}	7.95±0.29 ^{BCa}	8.20±0.18 ^{BCa}	8.27±0.15 ^{ABCa}	8.29±0.25 ^{ABCa}
	12	ND	7.92±0.29 ^{Ba}	7.80±0.12 ^{Ca}	8.05±0.35 ^{BCa}	8.13±0.32 ^{BCa}	8.15±0.34 ^{BCa}
	16	ND	ND	ND	7.97±0.26 ^{Ca}	8.01±0.17 ^{Ca}	8.04±0.16 ^{BCa}
	18	ND	ND	ND	ND	7.92±0.26 ^{Ca}	7.98±0.27 ^{Ca}
Texture	0	8.46±0.11 ^{Aa}	8.50±0.27 ^{Aa}	8.46±0.25 ^{Aa}	8.62±0.16 ^{Aa}	8.66±0.11 ^{Aa}	8.69±0.25 ^{Aa}
	4	7.42±0.14 ^{Bb}	8.24±0.18 ^{ABa}	8.25±0.28 ^{ABa}	8.41±0.21 ^{ABa}	8.47±0.22 ^{ABa}	8.48±0.24 ^{ABa}
	8	5.99±0.22 ^{Cc}	7.95±0.28 ^{BCb}	8.02±0.16 ^{Bab}	8.17±0.17 ^{BCab}	8.33±0.27 ^{ABCab}	8.39±0.11 ^{ABCa}
	12	ND	7.78±0.17 ^{Cab}	7.61±0.16 ^{Cb}	8.03±0.22 ^{Ca}	8.11±0.24 ^{BCa}	8.14±0.32 ^{BCa}
	16	ND	ND	ND	7.92±0.28 ^{Ca}	7.99±0.27 ^{Ca}	8.01±0.17 ^{Ca}
	18	ND	ND	ND	ND	7.91±0.19 ^{Ca}	7.94±0.28 ^{Ca}
Taste	0	8.39±0.12 ^{Aa}	8.40±0.13 ^{Aa}	8.38±0.32 ^{Aa}	8.51±0.17 ^{Aa}	8.56±0.14 ^{Aa}	8.61±0.17 ^{Aa}
	4	7.35±0.14 ^{Bb}	8.11±0.27 ^{ABa}	7.98±0.15 ^{ABa}	8.30±0.15 ^{ABa}	8.33±0.25 ^{ABa}	8.35±0.23 ^{ABa}
	8	5.91±0.22 ^{Cb}	7.87±0.28 ^{BCa}	7.70±0.28 ^{Ba}	8.05±0.23 ^{Ba}	8.09±0.33 ^{Ba}	8.11±0.19 ^{Ba}
	12	ND	7.71±0.23 ^{Ca}	7.58±0.34 ^{Ba}	7.97±0.31 ^{Ba}	8.03±0.25 ^{Ba}	8.05±0.18 ^{Ba}
	16	ND	ND	ND	7.93±0.26 ^{Ba}	8.00±0.19 ^{Ba}	8.01±0.34 ^{Ba}
	18	ND	ND	ND	ND	7.95±0.26 ^{Ba}	7.98±0.25 ^{Ba}
Overall acceptability	0	8.44±0.13 ^{Aa}	8.47±0.17 ^{Aa}	8.43±0.25 ^{Aa}	8.58±0.17 ^{Aa}	8.63±0.17 ^{Aa}	8.66±0.15 ^{Aa}
	4	7.43±0.24 ^{Bb}	8.20±0.26 ^{ABa}	8.07±0.32 ^{Ba}	8.36±0.24 ^{ABa}	8.43±0.18 ^{ABa}	8.45±0.12 ^{ABa}
	8	5.97±0.28 ^{Cb}	7.94±0.18 ^{BCa}	8.04±0.17 ^{Ba}	8.12±0.35 ^{BCa}	8.18±0.26 ^{BCa}	8.20±0.27 ^{BCa}
	12	ND	7.79±0.24 ^{Cab}	7.62±0.18 ^{Cb}	8.00±0.24 ^{BCa}	8.08±0.17 ^{BCa}	8.11±0.19 ^{BCa}
	16	ND	ND	ND	7.92±0.12 ^{Ca}	7.99±0.29 ^{Ca}	8.01±0.23 ^{Ca}
	18	ND	ND	ND	ND	7.93±0.24 ^{Ca}	7.95±0.27 ^{Ca}

Values are presented as mean±standard deviation, n = 3. Values with different lowercase superscripts within the same row and values with different uppercase superscripts within the same column are significantly different ($P<0.05$). PE: pomegranate peel extract, GE: guava leaves extract, LA: lactic acid, ND: not determined due to sensorial rejection or spoilage.

the drastic changes in the TSS of fruits by decreasing the microbial growth, reducing the respiration rate, and preventing the hydrolysis of polysaccharides. The trends obtained in the present research are consistent with a recent study undertaken by Zhou *et al.* (2021), who observed that the TSS of mango increased gradually with the advancement of storage time.

3.6 Sensory evaluation

The sensory quality attributes such as colour, odour, texture, and taste are crucial factors affecting consumers' acceptance (Chen *et al.*, 2019). The changes in sensory scores of FAS treated with different natural antimicrobials during cold storage are given in Table 6. The sensory scores of colour, odour, texture, taste and overall acceptability for all samples decreased significantly ($P < 0.05$) with the extended storage. Generally, the decrease in colour scores is related to enzymatic browning, whereas the decline in texture scores is mainly due to the degradation of pectic compounds. Likewise, odour and taste changes are related to respiratory metabolism and microbial growth. However, no significant ($P > 0.05$) differences in sensory scores were observed between the FAS samples throughout the storage period, as they showed the same trend. It is worth noting that the decrease in sensory scores of the control was higher than that of the treated samples. Moreover, the control exhibited the lowest overall acceptability score (5.97) at the end of storage (8 days). In contrast, the samples treated with LA 1% + PE 0.1% and LA 1% + GE 0.1% showed better storage stability and retained higher sensory scores throughout the storage compared to other treatments with overall acceptability scores of 7.93 and 7.95, respectively at the end of storage (18 days). It is noteworthy that these scores were higher than the limits (≥ 6) prescribed for fruits and vegetables (Huang *et al.*, 2021). The extended shelf life of these treatments could be due to the synergistic effect of natural plant extracts from guava leaves and pomegranate peel with lactic acid by inhibiting microbial growth, preventing enzymatic browning, reducing the decomposition of pectic substances, and decreasing the respiration rate. Such effects reduced the changes in colour, odour, texture, and taste of FAS samples during storage. The results reported in the current research are in harmony with the study conducted by Kumar *et al.* (2021) who observed that pomegranate extracts retained the sensory characteristics with a slight decrease during the cold storage of bell pepper.

4. Conclusion

The current investigation revealed that the treatments

of LA 1% + PE 0.1% and LA 1% + GE 0.1% were effective in retaining the quality and extending the shelf life of fresh-cut apples during storage. Such treatments exhibited better microbial, physicochemical, and sensorial quality of fresh-cut apple slices over a storage period of 18 days under refrigerated conditions compared to the control sample. The samples treated with natural plant extracts along with lactic acid were free of pathogenic bacteria up to the end of storage reflecting good hygienic quality. The results reported in the present study suggest that the combined effect of natural plant extracts from guava leaves and pomegranate peel with lactic acid could be considered an effective and safe preservation method to retain the quality and prolong the shelf life of a fresh-cut apple.

Conflict of interest

The authors have declared no conflicts of interest.

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