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COMPARISON OF THE CONTENTS OF BIOACTIVE COMPOUNDS AND QUALITY PARAMETERS IN SELECTED MANGO CULTIVARS

PERIYAR SELVAM SELLAMUTHU[1], GABRIELA INÉS DENOYA[2], DHARINI SIVAKUMAR[1,3], GUSTAVO ALBERTO POLENTA[2] and PUFFY SOUNDY[1]

[1]Postharvest Technology Group, Department of Crop Sciences, Tshwane University of Technology, Pretoria, South Africa
[2]Laboratorio de Compuestos Proteicos Instituto de Tecnología de Alimentos Centro de Investigacion de Agroindustria Instituto Nacional de Tecnología Agropecuaria (INTA), Morón, Buenos Aires, Argentina

[3]Corresponding author.
TEL: +27 12 382 5303;
FAX: +27 12 3825869;
EMAIL: dharinisivakumar@yahoo.co.uk

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ABSTRACT

Mango cultivars Tommy Atkins, Zill, Peach, Sabre, Rosa and Phiva were analyzed for their quality parameters (fruit weight, flesh color chroma, L, h^0, total soluble solid [TSS]/titratable acidity [TA], firmness), bioactive compounds (total phenols, carotenoids, ascorbic acid, antioxidant activity) and polyphenol oxidase (PPO) activity. Cv. Sabre showed highest total phenolic content (76.43 mg gallic acid/ 100 g FW), carotenoids (9.90 mg/100 g of FW), ascorbic acid content (69.71 mg/ 100 g of FW) and antioxidant activity (1.2 mg of gallic acid/g of FW), whereas cv. Peach mango contained lower bioactive compounds. Multivariate principal component analysis analysis showed higher concentration of bioactive compounds in cv. Sabre mango, whereas cv. Tommy Atkins was firm and heavier. Cv. Rosa and Phiva were moderately rich in bioactive compounds and lower in fruit weight and firmness, whereas cv. Peach was higher in TSS/ TA. The PPO activity was higher in cvs. Rosa and Zill.

PRACTICAL APPLICATIONS

The variability in the quality parameters and bioactive components in five different mango cultivars was investigated. The results of this study provide valuable information to growers, distributors and consumers in selecting cultivars that are rich in bioactive components to consume as fresh fruit. Different cultivars were also identified for specific processing operations based on their quality parameters.

INTRODUCTION

Mango (*Mangifera indica* L. Anacardiaceae) is a popular subtropical fruit grown in the subtropical regions because of its high economical value. It is reported as the second largest tropical crop next to banana in terms of production, acreage and popularity (FAOSTAT 2010). The consumer acceptance of this fruit is high because of its excellent eating quality (bright color, sweet taste and luscious flavor) and nutritional composition such as total carotenoids (Godoy and Rodriguez-Amaya 1989), ascorbic acid (Franke *et al.* 2004) and polyphenols (Berardini *et al.* 2004).

Consumption of mango could provide significant amounts of bioactive compounds with antioxidant activity to the human diet. A large amount of provitamin A carotenoids can be obtained by including traditional plant sources such as mangoes in our daily diet. However, information on bioactive compounds and antioxidant properties of South African mangoes is not well documented in South African data food composition tables. On the other hand, deficiency in vitamin A is one of the most common problems, particularly in sub-Saharan Africa.

Mango cultivars Haden, Tommy Atkins, Kent and Keitt are popular (Bally 2011) because of their less fibrous nature and firm flesh properties, and are more suited for long-distance transportation (Sauco 2004). Mango is commercially produced in India, China, Thailand, Indonesia, Mexico, Brazil and others (FAOSTAT 2010). About 99% of South Africa's mango production is currently consumed locally as fresh or in processed products (Ntombela 2012).

Mango is consumed worldwide as either whole fruit, fresh-cut produce, processed juice, atchar pickle, dried fruit, chutney, pulp, paste, puree, jam, slices in brine or flour (Evans 2008; Anon 2011; Ntombela 2012). Fresh fruit quality needed for each of these products differs. For example, the stability of fresh-cut or dried mango during processing and storage depends primarily on fruit composition (or ripening stage) and certain postharvest-processing treatments. Fruits with high solid content are needed for mango puree, jam, pulp or paste to increase product yield. Increased total soluble solid (TSS) content is important to juice and concentrate manufacturers. Some level of firmness is required for high consumer appreciation of fresh-cut product. The flavor and color characteristics are virtually important to all end users of mango fruit. Fresh-cut mango was reported as a potential new product to the fresh produce sector (Djioua *et al.* 2010). However, during fresh cut processing, the mango fruit experiences softening and decrease in overall appearance because of tissue browning on the cut surface (Plotto *et al.* 2004). Cultivars with higher phenolic content and higher polyphenol oxidase (PPO) activity will be highly susceptible to flesh browning during cutting operation (Vásquez-Caicedo *et al.* 2002). It is interesting to compare the quality parameters and bioactive compounds of cv. Tommy Atkins, a widely consumed cultivar with other cultivar types knowing the potential of other alternative mango cultivars, can open new perspectives to farmers and to the local industry, benefiting the consumer by offering a great source of vitamin A.

Therefore, the aim of our study was to investigate some physicochemical properties (TSS, titratable acidity [TA], firmness, color value chroma, hue angle and fruit weight), bioactive compounds (total phenolics, total carotenoids, ascorbic acid, antioxidant activity) and PPO activity in five mango cultivars in South Africa.

MATERIALS AND METHODS

Fruits

Six cultivars of mangoes (Tommy Atkins, Zill, Peach, Sabre, Rosa and Phiva) were obtained at commercial maturity stage from the orchards of Bavaria Estates in Tzaneen, Limpopo Province in South Africa. Fruit was selected at the pack house of Bavaria Estates according to their size, color and appearance while discarding fruit with defects and physiological disorders. Total number of 40 fruits per cultivar type was obtained from Bavaria Estates. Thereafter, fruit was washed in chlorinated water (200 ppm NaOCl) and air dried. Out of 40 fruits, a set of 20 fruit per orchard was peeled, and the flesh was cut and frozen at −20C for biochemical analysis. The rest 20 fruits were subjected to fruit quality evaluation.

Fruit Quality Evaluation

Fruit flesh color was determined using a chromameter (Model CR-400; Minolta, Osaka, Japan) color analyzer calibrated to a white porcelain reference plate. The color space values L^*, hue angle (h^0) and chroma were measured on the two spots on opposite sides of the cut fruit surface, and the mean of the two measurements was considered as one reading. Fruit firmness was measured on opposing sides of each fruit with a penetrometer (Chatillon and Sons, New York, NY) equipped with a 6-mm diameter plunger capable of penetrating through the peel into the flesh. The TSS was measured with a digital refractometer (Atago Co., Tokyo, Japan) and expressed in percentages. Percentage TA was determined by titration of mango juice (10 mL) with 0.01 N NaOH solution to pH 8.1. The acidity was expressed as citric acid equivalent, and the TSS/TA ratio was determined.

Determination of Bioactive Compounds

Ascorbic Acid Content. Ascorbic acid content in the fruit juice was determined from 20 g flesh using the 2,6-dichlorophenolindophenol titrimetric method (AOAC 2000). The results were expressed as mg/100 g FW.

Carotenoid Content. Total carotenoid content was measured according to Rodriguez-Amaya (2001) with slight modifications. Cold pure acetone was mixed with mango pulp (4 g) sample and homogenized (extractor of pigment) until the residue became colorless. Thereafter, the homogenate was filtered under suction through a filter. Petroleum ether (30 mL) was added to the extract, and the acetone was removed from the extract by rinsing with distilled water (five to six times). Afterwards, carotenoids were collected from petroleum ether extract and then passed through a glass funnel containing anhydrous sodium sulfate. The determination was carried out by spectrophotometry at 453 nm and petroleum ether was used as a blank sample. Total carotenoid concentration was expressed as mg/100 g FW.

Extraction of Polyphenols. Mango pulp (20 g) was homogenized with 15 mL of 80% methanol (v/v) using a homogenizer (Ultra-Turrax, IKAWerk, Staufen, Germany). The homogenate was sonicated for 15 min and centrifuged at 10,000 × g at 5C for 15 min. Afterwards, the pulp mixture was filtered through Whatman no. 1 (Sigma Aldrich, Johannesburg, South Africa). In order to extract maximum polyphenols, the method was repeated twice. Thereafter, 80% methanol was added to the collected extracts to make up the volume to 50 mL, and further dilutions were made up with methanol (80%). The final concentration of the extract was 0.4 g/mL of the original mango pulp and was used for

total phenols and 2,2-diphenyl-1-picrylhydrazyl free radical DPPH assays (Robles-Sánchez *et al.* 2009; Sivakumar *et al.* 2012).

Total Phenolic Content. Total phenolic content (TPC) of the fruit extracts was determined using the Folin–Ciocalteu assay described by Singleton and Rossi (1965) with some modifications (Sivakumar *et al.* 2012). A 40-μL aliquot of diluted fruit extract was mixed with 1.8 mL of Folin–Ciocalteu reagent. After 5 min of equilibrium time at 25C, 1.2 mL of (7.5 g/100 mL) Na_2CO_3 solution was added to the extract. The solutions were mixed and allowed to stand for 1 h at 25C, and thereafter, the absorbance was measured at 765 nm using a Zenyth 200 rt microplate reader (Biochrom Ltd., Cambridge, U.K.). Total phenolic compounds were calculated using a standard curve of gallic acid and expressed as mg of gallic acid equivalents (GAEs)/100 g FW.

DPPH Free Radical-Scavenging Assay. Antioxidant scavenging activity of the mango extract was measured using DPPH (Sigma-Aldrich, Johannesburg, South Africa) scavenging assay according to Toit *et al.* (2001) with some modifications. Briefly, the extract was diluted with extraction solution (methanol : water [60:40]) to obtain different concentrations of sample. Thereafter, 250 μL of 90 μM DPPH was added in 96 well microplate with 28 μL of specific concentration of the sample extract. The mixture was sonicated and incubated in the dark for 60 min. Absorbance was read at 515 nm using a Zenyth 200 rt microplate reader. The results were expressed in mg of GAE/gram FW.

Assay for PPO Activity. PPO activity was estimated according to Soliva *et al.* (2001) with slight modification. In this study, 200 μL of 0.1 M phosphate buffer solution (pH 7) containing 1 mL of 0.5 M catechol and 20 μL of crude enzyme extract was pipetted according to sequence into the 96 well microplate, and the absorbance was measured at 410 nm for 5 min at 25C using a Zenyth 200 rt microplate reader. The enzyme activity was expressed as Δ $A410$ min/mg of protein. The protein content of enzyme extracts was estimated by dye-binding method of Bradford (1976) with bovine serum albumin as the standard. All the enzyme assays were carried out as six replicates per sample.

STATISTICAL ANALYSIS

The experiments were laid out as a completely randomized design. Twenty replicate fruits were used for each fruit quality or biochemical analysis separately. The experiment was repeated twice in order to confirm the observations. Data were subjected to analysis of variance (ANOVA) using the GenStat for Windows (2004) statistical package (VSN International, Hempstead, U.K.). Fisher's protected least significant difference at ($P < 0.05$) level of significance was performed. Principal component analysis (PCA) was carried out for the observations of fruit weight, flesh color chroma, L, h^0, TSS/TA, firmness, total phenols, carotenoids, ascorbic acid, antioxidant activity and PPO activity using XLSTAT Version 2011.2.05 (Kovach Computing Services, Wales, U.K.). The PCA was adopted using correlation matrix in this investigation to see whether the different mango cultivars can be grouped into different groups or clusters.

RESULTS AND DISCUSSION

Physicochemical characteristics of the five tested mango cultivars were determined at the commercial ripe stage. Fruit firmness and fruit weight were significantly ($P < 0.05$) higher in cv. Tommy Atkins and lower ($P < 0.05$) in cv. Sabre (Fig. 1a,b). According to Kader (2008), large fruit size, small seed size and firm-ripe mangoes are selected for fresh cut processing. The fully ripe mango fruits varied in their TSS and TA. The TSS/TA ratio differed according to the cultivar type at ripe stage. The TSS/TA was significantly higher ($P < 0.05$) in cv. Peach and lower ($P < 0.05$) in cv. Zill (Fig. 1c). Firmness and soluble solid content are important quality parameters for selecting mango fruits for fresh cut processing (Kader 2008).

The required firmness for cvs. Tommy Atkins and Kent mangoes meant fresh cut processing is 13.4–26.7 N (Rattanapanone *et al.* 2001). Allong *et al.* (2000) recommended half-ripe "Julie" and "Graham" mango with 13–16% soluble solids as ideal for fresh-cut purposes. Therefore, on this basis, it is evident from this investigation that Tommy Atkins can be recommended for fresh cut processing. The TSS/TA has a greater impact on taste, and the lower ratio of TSS/TA in cv. Zill implies more sweet–sour taste.

Color data (Fig. 1d) correspond to the stage of maturity for each cultivar showing that the fruits were ripe. Generally, flesh color changes in mango are known as a reliable parameter to note the extent of fruit ripening (Bender *et al.* 1995). Significantly higher ($P < 0.05$) chroma and lower h^0 values were noted in cv. Sabre, whereas cv. peach mango showed significantly ($P < 0.05$) lower chroma and higher h^0. Pulp color is an important parameter for processing and determining the consumer acceptance of fresh-cut mangoes. The chroma value is known as a measure of color intensity, and an increase in chroma during ripening is linked to carotenoid biosynthesis (Scaffer and Andersen 1994). The decrease in h^0 value was also reported to correspond with an increase in carotenoids synthesis during fruit ripening.

The ascorbic acid content ranged from 50.71 to 17.01 mg/100 g FW in all six mango cultivars. Ascorbic acid content

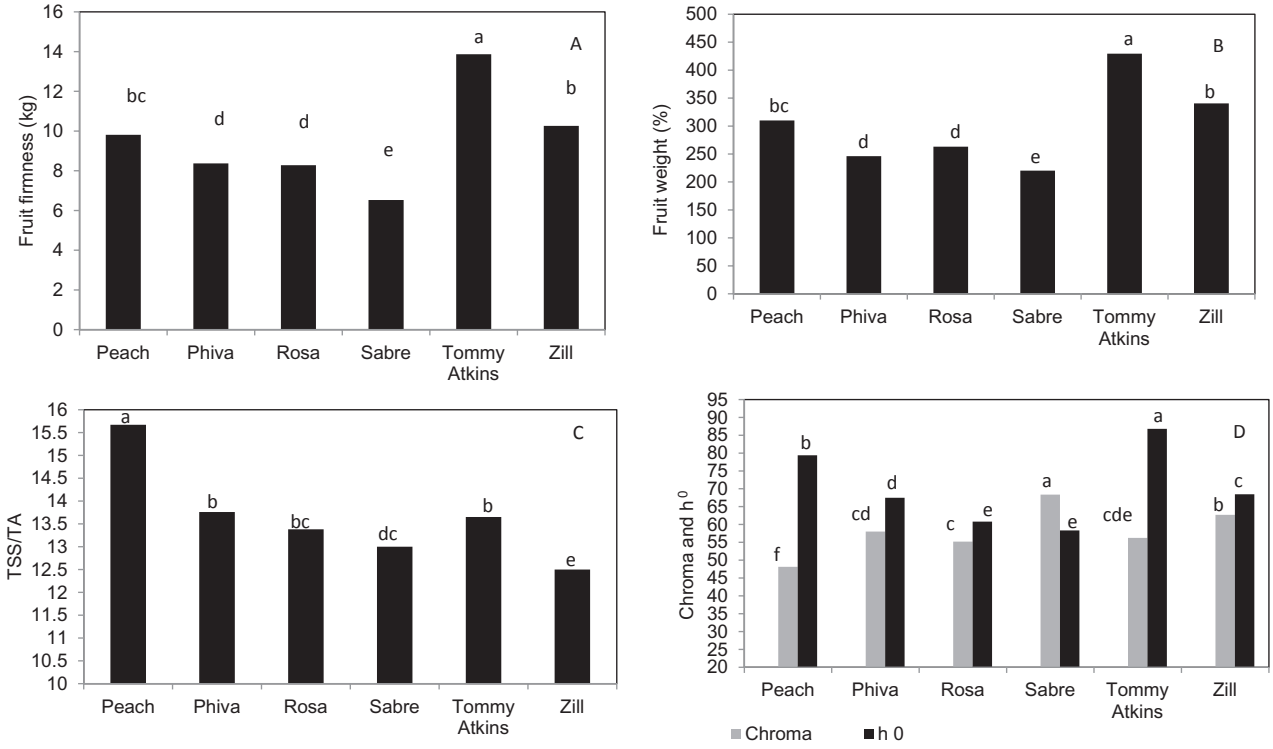

FIG. 1. QUALITY PARAMETERS (A) FRUIT FIRMNESS, (B) FRUIT WEIGHT, (C) TOTAL SOLUBLE SOLID (TSS)/TITRATABLE ACIDITY (TA) AND (D) COLOR VALUES (CHROMA AND H^0) IN DIFFERENT MANGO CULTIVARS AT COMMERCIAL MATURITY

was significantly higher ($P < 0.05$) in cv. Sabre and lower ($P < 0.05$) in cv. Tommy Atkins (Fig. 2a). According to the reports, the ascorbic acid content varied between the mango cultivars (Kent [11 mg/100 g FW] Robles-Sánchez *et al*. 2009; Palmer from Brazil [40.9 mg/100 g FW] Valente *et al*. 2011). Higher TA was reported to be linked with the stability of ascorbic acid in fruit (Toor and Savage 2006). Ascorbic acid plays a major role as an antioxidant in the detoxification of hydrogen peroxide, superoxide radicals (O2–) and hydroxyl radicals (OH•) that are generated from the different reactive oxygen species in the plant tissue (Moldau 1998).

It has to be noted that the protective benefits of the bioactive compounds are due to their ability to quench or remove free radicals, and thereby protecting the human body against abnormal oxidative changes (Toor and Savage 2006). Total phenolic compounds were reported to be responsible for the maintenance of antioxidant capacity or scavenging activity in both whole and fresh-cut mangoes (Robles-Sánchez *et al*. 2009). The total phenolics generally ranged between 32.54 and 208.7 mg GAE/100 g FW in mangoes (Ribeiro *et al*. 2007). In this study, the total phenolic varied from 30.36 to 76.43 mg GAE/100 g FW. Cultivar Sabre revealed significantly higher ($P < 0.05$) phenolic content, whereas cvs. Phiva, Rosa and Zill showed moderate phenolic content in the edible portion of the fruit. The observed differences in TPC in six cultivars could be related to the wide range of phenolic compounds such as gallic acid, ρ-hydroxybenzoic acid, ρ-coumaric acid, quercetin, ellagic acid, catechin and sinapic acid as reported by Liu *et al*. (2013).

The TPC was significantly ($P < 0.05$) lower in the edible portion of cv. Tommy Atkins mangoes (Fig. 2b). The total phenol content in cv. Sabre was higher than the cv. Keitt mango (50 mg/100 g FW) from Florida, USA (Mahattanatawee *et al*. 2006) and lower than cv. Ataulfo (175 mg/100 g FW mango from Mexico (Robles-Sánchez *et al*. 2009). Significant correlation was reported between TPC and antioxidant activity, and similar observations were reported by different researchers (Sellappan *et al*. 2002; Gorinstein *et al*. 2004; Vasco *et al*. 2008). The content and reactivity of phenolics in fruits could determine the potential browning reactions following tissue exposure to oxygen environment such as found in fresh cut processing (Vásquez-Caicedo *et al*. 2002).

Antioxidant activity of cv. Sabre was significantly ($P < 0.05$) stronger and weaker ($P < 0.05$) in cv. Tommy Atkins (Fig. 2c). According to Kivrak *et al*. (2009), higher antioxidant activity can be related to the content of phenolic compounds. Ribeiro *et al*. (2007) also demonstrated that the DPPH radical-scavenging activity strongly

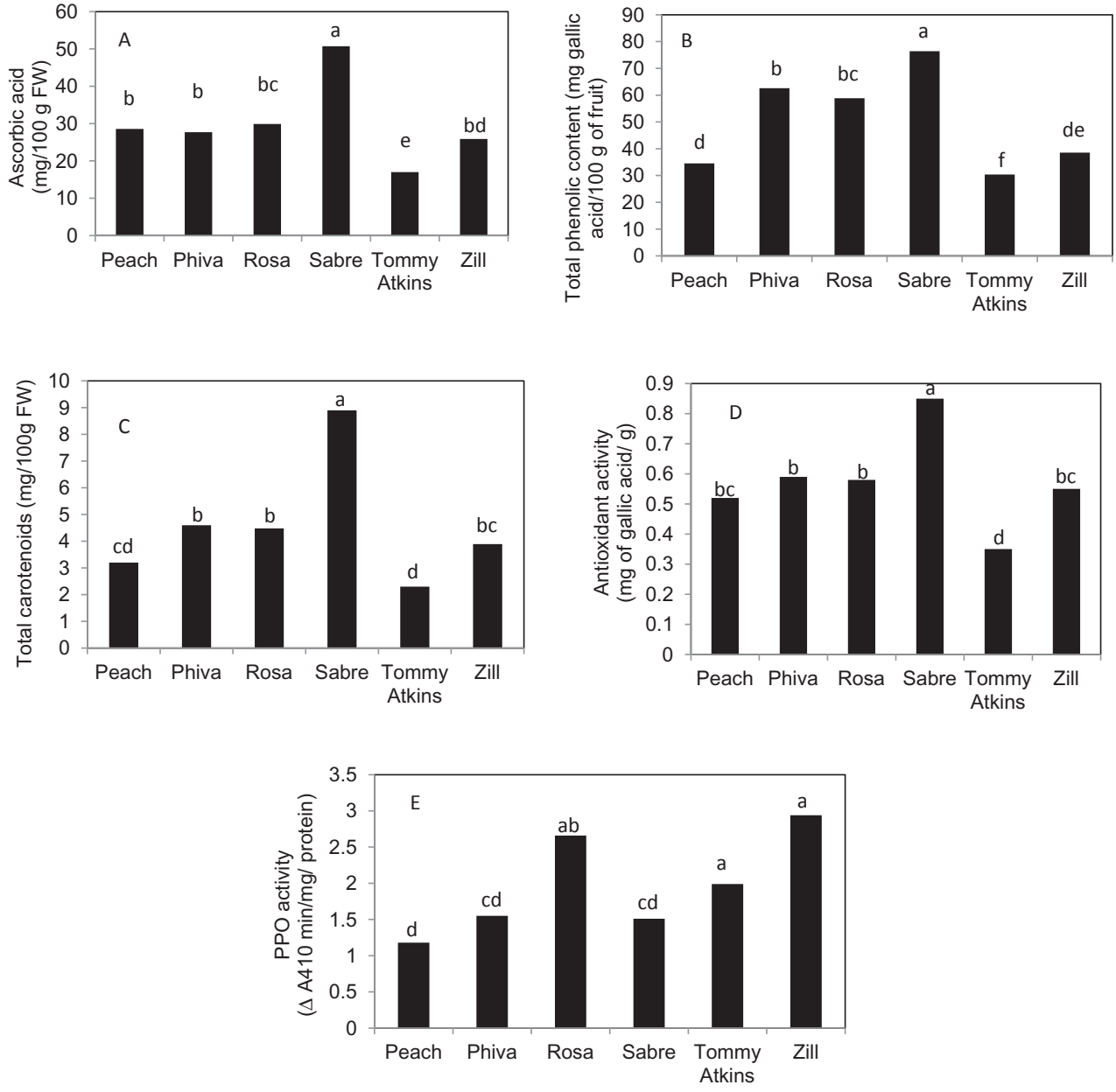

FIG. 2. BIOACTIVE COMPOUNDS (A) ASCORBIC ACID, (B) TOTAL PHENOLIC CONTENT, (C) ANTIOXIDANT ACTIVITY, (D) CAROTENOIDS AND (E) POLYPHENOL OXIDASE ACTIVITY IN DIFFERENT MANGO CULTIVARS AT COMMERCIAL MATURITY

correlated with the ascorbic acid content. Currently, antioxidant capacity or scavenging activity is shown as a desirable attribute for marketing the potential health benefits of fresh fruit and vegetables. Although the antioxidant activities of fruits are influenced by the cultivation conditions or preharvest factors (Scalzo *et al.* 2005), our study showed variation between the cultivars even though they were obtained from same location. The DPPH method was selected to determine the antioxidant activity because this method is sensitive, reproducible, rapid and easy to carry out large number of sample replicates using multiple samples that can be read by a multiplate reader (Miller *et al.* 2000).

Total carotenoid content of the different mango cultivars ranged from 0.9 to 9.20 mg/100 g FW (Litz 1997). Total carotenoid content was significantly ($P < 0.05$) higher in cv. Sabre, whereas moderate amounts were found in cvs. Rosa, Phiva, Zill and Peach with significantly ($P < 0.05$) lower content in cv. Tommy Atkins (Fig. 2d). It is well known that carotenoids are responsible for the color and antioxidant

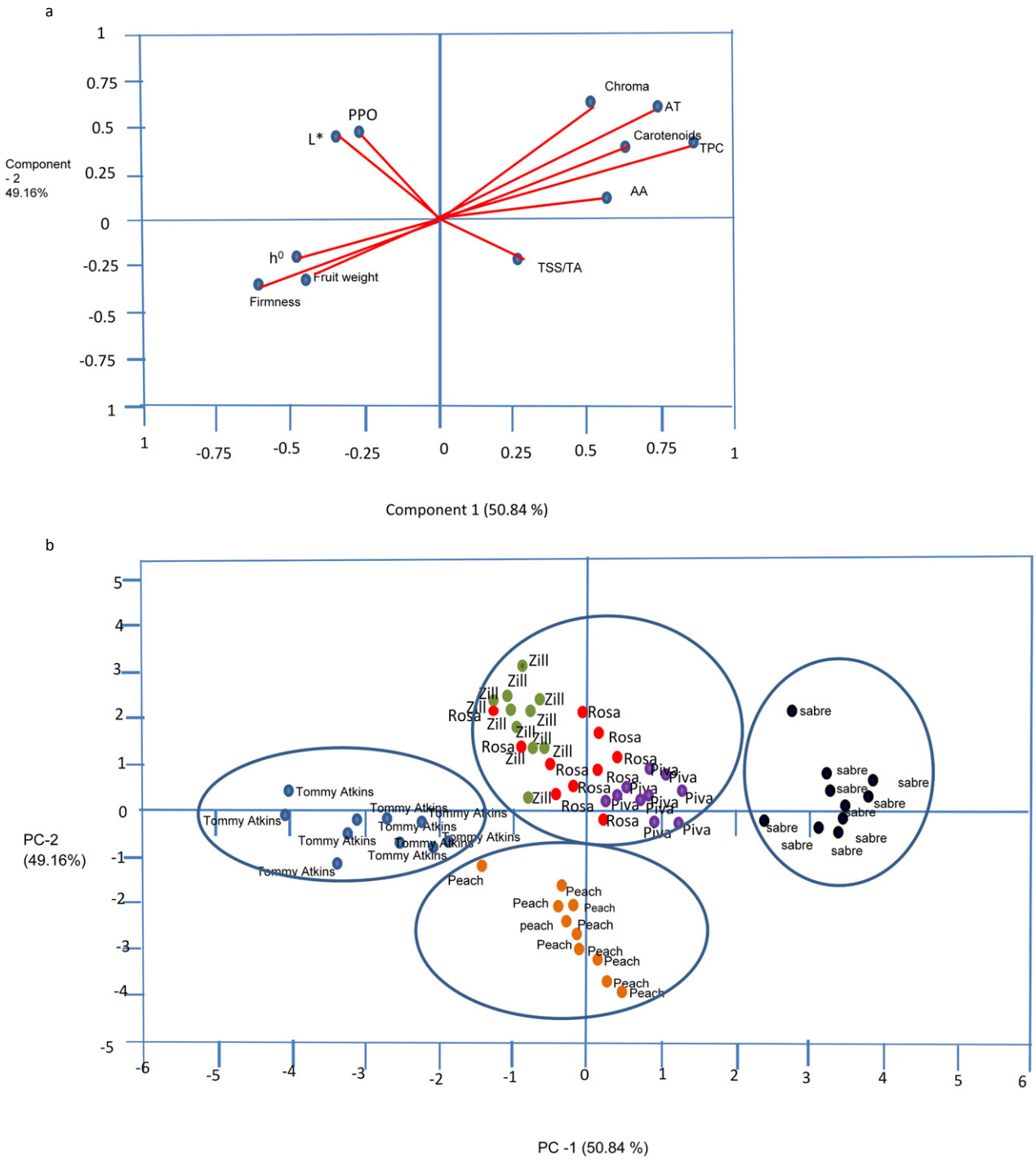

FIG. 3. PRINCIPAL COMPONENT ANALYSIS (PCA) SHOWING (A) CORRELATION LOADINGS AND (B) BIOACTIVE COMPOUNDS AND QUALITY PARAMETERS AND OTHER PROPERTIES IN DIFFERENT MANGO CULTIVARS

properties in biological systems. Carotenoid synthesis in mangoes coincides with the ripening process and production of ethylene (Vásquez-Caicedo *et al.* 2005). β-carotene is the predominant carotenoid contributing towards 50% of the total carotenoid content in a fully ripe mango fruit (Manthey and Perkins-Veazie 2009). PPO activity was significantly ($P < 0.05$) lower in cv. Piva and higher ($P < 0.05$) in cv. Zill and in Rosa (Fig. 3e). Oxidation of phenolic com-

pounds, catalyzed by the PPO enzyme, favors the production of brown colored pigments such as melanins that cause browning in fresh-cut surface or during mechanical damage and results in loss of quality (Vámos-Vigyázó 1981).

PCA Analysis

PCA was carried out to investigate the data stucture in order to establish a cultivar classification based on the obtained data including the physicochemical properties, bioactive compounds and PPO activity. With repective eigenvalues >1, two principal components (PCs) were obtained with their factor loading shown in Fig. 3a. Eighty-seven percent of the original variance in the data set of fruit properties (PC1 60.8 % and PC2 27.40%) was explained by the first two PCs.

The two PCs explained 87.6% of the *x* variables selecting 11 parameters, including total phenols, carotenoids, ascorbic acid, antioxidant activity, chroma, L^* h^0 fruit weight, firmness, TSS/TA and PPO activity. Total phenols, carotenoids, ascorbic acid, antioxidant activity, chroma and L^* h^0 were mainly accounted for with PC1, whereas fruit weight, firmness, TSS/TA and PPO activity mainly accounted for with PC2 as shown in Fig. 3a. The results of PCA analysis are shown in Fig. 3b, where different four classes (grouping) are suggested. As it is illustrated in Fig. 3a, bioactive compounds such as total phenols ($r = 0.84$), carotenoids ($r = 0.65$), antioxidant activity ($r = 0.72$), ascorbic acid content ($r = 0.58$), quality parameter firmness ($r = -0.54$) and fruit weight ($r = -0.52$) on PC1 and the quality parameters, TSS ($r = -0.57$), L ($r = 0.48$), chroma ($r = 0.63$), h^0 ($r = -0.54$) and PPO ($r = 0.51$) on PC2 helped to classify the cultivars in Fig. 3b into separate groups. The four classes or groups are the following: group A that includes cvs. Zill, Rosa and Phiva; group B that only includes cv. Sabre; group C, Tommy Atkins; and group D, only the cv. Peach.

Based on this analysis, cv. Saber was found to be rich in bioactive compounds (carotenoids, ascorbic acid, TPC and antioxidant-scavenging activity) and followed by cvs. Phiva and Rosa (intermediate), whereas cv. Tommy Atkins had less bioactive compounds. On the other hand, the flesh color (higher chroma) can be linked to the higher bioactive compounds especially with carotenoids. Similarly higher bioactive compounds and antioxidant activity were shown in a PCA plot for Tainong No1, a Chinese cultivar by Liu *et al.* (2013). The other important factors responsible for the group differentiation were fruit weight and fruit firmness which clearly showed that cv. Tommy Atkins is larger and firmer fruit. Fig. 3b also shows the moderately susceptible cultivars (Rosa and Zill) for flesh browning because of the higher PPO activity, whereas the PCA plot obtained from the investigations of Liu *et al.* (2013) shows that JinHwang mangoes from China shows higher PPO activity.

The taste can be predicted for these cultivars based on TSS/TA, where cv. Peach can be very sweet, and cv. Zill can be sweet and sour (more towards sour). However, in order to confirm this, sensory properties need to be evaluated. The cv. Saber also showed lower PPO activity and lower in fruit weight (small fruits) as cv. Tainong No1 shown in the PCA plot described by Liu *et al.* (2013).

CONCLUSIONS

Clear differences in the content of total phenols, carotenoids and antioxidant activity were determined between the different cultivars, and this information is valuable especially for the growers, processors and consumers when selecting mango cultivars for different purposes. The PCA analysis enabled to classify the cultivars in to different groups. Sabre cultivar is mainly noted for high bioactive compound content and high antioxidant activity and can be consumed as fresh fruit. Peach mangoes are typically noted for their TSS/TA ratio making them suitable for juice manufacturers. Cultivar Tommy Atkins are distinguished with higher flesh firmness and higher fruit weight and show potential for fresh cut processing.

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