J. Amer. Soc. Hort. Sci. 149(3):152–161. 2024. https://doi.org/10.21273/JASHS05346-23

Varied Attributes of Jalapeño Pepper Cultivars Influence Fresh-cut Product Quality

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Keywords. *Capsicum annuum*, electrolyte leakage, image analysis, modified atmosphere package, shelf-life, texture

Abstract. During the past 40 years, the US fresh-cut product market has experienced a consistent increase in demand because consumers prioritize health and convenience. Increased interest in fresh-cut products and ready-to-eat vegetables has led to innovations in breeding, product selection, and packaging. However, despite the increased popularity of bell pepper and chile pepper (*Capsicum annuum* L.), research of fresh-cut jalapeño pepper is limited. This study was conducted to identify jalapeño cultivars that could be suitable as a raw fresh-cut product and explore measures beyond tissue membrane electrolyte leakage (EL) of processed products that may be useful for the identification of cultivars suitable for fresh-cut applications. A total of 22 fresh-cut parameters were examined across five cultivars of jalapeño peppers and 10 intercrosses of these cultivars, including visual quality based on an image analysis via a computer vision system, package headspace gas composition, tissue membrane EL, and texture. Based on our results, the genotypes were grouped into five clusters using a cluster analysis. Variables including tissue softening (r^2 = 0.95), EL (r^2 = 0.95), total energy of the mesocarp (r^2 = 0.95), and package headspace carbon dioxide (CO_2) partial pressure (r^2 = 0.94) had strong associations with the cluster. A principal component analysis with biplots further confirmed the results. Cultivars Goliath and Emerald Fire and their hybrids in the first and second clusters showed good quality for fresh-cut applications. The fifth cluster, represented by a single cultivar, Jalapeño M, had the smallest physical size, rapid shelf-life decline, accumulated CO_2 partial pressures, increased EL, and rapid tissue softening in comparison with the other genotypes. All jalapeño cultivars except Jalapeño M maintained good quality until day 14 postprocessing, and some maintained good quality until 21 days postprocessing. Hybrid crosses suggested that two of the cultivars evaluated, Goliath and Emerald Fire, were useful as parents when transferring superior fresh-cut quality traits to progeny. Traditionally, the EL level has been used as an index of freshness (or tissue deterioration). Our results showed that other quality analyses, including measurements of tissue softening via an imaging analysis, and physical analyses of tissue firmness can also be used as indices for the freshness of fresh-cut jalapeños. The results suggest that fruit size, wall thickness, and skin toughness might be useful as predictive measures in the field for the selection of jalapeño genotypes with superior fresh-cut quality.

Capsicum species are members of the family *Solanaceae.* They are represented by five cultivated species with diverse pod types. Jalapeño pepper is a pod type of *Capsicum annuum* L., which is a species with a great diversity of pod types that are differentiated by their pod shape as well as their use (Bosland and Votava 2012; Howard and Hernandez-Brenes 1998; Weisenfelder et al. 1978). Jalapeño pepper is a good source of vitamin C, vitamin B6, and vitamin E, even though water comprises 92% of the fresh weight (US Department of Agriculture, Agricultural Research Service 2019). Genetic enrichment of jalapeño pepper has been researched and germplasm has been developed to endorse suitable traits for resistance to viruses and related diseases, attractive fruit according to physical size, wall thickness, corking, and higher yields (Bosland and Votava 2012; Stommel et al. 2016a, 2016b).

In 2021, the combined value of bell pepper and chile pepper production in the United Sates was more than $536 million. Bell pepper represented $461 million of this production value, and chile pepper represented $75 million of this production, which includes jalapeño pepper. Chile pepper production values for 2022 fresh and processing markets were $16.9 million and $60.2 million, respectively (US Department of Agriculture, National Agricultural Statistics Service 2023).

Fresh-cut produce (fruit and vegetable) is a type of minimally processed produce that has been physically transformed from its intact or whole form into a variety of cuts as a result of chopping, dicing, peeling, ricing, shredding, slicing, or spiralizing. Fresh-cut produce is typically washed, but it does not undergo any additional processing such as blanching or cooking before storage in commercial settings for up to 10 d (US Department of Health and Human Services, Food and Drug Administration 2008). The fresh-cut produce market has grown steadily since 1980; this growth has been driven by consumer demands for

Received for publication 29 Sep 2023. Accepted for publication 30 Jan 2024. Published online 13 May 2024.
We thank Michail Kozlov and Judith Dumm, Vaidehi Bhagat, Ellen Turner, Dr. Ellen R. Bornhorst, and Dr. Zi Teng, US Department of Agriculture, Agricultural Research Service, Beltsville, MD, USA, for assistance with field plot work, processing, physical measurements, and image analysis using the computer vision system.
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convenience, freshness, and health. Consumers are increasingly looking for ready-to-eat or prepared foods that are easy to prepare and offer nutritional benefits (Barrett et al. 2010; Delwiche et al. 2019; Giannakourou and Tsironi 2021; James and Ngarmask 2010; Nguz et al. 2005; Rico et al. 2007; Stommel et al. 2016a, 2016b).

Tissue electrolyte leakage (EL) has been used as an index of tissue damage and quality deterioration of fresh-cut produce (Delwiche et al. 2019; Kim et al. 2004; Luo 2007; Luo et al. 2004; Stommel et al. 2016a). Immediately after fresh-cut processing, there is a decline in tissue membrane EL. However, EL commonly increases during storage. This increase is attributed to cell disruption and loss of membrane integrity, which can lead to changes in color, texture, flavor, and microbial growth of fresh-cut produce (Delwiche et al. 2019; Luo et al. 2004; Picchioni et al. 1996; Saltveit 1997, 2000; Stommel et al. 2016a, 2016b).

Stommel et al. (2016a, 2016b) examined fresh-cut attributes of the following pepper genotypes with various fruit phenotypes selected based on pod types: sweet bell, large elongate (i.e., elongated bell to elongate conical); jalapeño; and serrano; these authors found that there was significant difference in fresh-cut attributes among and within these four types of peppers, as assessed by subjective and objective measurements. Furthermore, sweet bell and large elongate genotypes showed increasing EL over time, whereas jalapeño and serrano genotypes maintained stable electrolyte leakage levels. Another study (Delwiche et al. 2019) that used ultraviolet fluorescence hyperspectral imaging also identified differences among pepper pod types and enhanced jalapeño fresh-cut shelf life. Fluorescence imaging was useful for detecting large changes in the fresh-cut tissue integrity of jalapeño genotypes, but it was less sensitive than EL.

Stable EL observed during prior fresh-cut evaluations of jalapeño genotypes suggested their suitability as fresh-cut pepper products. During this study, we further evaluated the potential of jalapeño pepper as a fresh-cut product and explored additional measures to determine its utility by evaluating differences in the jalapeño fresh-cut shelf life. Variations in fresh-cut quality attributes were assessed among five diverse jalapeño cultivars and 10 intercrosses of these cultivars. Variations in these attributes were evaluated to determine their utility beyond tissue membrane EL for monitoring the quality of fresh-cut products during storage and whether some attributes may be useful predictive measures of the cultivar's fresh-cut performance.

Materials and Methods

Plant materials

Fruits of jalapeño peppers were grown and harvested at the mature green stage during pepper field trials in 2018 at the US Department of Agriculture, Agricultural Research Service, Genetic Improvement of Fruits and Vegetable Laboratory, Beltsville, MD, USA. Five commercial cultivars (Emerald Fire, Goliath, Jalapeño M, Mitla, Tricked You) and 10 hybrid breeding lines from intercrosses of these cultivars (Emerald Fire × Goliath, Emerald Fire × Jalapeño M, Emerald Fire × Mitla, Emerald Fire × Tricked You, Goliath × Jalapeño M, Goliath × Mitla, Goliath × Tricked You, Jalapeño M × Mitla, Jalapeño M × Tricked You, Mitla × Tricked You) were used for fresh-cut studies. Harvested fruits were stored in brown paper bags (3 replications × 10 fruits × 15 genotypes) at 10 °C with high relative humidity overnight to reduce water loss and shriveling before processing.

Fresh-cut processing

Whole fruits of each genotype were washed for 1 min in water containing 50 mg/L free chlorine (NaOCl) and adjusted to a pH of 6.0 to 7.0 with citric acid. Using a sharp knife, the fruit stem ends, seeds, and placental tissues were removed. Then, the fruits were sliced transversely into 1.0-cm-thick rings using an industrial slicer (Seven Chefs ECD-302; Nichimo International Inc., Tokyo, Japan). Sliced fruits were washed in a fresh solution of 50 mg/L NaOCl in water for 30 s. The washed fruit slices were centrifuged in a fresh produce centrifuge for 2 min at 300 rpm (0.1047 $rad \cdot s^{-1}$ to remove excess water (Model T-304; Meyer Machine Co, Watsonville, CA, USA). Then, 100-g (*SD*, ±2 g) samples were placed in polypropylene bags with an oxygen transmission rate of 29.5 (oxygen transmission rate, 400) $pmol \cdot s^{-1} \cdot m^{-2} \cdot Pa^{-1}$ (Packaging Concept Inc., Salinas, CA, USA) and heat-sealed. All sealed packages (n = 180; 15 genotypes × 3 replications × 4 timepoints) were stored in the dark at 5 °C for evaluation on days 0, 7, 14, and 21.

Fresh-cut evaluation

Gas composition. During each evaluation timepoint, a sample package was retrieved from the cold storage for each replication (n = 3 for each genotype) for destructive sampling. A septum was placed on the package surface. Headspace atmosphere (CO_2 and O_2) was determined using a gas analyzer (Checkmate II; PBI Dansensor Co., Ringsted, Denmark) after inserting the gas-tight needle through the septum (Delwiche et al. 2019; Kim et al. 2004; Stommel et al. 2016a).

Electrolyte leakage. A 30-g sample from each package evaluated for gas composition was transferred to 300 mL of reverse-osmosis water and submerged for 30 min at 5 °C; then, the electrical conductivity was measured using a conductivity meter (model 135A; Orion Research, Inc., Beverly, MA, USA). After measurement, the sample containers were placed in a −20 °C freezer for at least 24 h and then thawed. Tissue EL for each replication (n = 3) per genotype at each timepoint was expressed as a percentage of the total electrolytes (Delwiche et al. 2019; Luo 2007; Luo et al. 2004; Stommel et al. 2016a). This procedure was repeated two to three times for each sample until the total electrical conductivity was unchanged after repeated freeze–thaw cycles (Luo et al. 2004; Stommel et al. 2016a). Values measured on day 0 were assessed after cutting; even with time lapses between cutting and value measurements of all genotypes, the values were consistent.

Imaging. A computer vision system was used to measure color, size, and decay of whole fruits and fresh-cut peppers. The vision system consisted of a computer, camera, controlled lighting, and image postprocessing software. Images were captured using a camera (Nikon D 800; Nikon Inc., Melville, NY, USA) with a 60-mm lens and the following settings: F-20, 1/30 s, and ISO 640. Then, the images were color-corrected using ColorChecker Camara Calibration software (X-rite ColorChecker® Passport; X-rite Inc., Grand Rapids, MI, USA) and Adobe Lightroom (version 6.3; Adobe Systems Inc., San Jose, CA, USA). A method of segmenting the different regions of interest in the image was developed using Image Pro Premier software (version 9.3b; Media Cybernetics, Inc., Rockville, MD, USA) with the smart segmentation tool (Peng et al. 2021; Srinivasagan et al. 2017; Teng et al. 2019; Yamamoto et al. 2017).

Images were converted from the red, green, and blue color space to the L* a* b* color space (CIE-International Committee on Illumination, Viena, Austria). A color transformation to hue, saturation, and brightness was performed (Bulanon et al. 2002; Otsu 1979). The threshold value was calculated by the thresholding method, which was based on the grayscale histogram of the image (Patil and Bodhe 2011). This technique was applied to separate different regions of whole fruits. Then, the different regions were classified into five categories, and the number of pixels in each class was determined.

Whole-fruit image evaluation. Before fresh-cut processing, images of 10 whole fruits with three replications per genotype at each timepoint were photographed on a matte white background. For the color value of the whole fruits, segmentation was developed to classify pixels into the following five categories: background, dark green, medium green, light green, and cracking. Each pixel in the image was compared with the values of the reference object using Image-Pro Plus software. The physical size of the whole fruits was measured using a method of spatial calibration of images provided by the software program. The exact measurement by a ruler on images was used to calibrate the image (Yamamoto et al. 2016). For size measurements, the following five regions were acquired: area, perimeter, bounding polygon, length, and width.

Fruit slice image evaluation. At each sample period (0, 7, 14, and 21 d), images of 15 pepper slices separate from those used for EL from each package were captured. The samples were spread across a 40.5- × 21.5-cm area on a matte white background and photographed. There were three replications per genotype at each timepoint. Immediately after photographing, these samples were subjected to a texture analysis.

Deteriorated and healthy fresh-cut pepper tissues, as evidenced by tissue softening or the absence of softening, respectively, were determined based on tissue color differences acquired via a digital imaging system using Image-Pro Plus software (Arias et al. 2018; Luo and Tao 2003). Segmentation was classified by the pixel color difference and the class was categorized with defined reference objects. Data are reported as the percentage of the pixel color difference. Various segments of a region were shown depending on the number of classes, where one class showed all objects as one color and the background remained the same. The following four classes showed a range of color to represent different regions of the image while the background remained the same: cut pericarp surface (the part of the cut pepper in the cross-section that is not damaged or healthy tissue); cut external surface (fruit skin shown on the pepper slice); softening tissue (the part of the pepper slice in the cross-section that is damaged or unhealthy tissue softened by deterioration); and lignification (mesocarp that is lignified or whitening of tissue by water loss related to lignification) (Zhang et al. 2021) (Fig. 1). The colors on different regions were converted from red, green, and blue. A color transformation to hue, saturation, and intensity was applied to separate different regions by the four classes. The values for the hue, saturation, and intensity and the luminance, inphase, and quadrature models for color space were acquired by the software program. Additionally, a method of measuring wall thickness was developed on the fresh-cut pepper image.

Texture analysis. The firmness or skin strength of the pepper slices was determined using a TA-XT2 texture analyzer (Model TA.XT Plus; Stable Microsystems, Surrey, England) with a 5-kg load cell and a TA cylinder probe. Samples of five fruit slices from each of three replicates per genotype at each timepoint were trimmed to 1 cm × 1 cm using the special cutting device, and the flat portion was used for the texture analysis. Texture parameters of different pieces of each slice were measured and the fruit interior pericarp surface was measured using a 4-mm probe; measurements through the exterior skin surface were performed using a 3-mm probe. The puncture tests were performed at a speed of 1.0 $mm \cdot s^{-1}$ to a deformation depth of 3.5 mm. Textural parameters were extracted from the deformation curves using the manufacturer's software (Table 1) (Abbott et al. 1984, 2004; Lester and Saftner 2008; Park et al. 2018; Rodoni et al. 2015; Saftner and Lester 2009; Vincent 1990).

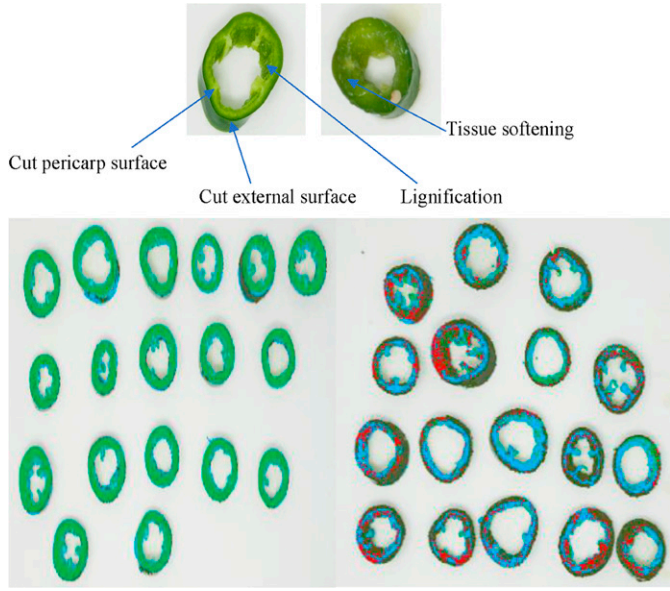

Fig. 1. Deteriorated and healthy fresh-cut pepper tissue, as evidenced by tissue softening or absence of softening, respectively, on fresh-cut jalapeño pepper slices. Softening scores were acquired based on color differences through a digital imaging system with segmentations classified by pixel colors: light green (cut pericarp surface); dark green (cut external surface); red (tissue softening); and light blue (lignification).

Statistical analysis

Data were analyzed using the mixed models procedure in SAS (version 12.1; Statistical Analysis Systems Inc., Cary, NC, USA). All data were initially analyzed as two-factor repeated-measures models with "day" as the repeated factor and "genotype" as the other factor. Mean comparisons were performed among genotypes within a day and across days within a genotype using Sidak adjusted P values at 0.05 (Stommel et al. 2016a, 2016b). The coefficients of determination (the square of the Pearson correlation coefficient) among parameters were derived from the correlation model.

A principal component analysis (PCA) of the multivariate association was performed to describe the relationship among the parameters. The PCA was performed to reduce the dimensionality of the data set using the Proc Princomp procedure in SAS (version 9.4; Statistical Analysis Systems Inc.). The biplots of two factors nominated by the Scree test were constructed to illustrate relationships among pepper characters and genotypes using SigmaPlot (version 13.0; Systat Software, Inc., San Jose, CA, USA).

Table 1. Acronyms of texture profiles extracted from the texture analyzer deformation curves examined across five cultivars of jalapeño peppers and 10 intercrosses of these cultivars.

Acronyms	Description and unit	Texture profile on pepper
MFX	Maximum force from the deformation curve on the mesocarp (N)	Hardness of pepper tissue
EFX	Maximum force from the deformation curve on the exocarp (N)	Strength of pepper skin
MTE	Area under the deformation curve on the mesocarp (N × sec)	Tissue softening
ETE	Area under the deformation curve on the exocarp (N × sec)	Toughness of skin
MP	Peak value in the mesocarp texture profile curve between the anchor at 0 mm and the anchor at 4 mm of penetration represents the points at which the food structure breaks (value)	Crispness or crunchiness of pepper tissue
EP	Peak value in the exocarp texture profile curve between the anchor at 0 mm and the anchor at 4 mm of penetration represents the points at which the food structure breaks (value)	Crispness or crunchiness of pepper skin
MLD	The linear distance is the distance under the deformation curve on the mesocarp determined using the length line joining all points in the selected region between the anchor at 0 mm and the anchor at 4 mm (no unit)	Specific sensory perception of tissue crunchiness (MLD)
ELD	The linear distance is the distance under the deformation curve on the exocarp determined using the length line joining all points in selected region between the anchor at 0 mm and the anchor at 4 mm (no unit)	Specific sensory perception of skin crispness (ELD)

Hierarchical clustering was performed using JMP® Pro 16 (version 16.0.0; Statistical Analysis Systems Inc.). The dendrogram was created to group the 15 test genotypes into clusters based on the following 22 variables scored at day 14 by their homogeneity: O_2 and CO_2 (gas composition); EL; ring width, wall thickness, cut pericarp surface, cut external surface, softening tissue, lignification, L*, a*, b*, hue angle, and chroma (image analysis); and maximum force from the deformation curve on the exocarp (EFX), peak value in the exocarp texture profile curve (EP), area under the deformation curve on the exocarp (ETE), the linear distance under the deformation curve on the exocarp (ELD), maximum force from the deformation curve on the mesocarp (MFX), peak value in the mesocarp texture profile curve (MP), area under the deformation curve on the mesocarp (MTE), and the linear distance under the deformation curve on the mesocarp (MLD) (texture analysis).

Results and Discussion

Instrumental measurement

Fresh-cut products have a shorter shelf-life than intact produce because tissue wounding that occurs during fresh-cut processing triggers the acceleration of tissue respiration (Abe and Watada 1991; Allende et al. 2004; Eskin 1990; Kim et al. 2004; Luo et al. 2004; Toivonen and Brummell 2008; Watada et al. 1996). The package atmospheric gas compositions and tissue EL of fresh-cut jalapeño fruits were significantly affected by pepper genotype and storage time (Table 2). Genotype × storage time interactions for CO_2 and EL were also significant. The O_2 concentration decreased from day 0 to day 14, and it remained relatively stable afterward, whereas the CO_2 concentration increased during the storage time in an inverse relationship with O_2 (Fig. 2A and B). Among the cultivars, Jalapeño M stood out. Packages with 'Jalapeño M' fresh-cut product had a significantly lower O_2 concentration and increased CO_2 concentration at day 7 (2.3% O_2 and 4.5% CO_2) and at day 14 (0.1% O_2 and 8.9% CO_2) compared with those of other cultivars. This indicated that 'Jalapeño M' had a higher respiration rate than the other genotypes. Packages with fresh-cut 'Mitla' fruit exhibited greater CO_2 concentrations by day 21 relative to other genotypes; however, significantly lower concentrations were observed for 'Jalapeño M'. The other genotypes maintained O_2 concentrations between 6.1% and 10.6% and CO_2 concentrations between 2.9% and 3.6% at day 7. After 14 d of storage, the O_2 concentrations of the other genotypes decreased to between 0.9% and 6.3%, and the CO_2 concentrations increased to between 3.6% and 5.7%.

The EL percentage was calculated by measuring electrical conductivity. All genotypes showed a slight decrease in the EL level from day 0 to day 7, followed by a plateau from day 7 to day 14; thereafter, a rapid increase occurred (Fig. 2C). Similar to the head space gas concentration, 'Jalapeño M' was the exception, and the EL of 'Jalapeño M' fresh-cut product increased more rapidly from day 14 to day 21 compared to that of other genotypes, reaching 13.1% and 44.6% at day 14 and day 21, respectively. The other 14 genotypes showed a more gradual increase in the EL level, ranging from 1.8% to 5.7% on day 14 and from 6.1% to 17.7% on day 21. Considering that EL, a

Table 2. Analysis of variance F-values for O_2, CO_2, and electrolyte leakage (EL) of fresh-cut pepper slices of jalapeño pepper fruits stored under passive modified atmosphere packaging conditions for 0, 7, 14 and 21 d.

Parameter	Source	df	SS	MS	F-value
O_2	Genotype	14	163.97	11.71	3.20***
	storage	2	1022.11	511.05	139.60****
	Genotype × storage	28	120.70	4.31	1.18
	Error	90	3.29.48	3.66	
	Total	134	1636.25		
CO_2	Genotype	14	250.25	17.88	8.17****
	storage	2	498.79	249.40	114.04****
	Genotype × storage	28	125.62	4.49	2.05**
	Error	90	196.83	2.19	
	Total	134	1071.50		
EL	Genotype	14	1806.79	129.06	12.03****
	storage	3	2694.79	898.26	83.76****
	Genotype × storage	42	2263.98	53.90	5.03****
	Error	120	1286.87	10.73	
	Total	179	8052.44		

****, ***, **, * Significant at $P \leq 0.0001$, $P \leq 0.001$, $P \leq 0.01$, and $P \leq 0.05$, respectively.
df = degrees of freedom; SS = sum of squares; MS = mean squares.

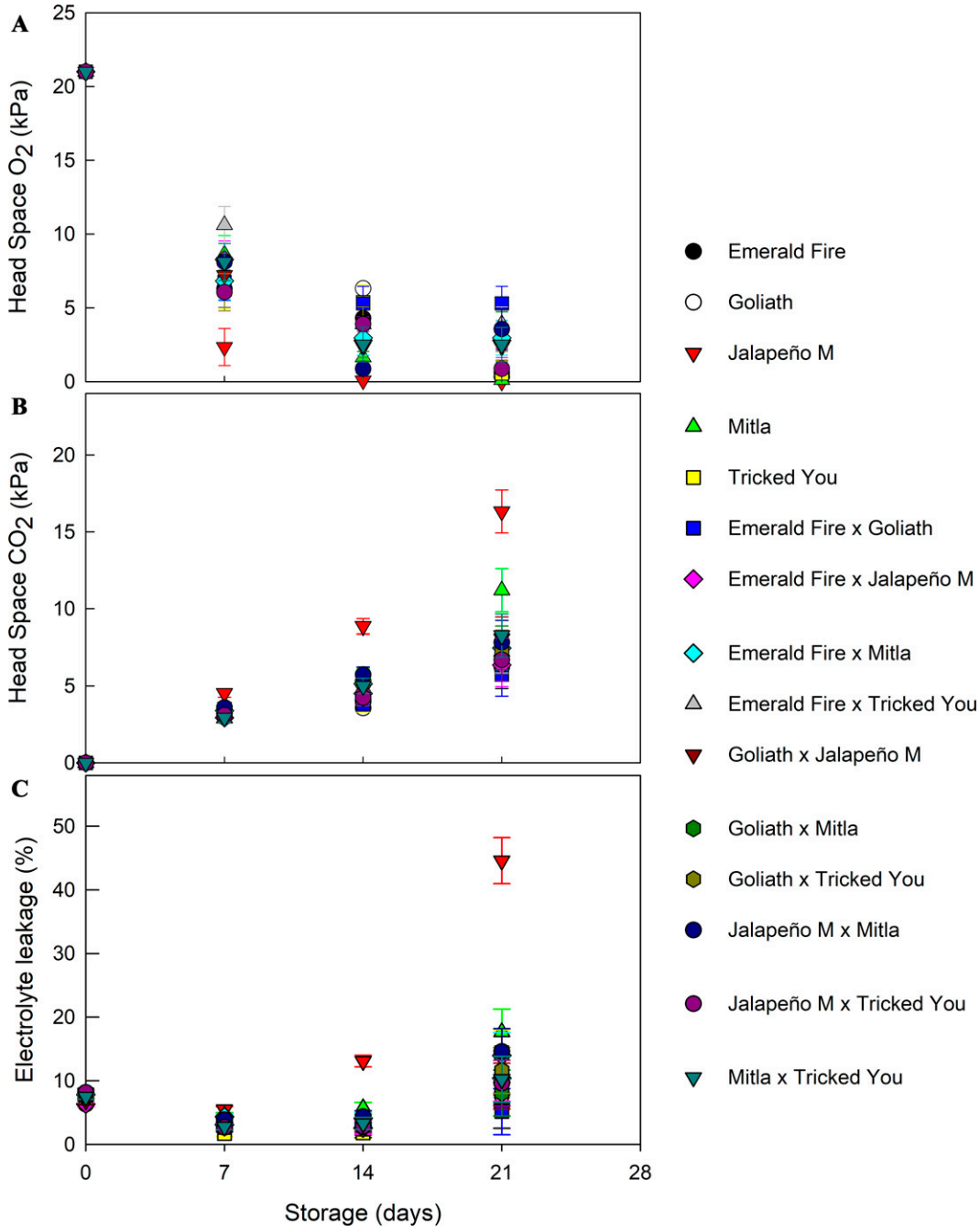

Fig. 2. Package headspace O_2 (**A**) and CO_2 (**B**) and electrolyte leakage (**C**) for fresh-cut jalapeño pepper slices from five cultivars and 10 intercrosses of these cultivars stored at 5 °C for up to 21 d (n = 3). Bars represent the pooled *SE* of the mean.

measure of membrane integrity, is often used as an index of tissue damage and quality deterioration of fresh-cut produce, 'Jalapeño M' is an inferior source of fruit for fresh-cut product.

Texture attributes of fruits and vegetables are closely related to cell strength and turgor pressure (Alvarez et al. 2000a, 2000b, 2020; Fillion and Kilcast 2002; Lin and Pitt 1986). Tissue softening is accelerated by the effects of tissue cutting and wounding on fresh-cut produce. Turgor pressure loss, starch degradation, and chemical modification in the cell decreases over time (in days) in storage, leading to loosening of the rigidity in pepper pericarp and an increase in tissue softening (Rao et al. 2011; Toivonen and Brummell 2008). During our study, an evaluation of texture parameters (Fmax, peak value, total energy, and linear distance) for fresh-cut jalapeño fruit slices during storage demonstrated that most genotypes maintained textural integrity during the storage period, even after 21 d (Supplemental Table 1). The hardest cultivar was Goliath, as indicated by its maximum force (Fmax). The softest cultivar in terms of flesh hardness was Tricked You. Intercross genotypes with 'Tricked You' also showed less hardness. However, even for those intercross genotypes, the original hardness did not deteriorate significantly over time in storage.

Skin toughness is a measure of a material's ability to absorb mechanical energy before it fails. It is measured by the area under the stress–strain curve up to the yield stress and has the dimensions

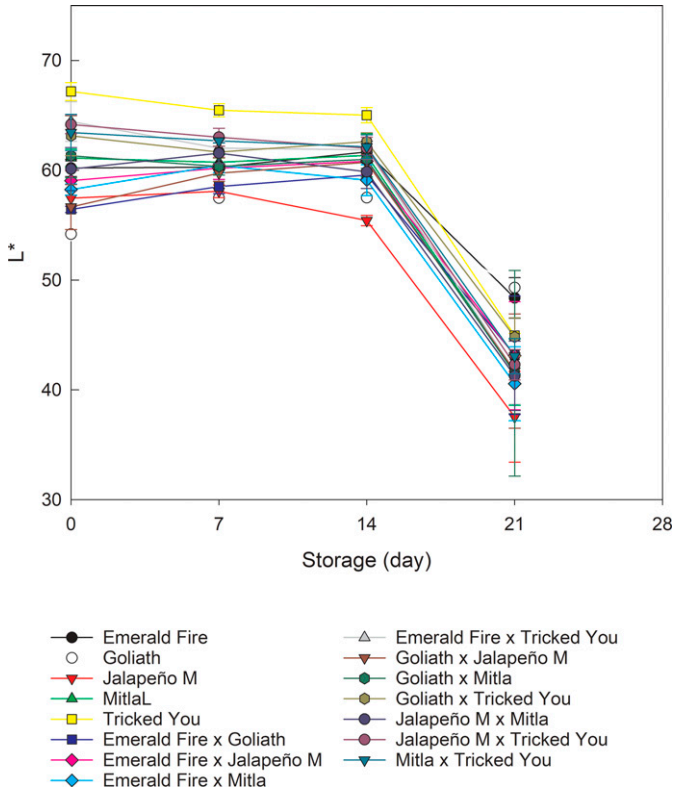

Fig. 3. The L* color space value for fresh-cut jalapeño pepper slices from five cultivars and 10 intercrosses of these cultivars stored at 5 °C for up to 21 d (n = 3). Bars represent the pooled *SE* of the mean.

of work per unit volume (Peleg 2019; Vincent 1990). During this study, genotypes that had harder flesh generally had tougher skin, with the exception of 'Jalapeño M' (Supplemental Table 1). 'Jalapeño M' was hard, comparable to other genotypes, but it had the lowest skin strength or toughness. These combined negative factors may have contributed to the difficulty measuring the texture of 'Jalapeño M' at 21 d, when it exhibited severe deterioration.

Visual quality by color transformation using image analysis

Color systems are useful parameters that are widely used to monitor the change in fresh-cut pepper color during storage (Barbosa et al. 2020). Changes of color and texture during storage are associated with biochemical changes in tissue (Sethu et al. 1996). Discoloration in green tissue is caused by chlorophyll degradation, whereas texture change is mainly caused by water loss (Barrett et al. 2010; Giannakourou and Tsironi 2021; Toivonen and Brummell. 2008). During our study, L* values for fresh-cut jalapeño remained consistent until day 14 of the fresh-cut product storage period; however, they declined rapidly between day 14 and day 21, indicating that the samples became darker in color. An analysis of variance showed that there were no significant differences among genotypes at day 14, except for 'Jalapeño M', which showed a significantly low L* value (Fig. 3; Supplemental Table 2). In comparison with other genotypes, 'Tricked You' fresh-cut product maintained higher L* values up to 14 d of storage, but they subsequently declined, coincident with the declines observed in other genotypes. Pepper fruits are susceptible to chilling injury (CI) below 7 °C, and symptoms vary considerably depending on the cultivar and fruit maturity (Lim et al. 2007). The surface pitting characteristic of CI in pepper fruits was not observed in our fresh-cut product. However, the observed decrease in tissue lightness is consistent with tissue senescence that may occur with pepper CI (Hanaei et al. 2022).

Fresh-cut a* values continued to increase slightly over storage time, and only 'Goliath' had a significantly low a* value at day 14. This indicated that 'Goliath' maintained its greenness, whereas the other genotypes became slightly less green and slightly more reddish. The b* values showed a slight decline over time, meaning that the samples changed their colors from slightly yellowish to more bluish. Tricked You and Jalapeño M exhibited significant declines in b* values between 14 and 21 d of storage relative to other cultivars for which b* was stable during latter days of storage. In comparison with other cultivars, Jalapeño M and Goliath exhibited the highest and lowest b* values, respectively, during up to 14 d of storage. Fruit color saturation also declined over the 21-d storage period, as evidenced by the declining chroma values (Supplemental Table 2). Despite its poor fresh-cut quality, as assessed by EL, 'Jalapeño M' maintained fruit color saturation through 14 d of storage, whereas other cultivars exhibited significant decreases in chroma after 7 or 14 d of storage.

Tissue softening, measured as the proportion of the pepper slice in the cross-section that is damaged or unhealthy tissue softened by deterioration, increased slightly until day 14. There was no significant difference between day 7 and day 14. After day 14, tissue softening increased rapidly. However, 'Jalapeño M' deteriorated exceptionally rapidly throughout the storage period (Fig. 4A).

Lignification is the process by which the mesocarp of peppers becomes lignified or hardened because of water loss and the formation of lignin. Lignification of wounded tissue eventually affects the texture of the fresh-cut pepper product (Luo et al. 2012; Zhang et al. 2021). In this study, lignification increased rapidly after processing until day 7, and then it slightly increased until day 14. After that, it decreased. 'Jalapeño M' lignification scores were significantly lower than those of other genotypes (Fig. 4B).

Correlation between visual quality and instrumental quality parameters

The assessment standard for the freshness of fresh-cut product has been regarded as EL. This study identified several

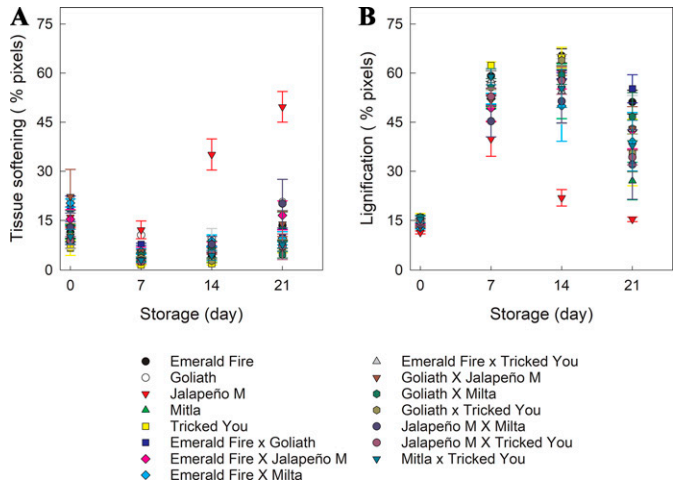

Fig. 4. Tissue softening (**A**) and lignification (**B**) classified using image analysis of fresh-cut jalapeño pepper slices from five cultivars and 10 intercrosses of these cultivars stored at 5 °C for up to 21 d (n = 3). Bars represent the pooled *SE* of the mean.

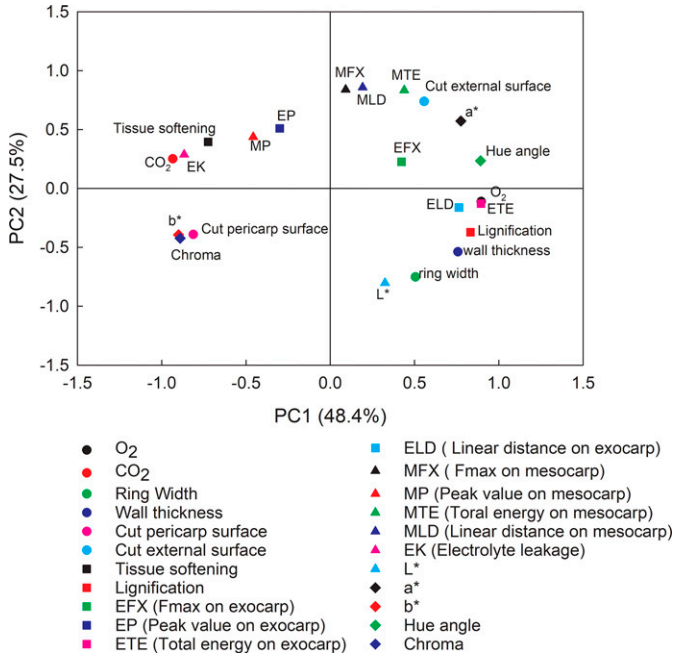

Fig. 5. Principal component analysis with biplots for 22 visual and instrumental measurements of fresh-cut quality parameters evaluated on jalapeño pepper slices after 14 d of storage at 5 °C. See Table 1 for definitions of texture profile acronyms.

parameters among the 22 variables studied that have a strong correlation with EL: CO_2 (r = 0.96; $P < 0.0001$), tissue softening (r = 0.93; $P < 0.0001$); lignification (r = −0.96; $P < 0.0001$), ring width (r = −0.76), and ETE (r = −0.69). The PCA supported a correlation between the parameters (Fig. 5). The CO_2 and tissue softening were located very closely together in the second quadrant, indicating that these parameters are strongly correlated to each other. Lignification occupied the fourth quadrant, directly opposite EL and tissue softening from origin, indicating a strong but negative relationship with EL and tissue softening. Ring width, wall thickness, ETE, and ELD were also negatively correlated with EL. Fresh-cut products of jalapeño genotypes with a larger ring width, which is associated with a larger fruit size, were less likely to deteriorate over the storage period. Genotypes with thicker walls were also less likely to deteriorate. Increased skin strength evidenced by ETE and ELD scores similarly reduced product deterioration. These are important parameters for assessing the freshness of fresh-cut jalapeños peppers. These relationships suggest that fruit size, wall thickness, and skin toughness might be useful as predictive measures in the field for the selection of jalapeño genotypes with superior fresh-cut quality.

A hierarchical cluster analysis grouped the 15 genotypes into five clusters based on the parameters evaluated after 14 d of storage. Cluster I (brown color group) consisted of one cultivar, Goliath, and one hybrid line, Emerald Fire × Goliath. Cluster II (red color group) consisted of one cultivar, Emerald Fire, and the following four hybrid lines: Emerald Fire × Jalapeño M, Goliath × Mitla, Goliath × Tricked You, and Goliath × Jalapeño M. Cluster III (green color group) comprised one cultivar, Tricked You, and the following three hybrid lines: Emerald Fire × Tricked You, Jalapeño M × Tricked You, and Mitla × Tricked You. Cluster IV (blue color group) consisted of one cultivar, Mitla, and the following two hybrid lines: Emerald Fire × Mitla

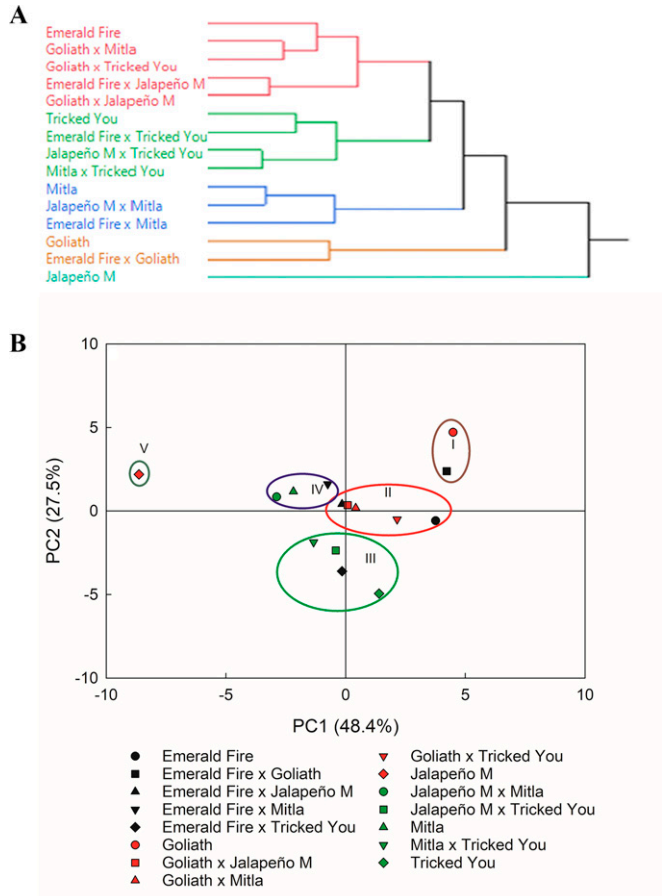

Fig. 6. Hierarchical cluster analysis (**A**) and principal component analysis with biplots (**B**) of five jalapeño pepper cultivars and 10 intercrosses of these cultivars evaluated using 22 fresh-cut quality parameters after 14 d of storage at 5 °C.

and Jalapeño M × Mitla. Cluster V (dark green color group) contained one cultivar, Jalapeño M (Fig. 6A). Clusters reflected the influence of the cultivar on the fresh-cut performance of hybrid combinations. Cultivars generally grouped with their hybrids as a function of individual cultivar fresh-cut performance.

The dendrogram shows each cluster and the level of similarity between each genotype, as determined by the distance of the horizontal line in the dendrogram (Fig. 6A). Variables such as tissue softening ($r^2 = 0.95$), EL ($r^2 = 0.95$), total energy of the mesocarp ($r^2 = 0.95$), CO_2 ($r^2 = 0.94$), and lignification ($r^2 = 0.90$) have a very strong association with the clusters. Firmness ($r^2 = 0.77$), especially tissue hardness (MFX), O_2 ($r^2 = 0.75$), Chroma and L* ($r^2 = 0.72$, respectively), and wall thickness, a*, and b* ($r^2 = 0.71$, respectively) have strong associations with the clusters.

To investigate the characteristics of genotypes that grouped within each cluster, a PCA with biplots was performed (Fig. 6B). The first principal component (PC1), which explained 48.4% of the variance, is represented by tissue deterioration indicators such as the O_2 and CO_2 concentrations, a*, b*, and EL. The second principal component (PC2), which explained 27.5% of the variance, is represented by firmness-related indicators such as MP, MLD, MFX, and MTE. In the PCA plot, the greater the positivity of the PC1 scores, which are indicative of less deterioration, the better the fresh-cut product quality of the genotype. The greater

the positivity of the PC2 scores, which are indicative of the sample firmness, the better the fresh-cut product quality of the genotype.

The cultivar Goliath and the hybrid Emerald Fire × Goliath in cluster I, located in the middle of first quadrant with high PC1 and PC2 scores, were superior to other genotypes in terms of stored fresh-cut product quality. The prevalence of 'Goliath' hybrids in clusters I and II suggests that 'Goliath' has a positive influence on reducing product deterioration in hybrid combinations with other cultivars. Various instrumental measurements confirmed that these genotypes are well-suited for fresh-cut product applications. Tricked You was the least firm cultivar, but its poor firmness was mitigated in hybrid combinations. Jalapeño M in cluster V, located at the far left side of PC 1, where the parameters EL, CO_2, and tissue softening are located, was the worst cultivar in terms of fresh-cut product quality. In comparison with other cultivars, Jalapeño M had the smallest physical size, a rapid decline in the package shelf-life, accumulated CO_2 partial pressures, increased EL, and rapid tissue softening (Figs. 5 and 6B). In hybrid combinations, other cultivars mitigated the negative effects of Jalapeño M.

Clusters I and V are distinctively different from the other three clusters. However, the other three clusters can be considered either similar or different, depending on the analytical needs or research objectives. As the dendrogram shows, these three clusters are closely grouped. Several instrumental measurements also indicated that 12 genotypes in these clusters did not show any statistical difference among them. Despite their similarities, each cluster has distinct features. The five genotypes in cluster II, which are hybrids of 'Goliath' or 'Emerald Fire', appear to be the second-best group for fresh-cut product. They have the second highest PC1 scores and better PC2 scores than cluster III. Cluster III is distinctively composed of four hybrids of 'Tricked You'. 'Tricked You' and its hybrids showed less firmness than the other genotypes. Cluster IV is distinctively composed of 'Milta' and its hybrids.

In summary, all jalapeño genotypes, except 'Jalapeño M', exhibited promising potential as suitable candidates for fresh-cut products. These genotypes demonstrated excellent quality retention for storage periods up to 14 d, with some genotypes maintaining quality for up to 21 d. These findings corroborate earlier research that also highlighted the superior ability of jalapeño genotypes to maintain stable EL levels when compared with sweet bell pepper and other pepper classes (Delwiche et al. 2019; Stommel et al. 2016b) and provide useful insights into the value of measures beyond EL to assess fresh-cut product tissue integrity. Measurements of tissue softening via imaging analysis and physical analyses of tissue firmness are also useful indices of the freshness of fresh-cut jalapeños. Our results suggest that fruit size, wall thickness, and skin toughness might be useful as predictive measures in the field for the selection of jalapeño genotypes with superior fresh-cut quality. Among the five jalapeño cultivars evaluated here, the two that stand out as the most optimal choices for fresh-cut applications are 'Goliath' and 'Emerald Fire'. Remarkably, their hybrids displayed significantly enhanced quality, even in hybrid combinations with inferior cultivars, including Jalapeño M. This evidence underscores the potential of 'Goliath' and 'Emerald Fire' as superior parents for transmitting desirable fresh-cut quality traits to their progeny.

This research provides valuable information to plant breeders, the fresh-cut industry, and those involved in creating a jalapeño fresh-cut market and will help to ensure that fresh-cut jalapeño products remain of high quality and meet consumer expectations. It is hoped that this research will facilitate the development of superior jalapeño cultivars for fresh-cut applications, stimulate further exploration into the potential of jalapeño peppers for fresh-cut products, and help sustain continued growth of the fresh-cut industry.

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