THE JOURNAL OF AGRICULTURE
OF THE UNIVERSITY OF PUERTO RICO

Issued biannually by the Agricultural Experiment Station of the University of Puerto Rico, Mayagüez Campus, for the publication of articles and research notes by staff members or others, dealing with scientific agriculture in Puerto Rico and elsewhere in the Caribbean Basin and Latin America.

VOL. 107 2023 No. 2

Characterization and post-harvest management of *Cucurbita moschata* Duchesne exhibiting a novel yellow color and grown as a "baby" summer squash[1,2]

Julirys Guzmán-Laureano[3], *Rosa N. Chávez-Jáuregui*[4], *and Linda Wessel-Beaver*[5]

J. Agric. Univ. P.R. 107(2):75-97 (2023)

ABSTRACT

Although immature fruit of the domesticated species of *Cucurbita* (pumpkin and squash) are edible, it is usually fruit of *C. pepo* L. that are harvested as summer (immature) squash. *Cucurbita pepo* is not widely cultivated in Puerto Rico because of its susceptibility to a variety of diseases. By contrast, *C. moschata* Duchesne (tropical pumpkin) is well adapted and widely grown on the island but is used almost exclusively as a winter squash, harvested at maturity. We conducted physicochemical, nutritional, and sensory analyses of fresh and stored immature fruits of recently developed tropical pumpkin lines with a novel yellow color to determine their potential use as miniature ("baby") summer squash. Two-d-old fruits were stored at 5° C for 14 d or at 10° C for 16 d. The nutritional composition of fresh 2-d-old fruits was like that of *C. pepo* summer squash. Changes in O_2 and CO_2

---

[1] Manuscript submitted to Editorial Board 9 February 2023.

[2] This research was supported by funds from the United States Department of Agriculture, National Institute of Food and Agriculture (NIFA), Hatch Program Project 1003571. A portion of this research was conducted in partial fulfillment of the requirements for a M.S. degree by the first author.

[3] Former graduate student, Food Science and Technology Program, College of Agricultural Sciences, University of Puerto Rico, Mayagüez Campus, Mayagüez, PR.

[4] Professor, Department of Agro-environmental Sciences, College of Agricultural Sciences, University of Puerto Rico, Mayagüez Campus, Mayagüez, PR.

[5] Professor Ad-Honorem, Department of Agro-environmental Sciences, College of Agricultural Sciences, University of Puerto Rico, Mayagüez Campus, Mayagüez, PR. *Corresponding author. email: lindawessel.beaver@upr.edu.

composition within sealed bags, and physical and chemical characteristics were measured before and after storage. The effect of fruit age (1- to 5-d-old fruit) on weight and color was evaluated. A sensory panel evaluated the general appearance and flavor of immature tropical pumpkin fruits. Over the storage period, O_2 decreased, and CO_2 increased within sealed bags. The increase in CO_2 was greater at 10° C. Weight loss during storage ranged from 2.22% to 3.11%. There were decreases in luminosity and hue angle, but not chroma; fruits became more difficult to penetrate after storage. Fruits exhibited a decrease in pH, acidity, vitamin C, beta-carotene and antioxidant activity, and an increase in soluble solids and phenolics during storage. The effect of storage temperature on chemical characteristics was significant only for acidity, beta-carotene, and antioxidant activity; there were minimal changes in fruits stored at 10° C. Among 1- to 5-d-old fruit, older fruits tended to have a lower luminosity, with a somewhat more saturated orange color (increased chroma and decreased hue angle), compared to younger fruits. Panelists' reaction to the appearance of whole, uncooked fruit ranged from "slightly like" to "extremely like". The evaluation of the flavor of cooked pieces of fruit ranged from "moderately dislike" to "extremely like", with 82% of participants rating cooked fruit as "slightly like" or better. Tropical pumpkin in its immature state is a fruit with nutritional value, an attractive appearance and a flavor likely to be accepted by consumers. Two-day-old fruit stored at 10° C maintained better quality than did fruit stored at 5° C. The immature fruit of these uniquely colored tropical pumpkin lines are a potentially high-value product for Puerto Rico or other areas where *C. pepo* cannot be grown.

Keywords: *Cucurbita pepo,* tropical pumpkin, miniature vegetables, fruit storage

RESUMEN

Caracterización y manejo postcosecha de *Cucurbita moschata* Duchesne que exhibe un novedoso color amarillo y se cultiva como un calabacín "bebé"

Aunque los frutos inmaduros de especies domesticadas de *Cucurbita* (los calabacines y calabazas) son comestibles, casi siempre los frutos que son utilizados en su estado inmaduro son los de *C. pepo*, los cuales son llamados calabacines (*summer squash* en inglés). *Cucurbita pepo* (calabacín) no se cultiva ampliamente en Puerto Rico debido a su susceptibilidad a una variedad de enfermedades. La calabaza común de Puerto Rico (*C. moschata* Duchesne; *tropical pumpkin* en inglés) se cultiva extensamente en la isla como un tipo de *winter squash* y se cosecha en su estado maduro. Realizamos análisis fisicoquímicos, nutricionales y sensoriales de frutos inmaduros de nuevas líneas de calabaza que exhiben un color amarillo para determinar su potencial como un calabacín miniatura o "bebé". Para estudiar el efecto del tiempo de almacenamiento, frutos de 2 d de edad se almacenaron a 5° C por un periodo de 14 d, o a 10° C por 16 d. La composición nutricional de frutos inmaduros frescos de calabaza fue parecida a la del calabacín *C. pepo.* Antes y después del almacenamiento, se midieron los cambios en la composición de O_2 y CO_2 dentro de bolsas selladas y las características físicas y químicas de los frutos. Se evaluó el efecto de la edad del fruto (frutos de 1 a 5 d de edad) sobre el peso y color de los frutos. Un panel sensorial evaluó la apariencia y la aceptación general de frutos inmaduros de calabaza. A través del periodo de almacenamiento, el O_2 disminuyó y el CO_2 aumentó en las bolsas selladas. El aumento en CO_2 fue mayor a los 10° C. La pérdida de peso durante el almacenamiento varió de

2.22% a 3.11%. Hubo disminuciones en luminosidad y el ángulo de matiz, pero no en croma, y los frutos fueron más difíciles de penetrar luego del almacenamiento. Durante el almacenamiento, los frutos exhibieron una disminución en pH, acidez, vitamina C, betacaroteno y la capacidad antioxidante y un aumento en sólidos solubles y compuestos fenólicos. El efecto de la temperatura de almacenamiento en las características químicas fue significativo para la acidez, betacaroteno y la capacidad antioxidante; hubo cambios mínimos en los frutos almacenados a 10° C. Entre las frutas de 1 a 5 d de edad, las frutas más viejas tendieron a tener luminosidad más baja, con un color naranja algo más saturado (croma aumentado y ángulo de matiz disminuido), en comparación con las frutas más jóvenes. La opinión del panel sensorial sobre la apariencia general del fruto varió desde "gusta ligeramente" a "gusta extremadamente". La aceptación general de sabor de frutos cocidos fluctuó desde "no gusta ligeramente" hasta "gusta extremadamente", con 82% de los participantes indicando "gusta ligeramente" o mejor. La calabaza en su estado inmaduro es un fruto con valor nutricional, con una apariencia atractiva y con una aceptación general del consumidor. Los frutos de 2 d de edad almacenados a 10° C retuvieron mejor sus características químicas durante el almacenamiento que los almacenados a 5° C. El fruto inmaduro de estas nuevas líneas de calabaza con un color único es un potencial producto de alto valor para Puerto Rico u otras áreas donde *C. pepo* no se cultiva.

Palabras clave: *Cucurbita pepo,* calabaza, calabacín, hortalizas miniaturas, almacenamiento de frutos

INTRODUCTION

High quality fruits of summer squash are typically harvested about seven days after flowering and are characterized by their smooth, thin, shiny skin and firm, but tender, texture (McCollum, 2016). Summer squash can be harvested from any of the five domesticated species of *Cucurbita*; however, most commercially produced summer squash cultivars are *C. pepo*.

Postharvest storage practices for summer squash have been studied by several researchers. Summer squash has been shown to be susceptible to chilling injury when stored at temperatures <5° C (Massolo et al., 2019; McCollum, 1989, 1990; Megías et al., 2015, 2017; Nunes et al., 2003; Sherman et al., 1985), although the degree of susceptibility can vary depending on the cultivar (Megías et al., 2014, 2015, 2017). McCollum (2016) recommended summer squash be stored at 5 to 10 °C while Nunes et al. (2003) concluded that the best storage temperature was 10 to 15 °C.

While most summer squash is harvested about 7 d after flowering, there has been increasing interest in the past decade and a half in marketing miniature or "baby" vegetables, including baby squash. Summer squash used for this purpose is harvested from flowering through several days after flowering. Baby squash is more sensitive to chilling injury than summer squash harvested at a larger size. Brew et al. (2006) reported that, compared to baby squash stored at 5 or 7 °C for 14 d, fruits stored at 10° C maintained better quality. After 14 d fruits exhibited damage due primarily to microbial growth.

Most cultivars of summer squash are adapted to temperate zones or, in the tropics, to high elevations (>1000 m). In the lowland humid tropics of Puerto Rico and similar locations, *C. pepo* is seldom produced on a commercial scale because of susceptibility to a variety of diseases. By contrast, tropical pumpkin (*C. moschata*) is widely planted in Puerto Rico and other areas of the humid lowland tropics, but as a winter (mature) squash. The breeding program of the Agricultural Experiment Station (AES) of the University of Puerto Rico at Mayagüez (UPRM) has developed various lines of tropical pumpkin that exhibit unique yellow skin colors. Many cultivars of *C. pepo* summer squash also produce yellow fruit, and a mix of yellow and green summer squash is attractive in prepared dishes. The overall objective of our research was to study the potential for harvesting immature fruit of newly developed, yellow-skinned lines of tropical pumpkin as a substitute for *C. pepo* baby summer squash. More specifically, we determined the nutritional value of immature tropical pumpkin fruits, studied the physical and chemical changes in immature fruits of tropical pumpkin during storage, evaluated the changes that occur as fruits mature from anthesis through 5 d of age, and presented fruit samples to a sensory panel.

MATERIALS AND METHODS

Collection of fruit samples. Sixteen lines of tropical pumpkin that produce unique yellow fruit were transplanted to the field at the Lajas Substation of the Agricultural Experiment Station, University of Puerto Rico at Mayagüez (AES-UPRM) on 13 February 2018 using a randomized complete block design with two replications. Plots consisted of a single row of five plants spaced 1.8 m apart on raised beds with drip irrigation and covered with grey plastic mulch. Prior to transplanting, 45 kg/ha of N and P, and 36 kg/ha of K were broadcast and incorporated. Later on, 45 kg/ha of N and P, and 36 kg/ha of K was supplied via drip irrigation. Once flowering commenced (late April 2018), immature fruits were harvested three times a week for five weeks to supply enough fruits for our study. Fruit age at harvest was determined by the state of the corolla (very slightly deteriorated at 1 d after flowering to completely deteriorated or fallen off at 5 d after flowering). Fruits were transported in egg cartons to the laboratory at UPRM. Corollas were removed, then fruits were washed with distilled water and gently cleaned with paper towels (Kimwipes, Kimberly-Clark Corporation, Irving, TX)[6]. To disinfect, fruits were placed in a 0.02% sodium hypochlorite solution for 3 min, then washed one more time with distilled water.

Proximate and mineral analyses. A random sample of five to seven 2-d-old fruits from the Lajas planting was harvested on each of three different dates in May 2018. At each harvest date, fruits were dried at 70° C for 24 h in a conventional oven, then ground to homogenize the sample. A proximate analysis (macronutrient content) was carried out using AOAC method 925.04 for moisture, method 923.03 for ash, method 991.20 for protein, method 992.16 for total fiber, 991.43 for dietary fiber, and method 920.39 for fat (AOAC International, 2012). Mineral content was determined following PerkinElmer (1996). Samples of 0.4 g were incinerated in crucibles at 500° C for 4 h, then allowed to cool overnight. The incinerated samples were digested with 20 mL of 33% HCl until 10 mL of solution remained in the crucible. After digestion was completed, each sample was filtered through Whatman No. 541 filter paper into a 100 mL volumetric flask. Concentrations of aluminum, boron, calcium, iron, potassium, magnesium, manganese, phosphorus, and zinc were obtained using an inductively coupled plasma optical spectrometer (ICP-OES Optima 8000DV; PerkinElmer, Inc., Waltham, MA).

Changes in O_2 and CO_2 in packaging during storage. Three to six (depending on fruit size) 2-d-old fruit were placed into 20.3 x 20.3 cm plastic bags (FoodSaver®, Jarden Consumer Solutions, Boca Raton, FL) that were then sealed without vacuum (Food Saver® VS3180, Jarden Consumer Solutions, Boca Raton, FL). Bags were stored at 5 or 10 °C for 6 d (first replication or run) or 7 d (second run). For each storage temperature, and during each of the 6 or 7 d of storage, O_2 and CO_2 concentrations in each bag were measured with a gas analyzer (Servomex Company Inc., Brighton, East Sussex, UK.

Changes in physical characteristics during storage. In early May 2018, a pool of 2-d-old fruit (2 d post-flowering) was harvested from all the experimental lines in Lajas then transported and cleaned as described above. A random sample of 16 unblemished fruits was selected for evaluation on the day of harvest (day 0) and after 14 d of storage at 5° C. At a second May 2018 harvest date, another pool of 2-d-old fruit was harvested and processed, and a random sample of 40 fruits was selected for evaluation on the day of harvest (day 0) and after 16 d of storage at 10° C. A preliminary study had determined that

---

[6] Company or trade names in this publication are used only to provide specific information. Mention of a company or trade name does not constitute an endorsement by the Agricultural Experiment Station of the University of Puerto Rico, nor is this mention a statement of preference over other equipment or materials.

fruit stored at 10° C could be stored several days longer than those stored at 5° C. An identification number was written on each fruit using a felt-tip pen, and each fruit was evaluated for visual quality, weight and skin color before and after storage at 5 or 10 °C. For storage, fruits were placed in 137 x 178 x 35 mm (length x width x depth) black polypropylene trays with absorbent pads at the bottom of each tray to minimize condensation. Four to six fruits were placed in each tray and trays were sealed with polypropylene film using a packaging machine (KOCH Ultra Source LLC, Kansas City, MO, USA) before being stored. Two persons evaluated visual quality using a 0 to 4 scale, where 0 = no defects or scars, 1 = minor wounds or scars (very slight deterioration), 2 = slight deterioration, 3 = moderate deterioration, and 4 = heavy deterioration (unmarketable). Signs of deterioration included shrinkage, softening, pitting of the skin, and decomposition. Fruits were weighed. Length (from base of stem to base of fruit) and width (at the widest part of the fruit) were measured using a digital caliper (ABSOLUTE Digimatic Caliper Series 500, Mitutoyo, Kanagawa, Japan). A colorimeter (Color Flex EZ, Hunter Associates Laboratory, Inc., Reston, VA) was used to measure L*a*b* tridimensional color space on the fruit surface; a* and b* values were used to calculate hue angle (shade of color) and chroma (color saturation) using formulas from McGuire (1992).

Firmness, chemical and nutritional characteristics. Firmness, pH, acidity, soluble solids, vitamin C, beta-carotene, phenolics, and antioxidant capacity were measured in 2-d-old fruit before and after storage. Fruits were transported, cleaned, and packaged as described for physical characteristics, but the harvesting protocol was slightly different. Harvests of 2-d-old fruit took place on three different dates during May 2018. At each harvest, three unblemished fruits were randomly assigned to each one of the three treatments:(1) fresh fruit at day 0, (2) storage for 14 d at 5° C and (3) storage for 16 d at 10° C. Each date was considered a block (replication). Fruit firmness was measured with a texture analyzer (TA.XT*Plus*, Stable Micro Systems Ltd., Surrey, UK) using a cylindrical 4-mm diameter probe, a penetration depth of 7 mm and a speed of 0.83 mm/s (Martínez-Valdivieso et al., 2015).

Once firmness was measured, the three fruits of a treatment replication were macerated in a food processor in bulk. The juice was used to determine pH (AOAC method 982.12), titratable acidity (AOAC method 942.15), soluble solids concentration [AOAC method 932.14 (C) using a refractometer, and expressed as °Brix] and vitamin C (ascorbic acid) using the 2,6-dichloroindophenol (DCIP) titrimetric method (AOAC Method 967.21) (AOAC International, 2012).

Beta-carotene content was determined using the method proposed by Nagata and Yamashita (1992): 1.0 g of macerated pulp was homogenized with 10 to 20 mL of acetone:hexane (4:6) solvent by mixing for 2 min with a stand homogenizer (Polytron PT 2500 E, Kinematica Inc., New York, NY) set at 18,000 rpm. The absorbency of the supernatant was measured at wavelengths of 663, 645, 505 and 453 nm using a spectrometer (UV-3100PC Spectrophotometer, VWR International, Leuven, Belgium). Beta-carotene content was determined using the following formula: β-carotene (mg/100 mL) = $0.216A_{663} - 1.22A_{645} - 0.304A_{505} + 0.452A_{453}$. Results were converted to mg/g of fresh weight (FW).

Samples used to determine total phenolics and antioxidant capacity were prepared following Granciero (2015). The remaining macerated pulp was lyophilized. For phenolic content, a 0.5 g sample of lyophilized pulp was diluted with 10 mL of distilled water and placed in a shaker for 30 minutes. The Folin-Ciocalteau (FC) colorimetric method of Singleton and Rossi (1965) was used to measure phenolics content. Readings were reported in milligrams of gallic acid equivalents (GAE)/100 g FW. The 2,2-diphenyl-1-picryl-hydrazyl-hydrate (DPPH) method, described by Brand-Williams et al. (1995), was used to measure antioxidant capacity. Antioxidant capacity was reported as micrograms of Trolox equivalents/g FW. For each treatment at each harvest date measurements were taken in triplicate and then averaged for that date.

Effect of fruit age on physical characteristics. Samples of 1- to 5-d-old fruit were harvested in plots of six of the 16 experimental lines at the Lajas substation and transported and handled as described above. Within a line, two to 13 fruits (usually three to four) were sampled for each fruit age. Fruits were evaluated in the laboratory approximately 4 h after harvest. Fruit weight and color (luminosity, hue angle and chroma measured as described earlier) were determined.

Sensory panel. General appearance and flavor of 2-d-old fruit harvested from the experimental lines at the Lajas substation were evaluated by a panel of 100 volunteers consisting of students and staff at UPRM who indicated that they liked vegetables. Appearance and flavor were evaluated on separate days. For appearance, each panelist was presented with a fresh 2-d-old fruit. For taste, 2-d-old fruits were boiled in unsalted water, and panelists were presented with two pieces of cooked fruit, approximately 2.54 x 2.54 cm in size. For both appearance and taste, panelists rated fruit samples on a nine-point scale, where 1 = extremely dislike, 2 = dislike very much, 3 = moderately dislike, 4 = slightly dislike, 5 = neither like nor dislike, 6 = slightly like, 7 = moderately like, 8 = like very much, and 9 = extremely like.

Statistical analyses. All analyses were carried out using InfoStat (version 2018, Di Rienzo et al., 2018). Changes in O_2 and CO_2 during storage were analyzed by analysis of variance (ANOVA) of a complete block design with two replications (runs). A separate ANOVA was carried out for each storage temperature. A paired t-test (16 pairs for storage at 5° C; 40 pairs for storage at 10° C) was used to study the effect of storage (before vs. after) on physical characteristics. Firmness and chemical characteristics were analyzed using a randomized complete block design with three replications (fruit harvest dates). Single-degree-of-freedom contrasts were used to compare means for (1) before vs. after storage, and (2) storage at 5° C vs. 10° C. Regression was used to analyze the effect of fruit age from 1 to 5 d after flowering on fruit weight, diameter, length, L* (luminosity), hue angle, and chroma. For the study on the effect of fruit age (1 to 5 d after flowering), data was collected from only six of the 16 experimental lines and each line was sampled and analyzed separately. Linear and quadradic regressions were carried out and graphs of the best fitting models are presented.

RESULTS

Fruit description: The experimental lines of tropical pumpkin produced fruits with a variety of shapes including globe (flattened), round and ovoid (Figure 1). Although most lines consistently produced fruits with a yellow color, plants within some lines segregated for fruit color producing yellow, bicolor (yellow and green) and green fruit. Only yellow fruits were harvested and evaluated in this experiment, but the tone of the color was variable from light to bright yellow. Stem color also varied both within and among lines: some lines produced fruits with yellow stems while others had green stems.

Proximate and mineral analyses. Fruits of 2-d-old tropical pumpkin produced a proximate and mineral analysis similar to that of the USDA's analysis for raw zucchini summer squash (U.S. Department of Agriculture, 2018) (Tables 1 and 2). The higher moisture content in our fruits was likely due to the fruits' young age (2 d). Although the documentation doesn't specify the age of fruits analyzed by the USDA, zucchini is typically harvested when fruits are closer to 7-d-old. We observed a higher percentage of fat and a lower percentage of protein than in the USDA standard for zucchini. However, summer squash, in general, is not an important source of either of those nutritional components. The mineral analysis of immature tropical pumpkin was even more similar to zucchini, with the exception of Mn, which was much lower in tropical pumpkin than in zucchini.

FIGURE 1. Examples of stem and skin color and of fruit shape of immature fruits from lines of tropical pumpkin (*Cucurbita moschata*) with a novel yellow color. The age of fruits ranges from 1 to 8 d after flowering.

Changes in O_2 and CO_2 in packaging during storage. At both 5 and 10 °C, the percentage of O_2 within sealed packages of fruit dropped quickly between day 0 and day 2 and then remained at 5% or less during the remaining 7 d of the experiment (Figure 2). Over the 7-d storage period, CO_2 increased from 0% to about 40% in packages stored at 5° C and to over 60% for packages at 10° C.

Changes in physical characteristics during storage. Significant changes in fruit quality, weight and color occurred over the storage period of 14 to 16 d (Table 3). Fruit quality deteriorated over time at both 5 and 10 °C, but the change was less at 10° C. Although significant deterioration was evident after storage, it was rated as “slight” or “very

TABLE 1.—*Proximate analysis of immature fruit (2 d post-anthesis) of yellow fruited tropical pumpkin (Cucurbita moschata) harvested at Lajas, Puerto Rico compared to the USDA proximate analysis for raw summer squash* (C. pepo), *with skin (US Department of Agriculture, 2018).*

	Analysis of immature tropical pumpkin		USDA standard analysis of summer squash
Component	%	SD	%
Moisture	96.96	± 0.05	94.70
Ash	0.62	± 0.0019	0.58
Fat	0.83	± 0.01	0.32
Protein	0.64	± 0.0012	1.21
Carbohydrates	4.62	± 0.04	3.11
Total fiber	0.33	± 0.0027	—
Dietary fiber	1.41	± 0.10	1.00
Soluble fiber	0.29	± 0.02	—
Insoluble fiber	0.79	± 0.05	—

slight" (quality scores of 2.13 and 1.38 for 5 and 10 °C, respectively). Loss of fruit weight during storage, although significant, was slight: less than 3%. Likewise, color changes, although usually statistically significant, were minor. Fruits were slightly darker (lower luminosity) at the end of the storage period, somewhat more orange in color (decreased hue angle) and, for 5° C only, color was less saturated (lower chroma).

Firmness, chemical and nutritional characteristics. Firmness, pH, total acidity and soluble solids of fruits changed during storage, but storage temperature only affected firmness and total acidity (Table 4).

TABLE 2.—*Mineral analysis of immature fruit (2 d post-anthesis) of yellow-fruited tropical pumpkin (Cucurbita moschata) harvested at Lajas, Puerto Rico, compared to the USDA mineral analysis for raw summer squash* (C. pepo), w*ith skin (U.S. Department of Agriculture, 2018).*

Mineral	Analysis of immature tropical pumpkin (mg/100 g)	USDA standard analysis of summer squash (mg/100 g)
Aluminum	0.19	—
Boron	0.14	—
Calcium	10.02	16
Iron	0.23	0.37
Potassium	247.34	262
Magnesium	16.07	18
Manganese	0.06	0.177
Phosporus	38.70	38
Zinc	0.39	0.32

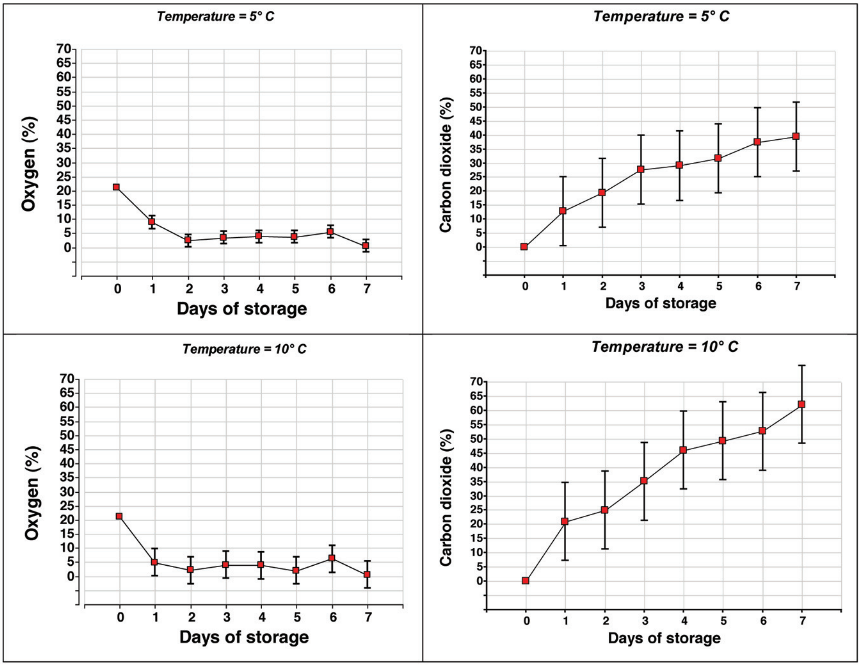


FIGURE 2. Change in percentage of oxygen and carbon dioxide inside packages of 2-d-old tropical pumpkin (*Cucurbita moschata*) fruit sealed in plastic and stored at 5 and 10 °C for 7 d.

More force was needed to penetrate fruits after storage, and this difference was greater at 5° C compared to 10° C. The pH of fruits decreased during storage, but the change was very small and not affected by storage temperature. Total acidity increased, especially at the 5° C storage temperature. Soluble solids increased during storage, and by the same amount under both storage temperatures.

Storage had a marked effect on the nutritional status of fruits. At both storage temperatures vitamin C content after 14 to 16 d of storage was less than half of that of fresh fruits (Table 5). Beta-carotene content after 16 d at 10° C was less than 50% of that of fresh fruit. After being stored at 5° C for 14 d, fruit averaged less than 15% of the beta-carotene of fresh fruit. By contrast, phenolics content increased by about 25% on average in stored fruits. Antioxidant capacity of stored fruit remained at the same level as fresh fruit when stored for 16 d at 10° C, but decreased nearly 20% in fruits stored for 14 d at 5° C.

Effect of fruit age on physical characteristics. The increase in fruit weight from one to five days after flowering closely fit logistic regression models with coefficients of determination (R^2) ranging from 0.89

TABLE 3.—*Means, standard errors (SE), and t-test probabilities for fruit weight and skin color (luminosity, hue angle and chroma) in immature fruit (2 d post-anthesis) of tropical pumpkin (*Cucurbita moschata*) evaluated before storage, and after storage in sealed plastic bags at 5° C (14 d) or 10° C (16 d).*

Treatment	Visual quality[1]	Weight (g)	Luminosity (L*)	Hue angle (°)	Chroma
5° C					
Before storage	0.38	75.81	79.45	81.96	54.85
After 14 d of storage	2.13	74.10	73.93	78.30	49.78
SE	0.17	0.12	0.35	0.34	1.24
t-test[2]	<0.0001	<0.0001	<0.0001	<0.0001	0.0113
10° C					
Before storage	0.01	62.88	79.27	82.39	55.89
After 16 d of storage	1.38	61.01	74.52	78.90	56.78
SE	0.08	0.05	0.20	0.11	0.40
t-test[3]	<0.0001	<0.0001	<0.0001	0.0001	0.1291

[1] A measure of deterioration on a 5-point scale where 0=no deterioration or blemishes on fruit surface, 4=strong deterioration.

[2] Probability of error in a paired t-test with 15 degrees of freedom.

[3] Probability of error in a paired t-test with 39 degrees of freedom.

to 0.95 (Figure 3). Using these models, we estimated that immature fruits increased their weight by 74 to 109% per day, on average, over the first 5 d after flowering. Average fruit weight within the six lines varied from 20 to 32 g for 1-d-old fruit to 264 to 465 g for 5-d-old fruit.

Increasing fruit age resulted in changes in the luminosity of fruit skin in only three of the six lines sampled (Figure 4). Changes in lu-

TABLE 4.—*Means, SE, F test probability, and single-degree-of-freedom linear contrasts for firmness, pH, total acidity and soluble solids of immature fruit (2 d post-anthesis) of tropical pumpkin (*Cucurbita moschata*) evaluated before and after storage in sealed plastic bags at 5° C (14 d) or 10° C (16 d).*

Treatment	Firmness (N)	pH	Total acidity (%)	Soluble solids (%)
Before storage (fresh)	6.35	6.84	0.05	5.1
After storage (14 d, 5° C)	9.59	6.65	0.09	5.5
After storage (16 d, 10° C	7.58	6.64	0.06	5.5
SE	0.2098	0.0212	0.0072	0.0638
F test probability[1]	0.0001	0.0009	0.0243	0.0062
Contrasts[2]:				
Before vs. after storage	0.0001	0.0003	0.0493	0.0022
5 vs. 10 °C (after storage)	0.0005	0.8312	0.0257	0.4881

[1] Degrees of freedom for treatment = 2; degrees of freedom for error = 6.

[2] Reported values are probabilities in F-tests of single-degree-of-freedom linear contrasts using means shown above in the same column.

TABLE 5.—*Means, SE, F test probabilities, and single-degree-of-freedom linear contrasts for nutritional variables of immature fruit (2 d post-anthesis) of tropical pumpkin (*Cucurbita moschata*) evaluated before and after storage in sealed plastic bags at 5° C (14 d) or 10° C (16 d).*

Treatment	Vitamin C (mg/100 g FW)	Beta-carotene (mg/g FW)	Phenolics (mg/100 g FW)	Antioxidant capacity (µg Trolox/g FW)
Before storage (fresh)	6.76	2.96	2,559.78	147.67
After storage (14 d, 5 °C)	3.17	0.39	3,110.89	120.22
After storage (16 d, 10° C	3.16	1.45	3,288.89	146.65
SE	0.3918	0.2403	62.54	1.04
F test probability[1]	0.0009	0.0008	0.0004	0.0001
Contrasts[2]:				
Before vs. after storage	0.0003	0.0004	0.0002	0.0001
5 vs. 10 °C (after storage)	0.9908	0.0209	0.0908	00001

[1] Probabilies in F tests (degrees of freedom for treatment = 2; degrees of freedom for error = 6).

[2] Reported values are probabilities in F-tests of single-degree-of-freedom linear contrasts using means shown above in the same column.

FW = fresh weight.

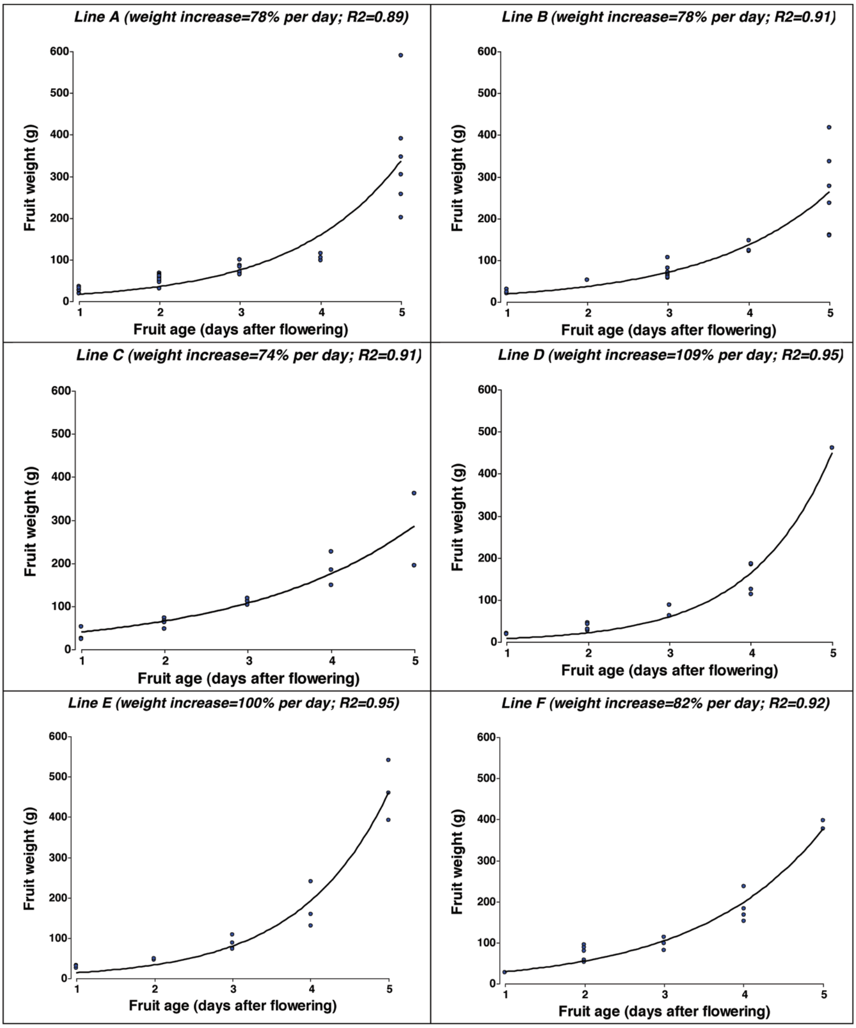
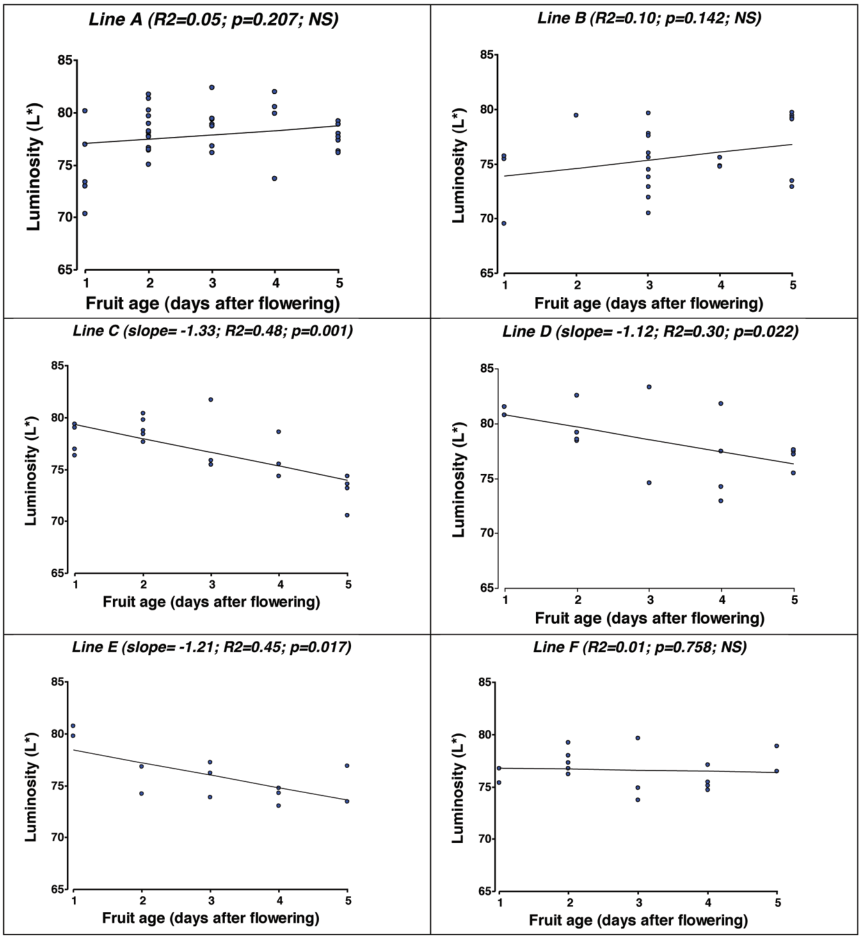


FIGURE 3. Curves represent a logistic regression model of the effect of fruit age (1 to 5 d after flowering) on fruit weight of immature fruits of six lines of tropical pumpkin (*Cucurbita moschata*) with a novel skin color. The increase in weight per day was estimated from the logistic models and all regressions were significant at the 0.0001 probability level. R2 = R^2.

minosity were small: a decrease of 1.12 to 1.33 units per day. Three of the six lines exhibited changes in chroma with increasing fruit age (Figure 5). Additionally, the other three lines also showed a similar, though non-significant, tendency of increased chroma value with increasing fruit age. Greater chroma values reflect an increase in the saturation or



Line A (R2=0.05; p=0.207; NS)

Line B (R2=0.10; p=0.142; NS)

Line C (slope= -1.33; R2=0.48; p=0.001)

Line D (slope= -1.12; R2=0.30; p=0.022)

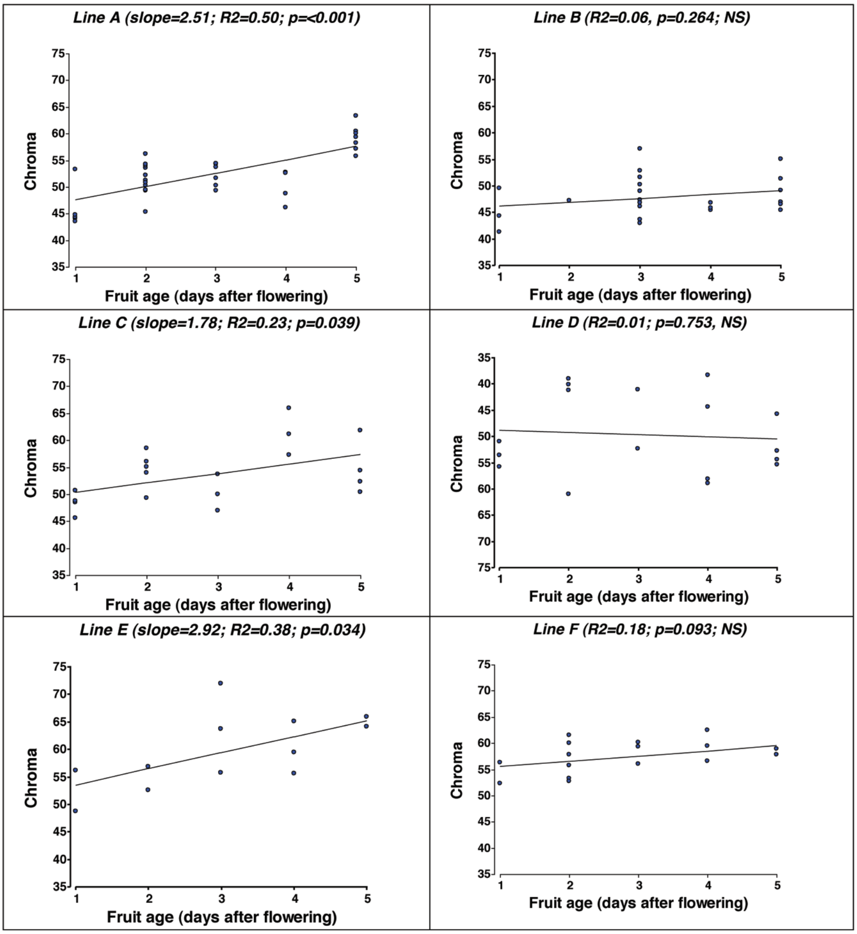
Line E (slope= -1.21; R2=0.45; p=0.017)

Line F (R2=0.01; p=0.758; NS)

FIGURE 4. Regression analysis of the effect of fruit age (1 to 5 d after flowering) on luminosity of skin of immature fruits of six lines of tropical pumpkin (*Cucurbita moschata*) with a novel skin color. The slope in the linear regression analysis is presented for all lines except A for which a quadratic regression gave the best fit. R2 = R^2. NS = not significant at the 0.05 probability level.

purity of skin color. Hue angle decreased in all but one of the lines we sampled, with changes ranging from an angle decrease of 1.93 to 0.65° (Figure 6). A hue angle of 90° corresponds to yellow, while a hue angle of 45° corresponds to orange. Fruit harvested 5 d after flowering had skins that were more yellow-orange compared to that of 1-d-old fruit.

Sensory panel. Panelists had a favorable reaction to both the general appearance of fresh immature fruits and the flavor of cooked immature fruits of yellow fruited tropical pumpkin. All panelists liked the appear-



Line A (slope=2.51; R2=0.50; p=<0.001)

Line B (R2=0.06, p=0.264; NS)

Line C (slope=1.78; R2=0.23; p=0.039)

Line D (R2=0.01; p=0.753, NS)

Line E (slope=2.92; R2=0.38; p=0.034)

Line F (R2=0.18; p=0.093; NS)

FIGURE 5. Regression analysis of the effect of fruit age (1 to 5 d after flowering) on chroma of skin of immature fruits of six lines of tropical pumpkin (*Cucurbita moschata*) with a novel skin color. The slope in the linear regression analysis is presented for all lines except A for which a quadratic regression gave the best fit. R2 = R^2. NS = not significant at the 0.05 probability level.

ance of the fruits (Figure 7). A large majority (82%) of panelists gave a rating of "slightly like" or better when tasting a cooked fruit sample (Figure 8). Only 5% of panelists expressed a dislike for the taste of cooked fruit.

DISCUSSION

The skin color of immature fruit of *C. moschata* typically ranges from pale to dark green, with or without broken stripes of a lighter

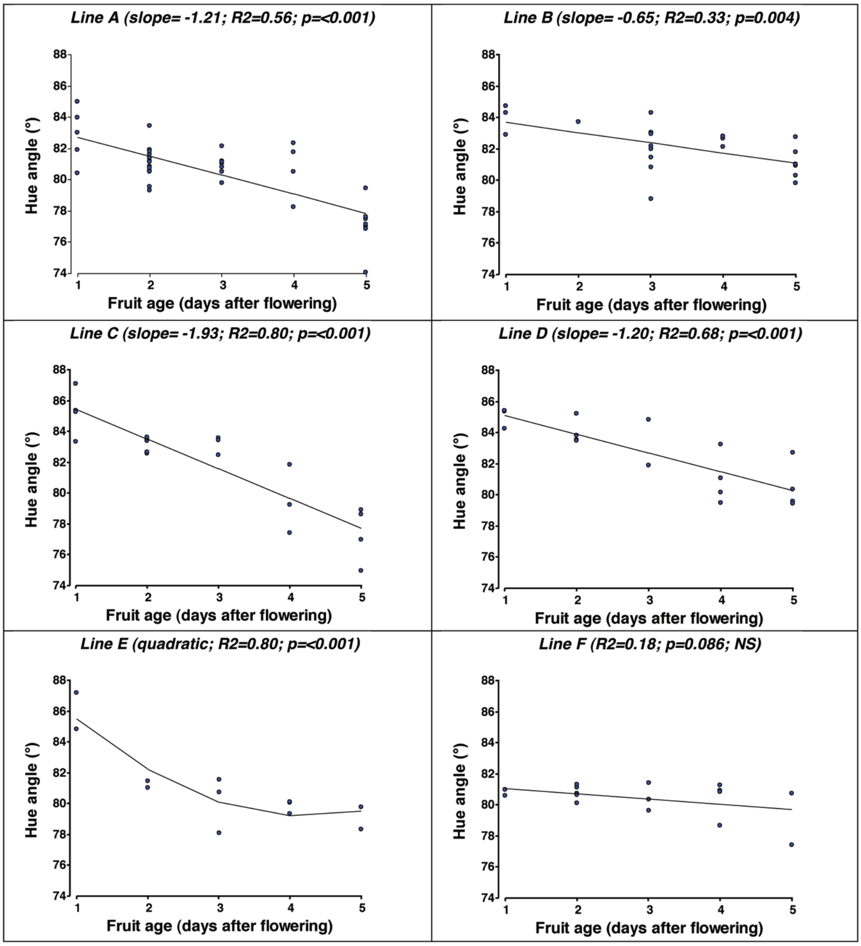


FIGURE 6. Regression analysis of the effect of fruit age (1 to 5 d after flowering) on hue angle of skin of immature fruits of six lines of tropical pumpkin (*Cucurbita moschata*) with a novel skin color. The slope in the linear regression analysis is presented for all lines except A for which a quadratic regression gave the best fit. R2 = R^2. NS = not significant at the 0.05 probability level.

green color. Some of our lines produced fruit with green skin color although most lines produced exclusively yellow fruit (Figure 1). Only yellow fruit was used in our study. The novel yellow skin color of our lines is similar to cultivars of *C. pepo* carrying the *Bicolor* (*B*) gene. The intense yellow color of several *C. pepo* summer squash cultivars is a result of the presence of this gene. Cultivars carrying this gene have "precocious yellow" fruit pigmentation since the yellow color is present well before anthesis (Paris and Brown, 2005). Yellow fruit color

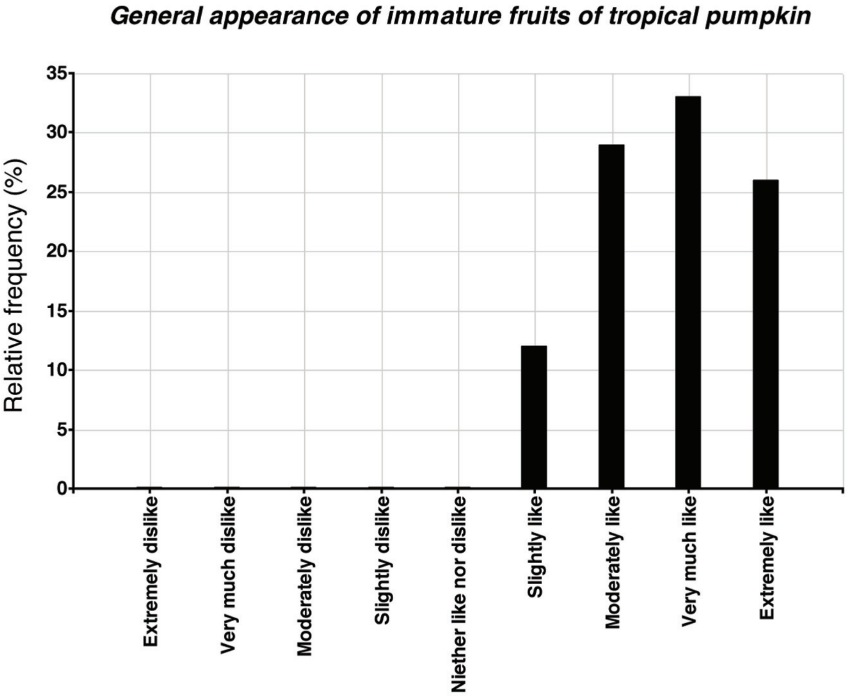


FIGURE 7. Sensory panelists' evaluation of the general appearance of immature fruits (2 d post-anthesis) of tropical pumpkin (*Cucurbita moschata*) with a unique yellow color.

is sometimes expressed only near the stem resulting in a bicolor fruit with yellow at the stem and green at the distal end. Bicolor fruit was evident in some of our lines as well. Both yellow and green stems occurred in our fruits, depending on the line. We judged yellow fruits with dark green stems to be particularly attractive.

The *B* gene has been backcrossed into some genotypes of *C. moschata* but it also is likely that the *B* gene (or a gene specific to *C. moschata* that functions in a similar way) occurs naturally in some landraces of *C. moschata*. The novel yellow color in our lines is derived from a landrace originating from Colombia. In Brazil, Boiteux et al. (2007) developed a bicolor cultivar derived from a bicolor landrace of *C. moschata*. Future studies could clarify the genetics of the novel fruit color in our lines.

The *B* gene appears to have a detrimental effect on the quality of stored fruit. Sherman et al. (1987) evaluated chilling injury in fruits of near-isogenic *C. pepo* lines, with and without the *B* gene, stored at 5° C. Fruits with the *B* gene deteriorated more rapidly. Sherman et al.

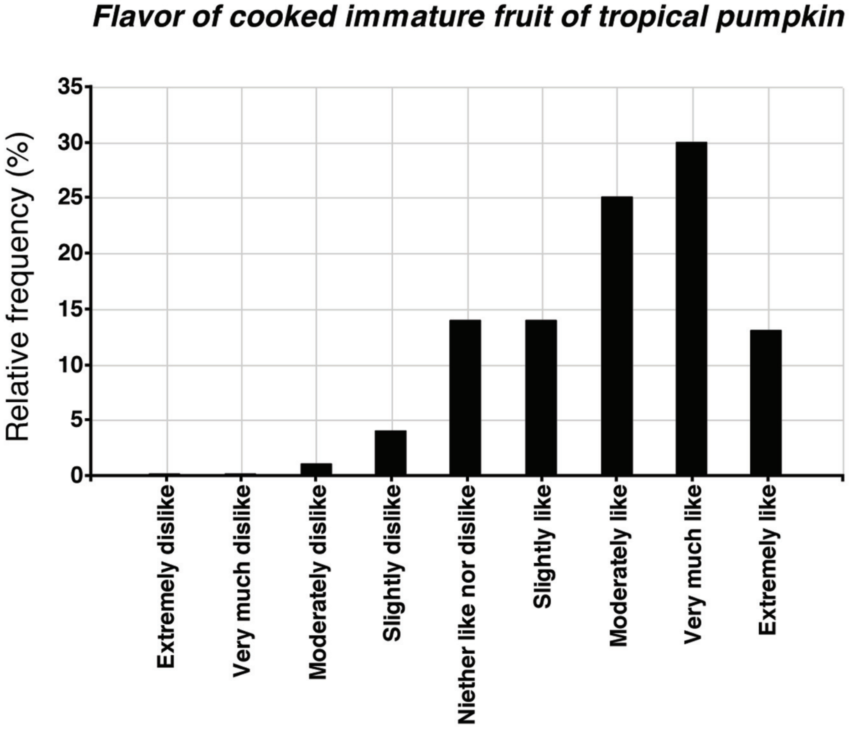


FIGURE 8. Sensory panelists' evaluation of the flavor of immature fruit (2 d post-anthesis) of tropical pumpkin (*Cucurbita moschata*) with a unique yellow color.

(1985) compared regular yellow squash cultivars Goldbar and Seneca Butter with Multipik which carries the *B* gene. 'Multipik' had greater weight loss and a poorer appearance after 5 d of storage at both 5 and 10 °C. McCollum (1990) noted that the presence of the *B* gene increased chilling injury in both scallops and marrows (market classes of *C. pepo* summer squash). Grisales et al. (2020) found that 'Golden', a zucchini with the *B* gene, had more damage during storage than traditional green types.

Our study monitored changes in the concentrations of O_2 and CO_2 in packages during 7 d of storage. Fruit respiration caused O_2 to be consumed and CO_2 to be produced. Respiration was greater at 10° C than at 5° C as is evidenced by the higher concentration of CO_2 after 7 d (Figure 2). High levels of respiration favor deterioration of fruit in storage. The use of active modified atmosphere packaging, where concentration of O_2 is maintained at a lower level and CO_2 at a higher level than normal atmosphere concentrations, has been used successfully in the storage of fruits and vegetables to control respiration. However, Mencarelli et al. (1983) observed that a low concentration of atmospheric O_2 had little to no effect on storage quality of zucchini squash at both chilling and non-chilling temperatures.

Fruit quality of the immature fruits of the *C. moschata* lines in our study deteriorated more rapidly in fruits stored at 5° C than in fruits stored at 10° C. Previous work with cultivars of *C. pepo* also concluded that storage temperatures of about 10° C result in better quality fruit (Balbierz and Kolata, 2015; Brew et al., 2006; Nunes et al., 2003; Sherman et al., 1985). A shiny fruit surface is an important market attribute of summer squash (McCollum, 2016). After 14 to 16 d of storage, we observed only a slight reduction in the luminosity of the fruit surface; likewise, changes in hue and chroma were also small (Table 3). Fruits maintained a reasonable appearance at the end of the storage period at 10° C; on average they were judged to have a visual quality of 1.38 - very slight to slight deterioration. By contrast, there was a marked decrease in nutritional quality of fruits during storage and the decline tended to be greater at 5° C. Vitamin C, beta-carotene and antioxidant capacity all decreased over time (Table 5). Balbierz and Kolota (2015) observed similar changes in their study with scallop squash (*C. pepo*) cultivars. Total phenolics content increased over time in our study. Massolo et al. (2016) also observed this in vegetable marrow (*C. pepo*). The accumulation of phenolics may contribute to fruit deterioration (Massolo et al., 2014).

There are various fruit preservation methods that could be considered in future studies with fruits from our novel lines. Mercado-Ruiz et al. (2010) and Megías et al. (2015) found that chilling injury can be reduced in summer squash by shrink-wrapping individual fruits in polyethylene film before storage. This technique is already commonly used on English cucumbers. Massolo et al. (2014) sprayed summer squash fruits with benzylaminopurine (BAP), a cytokinin, prior to storage at 5° C. Fruits with BAP exhibited less deterioration over a 25-d storage period compared to unsprayed fruits. The use of BAP appeared to decrease pectin solubilization (disassembly) in cells. Carvajal et al. (2015) demonstrated that solubilization and depolymerization of pectin was induced by low temperatures but that this effect was reduced when squash was preconditioned by holding fruit at 15° C for 48 h before storing at 5° C.

Dipping squash into solutions of salicylic acid and sodium nitroprusside was found to be effective in reducing chilling injury (Kannaujia et al., 2019).

The fruits evaluated in our study ranged from “baby” squash (1- to 3-d-old fruit) to a size approaching that of traditional summer squash (4- to 5-d-old fruit). We studied the effect of storage only on 2-d-old fruit, but 3- to 5-d-old fruit would likely maintain even better quality during storage. In all the sampled lines, fruits grew rapidly from 1 to 5 d after flowering (Figure 3). Overall, we judged that the changes in color (including luminosity, hue and chroma) of fruits as they aged from 1 d after flowering to 5 d after flowering did not substantially change, and sometimes improved the marketability of the fruits. Luminosity decreased very slightly (Figure 4), but skin color often became more saturated or pure (increased chroma; Figure 5). Hue angle decreased resulting in an even more attractive yellow-orange color (Figure 6).

Panelists judged immature *C. moschata* fruits with a yellow color as very attractive, and cooked fruits as having a very acceptable flavor (Figures 7 and 8). Only a small percentage of panelists disliked cooked immature *C. moschata* fruits. Although these fruits lose some of their nutritional qualities and deteriorate somewhat in appearance during storage, they maintain marketable quality for about two weeks. In Puerto Rico and other parts of the humid tropics, *C. moschata* is typically harvested at maturity and consumed as a tropical pumpkin (a “winter squash”). Production of traditional *C. pepo* cultivars of summer squash is very challenging in the humid tropics. By contrast, the uniquely colored tropical pumpkin (*C. moschata*) lines evaluated in this study are well-adapted to the humid tropics and offer an opportunity for a new use of tropical pumpkin, either as “baby squash” or traditionally sized summer squash.

LITERATURE CITED

AOAC International, 2012. Official methods of analysis of AOAC International, 19th ed., Gaithersburg, MD, USA.

Balbierz, A. and E. Kolota, 2015. Pre- and postharvest nutritional value and storage ability of scallop squash cultivars. *J. Hort. Res.* 23(2): 105-110 https://doi.org/10.2478/johr-2015-0021.

Boiteux, L.S., W.M. Nascimento, M.E.N. Fonseca, M.M. Lana, A. Reis, J.L Mendonça, J.F. Lopes, and F.J.B. Reifschneider, 2007. ‘Brasileirinha’: cultivar de abóbora (*Cucurbita moschata*) de frutos bicolores com valor ornamental e aptidão para consumo verde. *Horticultura Brasileira* 25: 103-106. https://doi.org/10.1590/S0102-05362007000100020.

Brand-Williams, W., M.E. Cuvelier, and C. Berset, 1995. Use of a free radical method to evaluate antioxidant activity. *LWT-Food Sci. Tech.* 28: 25-30. http://dx.doi.org/10.1016/S0023-6438(95)80008-5

Brew, B.S., A.D. Berry, S.A. Sargent, N.L. Shaw, and D. J. Cantliffe, 2006. Determination of optimum storage conditions for ‘baby’ summer squash fruit (*Cucurbia pepo*). *Proc. Fla. State Hort. Soc.* 119: 343-346.

Carvajal, F., F. Palma, M. Jamilena, and D. Garrido, 2015. Cell wall metabolism and chilling injury during postharvest cold storage in zucchini fruit. *Postharvest Biol. and Tech.* 108: 68-77.

Di Rienzo, J.A., F. Casanoves, M.G. Balzarini, L. González, M. Tablada, and C.W. Robledo, 2018. InfoStat version 2018. Grupo InfoStat, FCA, Universidad Nacional de Córdoba, Argentina. http://www.infostat.com.ar.

Granciero, L.B., 2015. Compostos bioativos e capacidade antioxidante em aboboras-gila (*Cucurbita ficifolia* Bouché). Tesis de maestría. Universidade de Brasilia, Facultad de Ciencias Da Saude, Programa de Pos-graduacao em Nutricao Humana.

Grisales, N.Y., J.C. Henao-Rojas, L.M. Q. Quintero, G. Franco, and J. Jaramillo, 2020. Influencia de las condiciones de almacenamiento sobre la calidad de calabacín (*Cucurbita pepo* L.) en Antioquia – Colombia. Revista Iberoamericana de Tecnología Postcosecha 21(1): 99-112. http://www.redalyc.org/articulo.oa?id=81363356010

Kannaujia, P.K., R. Asrey, A.K. Singh, and E. Varghese, 2019. Postharvest treatments to reduce chilling injury in summer squash (*Cucurbita pepo*) fruits during storage. *Indian J Agric. Sci.* 89(10): 1633-1637.

Martínez-Valdivieso, D., P. Gómez, R. Font, A. Alonso-Moraga, and M. del Río-Celestino, 2015. Physical and chemical characterization in fruit from 22 summer squash (*Cucurbita pepo* L.) cultivars. *LWT-Food Sci. Tech.* 64(2): 1225-1233. https://doi.org/10.1016/j.lwt.2015.07.023

Massolo, J.F., M.L. Lemoine, A.R. Chaves, A. Concellón, and A.R. Vicente, 2014. Benzylaminopurine (BAP) treatments delay cell wall degradation and softening, improving quality maintenance of refrigerated summer squash. *Postharvest Biol. Tech.* 93: 122-129.

Massolo, J.F., J.M. Zarauza, J.H. Hasperué, L.M. Rodoni, and A.R. Vicente, 2019. Maturity at harvest and postharvest quality of summer squash. *Pesquisa Agropecuária Brasileira*, v.54, e00133, 2019. DOI: https://doi. org/10.1590/S1678-3921.pab2019.v54.00133.

McCollum, T.G., 1989. Physiological changes in yellow summer squash at chilling and nonchilling temperatures. *HortScience* 24 (4): 633-635.

McCollum, T.G., 1990. Gene B influences susceptibility to chilling injury in *Cucurbita pepo*. *J. Amer. Soc. Hort. Sci.* 115(4): 618-622.

McCollum, T.G., 2016. Squash, pages 556-558. *In*: (K.C. Gross, C.Y. Wang, and M. Saltveit, eds.) The commercial storage of fruits, vegetables, and florist and nursery stocks. USDA-ARS, Agriculture Handbook 66. https://www.ars.usda.gov/arsuserfiles/oc/np/commercialstorage/commercialstorage.pdf

McGuire, R.G., 1992. Reporting of objective color measurements. *HortScience* 27(12): 1254-1255.

Megías, Z., C. Martínez, S. Manzano, A. Barrera, R. Rosales, J.L. Valenzuela, D. Garrido, and M. Jamilena, 2014. Cold-induced ethylene in relation to chilling injury and chilling sensitivity in the non-climacteric fruit of zucchini (*Cucurbita pepo* L.). *Food Sci. Tech.* 57(1): 194-199.

Megías, Z., S. Manzano, C. Martínez, A. García, E. Aguado, D. Garrido, M. del Mar Rebolloso, J. L. Valenzuela, and M. Jamilena, 2017. Postharvest cold tolerance in summer squash and its association with reduced cold-induced ethylene production. *Euphytica* 213: 9. https://doi.org/10.1007/s10681-016-1805-0.

Megías, Z., C. Martínez, S. Manzano, A. García, M. del Mar Rebolloso-Fuentes, D. Garrido, J.L. Valenzuela, and M. Jamilena, 2015. Individual shrink wrapping of zucchini fruit improves postharvest chilling tolerance associated with a reduction in ethylene production and oxidative stress metabolites. *PLoS ONE* 10(7): e0133058. doi:10.1371/journal.pone.0133058.

Mencarelli, R., W.J. Lipton, and S.J. Peterson, 1983. Response of 'zucchini' squash to low O_2 atmospheres at chilling and non-chilling temperatures. *J. Amer. Soc. Hort. Sci.* 108: 884-890.

Mercado-Ruiz, J.N. and M.A. Martínez-Téllez, 2010. Características sensoriales de la calabaza zucchini (*Cucurbita pepo* L.) envasada individualmente y conservada en refrigeración. *BIOtecnia* 12(2): 29-39.

Nagata, M. and I. Yamashita, 1992. Simple method for simultaneous determination of chlorophyll and carotenoids in tomato fruit. *Soc. Food Sci. Technol. (Nippon Shokuhin Kogyo Gakkaishi)* 39(102): 925-928.

Nunes M.C.N., E. Proulx, J.P. Émond, and J.K. Brecht, 2003. Quality characteristics of 'Horn of Plenty' and 'Medallion' yellow summer squash as a function of the storage temperature. *Acta Hort.* 628: 607-614.

Paris H.S and R.N. Brown, 2005. The genes of pumpkin and squash. *HortScience* 40(6): 1620-1630.

PerkinElmer, 1996. Analytical methods for atomic spectrometry. The PerkinElmer Corporation, Norwalk, Connecticut, USA. 310 p.

Shaw, N.L. and D.J. Cantliffe, 2005. Hydroponic greenhouse production of "baby" squash: Selection of suitable squash types and cultivars. *HortTechnology* 15(3): 722-728.

Sherman, M., G.W. Elmstrom, and J.J. Allen, 1985. Storage characteristics of three cultivars of yellow summer squash (*Cucurbita pepo* L.). *Proc. Fla. State Hort. Soc.* 98:216-218.

Sherman, M., H.S. Paris, and J.J. Allen, 1987. Storability of summer squash as affected by gene B and genetic background. *HortScience* 22(5): 920-922.

Singleton, V.L. and J.A. Rossi, 1965. Colorimetry of total phenolics with phosphomolybdic -phosphotungstic acid reagents. *American J. of Enology and Viticulture* 16: 144-158.

U.S. Department of Agriculture, 2018. Agricultural Research Service, FoodData Central. Squash, summer, zucchini, includes skin, raw. https://fdc.nal.usda.gov/fdc-app.html#/food-details/169291/nutrients.