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# Asynchronous ripening behavior of cactus pear (*Opuntia ficus-indica*) cultivars with respect to physicochemical and physiological attributes

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ABSTRACT

Physicochemical and physiological ripening events in cactus pear (*Opuntia ficus-indica*) fruit of cultivars 'Ntopia' and 'Hercules' were profiled against skin coloration from mature-green (S1) to over-mature (S5). Fructose and glucose accumulation were linear in 'Ntopia' but peaked near S3 in 'Hercules' synchronously to the appearance of sucrose. Betalains increased steadily in 'Ntopia' (103.2 mg/l) but peaked before full skin coloration in 'Hercules' (49.7 mg/l); whereas phenolic content remained invariable and ascorbate content peaked near S5 in both 'Ntopia' (108.6 μg/g) and 'Hercules' (163.1 μg/g). Cell wall material diminished with maturity though textural changes with ripening appeared not related to pectin solubilization but to weakening of glycan bonding and loss of neutral sugars. Fruit firmness rather was correlated to seed weight ($r = 0.89$) and seed-to-pulp ratio ($r = 0.73$). Cultivar differences highlighted in the chronology of ripening events are critical for defining optimum harvest maturity and postharvest handling protocols for premium quality cactus pear fruit.



## 1. Introduction

Resilience to conditions of prolonged drought and high temperature, owing mainly to crassulacean acid metabolism, make the *Cactaceae* family subject of renewed agronomic interest for cultivation under arid and semi-arid climates, expedited by the ensuing climate change (Sáenz, 2013). The growing exploitation of cactus fruits and cladodes, in particular of *Opuntia* species, in the cosmetics and neutraceutical industries, and the heightened interest in their fresh fruits, widely referred to as cactus pears, stem from the unique functional properties and composition of this fruit (El-Mostafa et al., 2014; Kuti, 2004; Martínez, Esparza, & Fragoso, 2014; Pimienta- Barrios, 1994; Stintzing et al., 2005; Sáenz, 2013; Sáenz & Sepúlveda, 2001). While expansive genetic diversity within *Opuntia* is found in the countries of South America, prominently in Mexico, introduction of these fruiting species into the Old World, possibly in the 16th century, has long provided a naturalized genetic basis for the resurgence and valorization of this crop in southern Europe over the last two decades (Allegra et al., 2015; Piga, Del Caro, Pinna, & Agabbio, 2003). Cultivation of mostly *Opuntia ficus-indica* L. genotypes is now widespread in southern Europe, particularly on the islands of Malta, Sicily, Crete and Cyprus (Lydakis, Pompodakis, Markellou, & Lionakis, 2005; U.S. National Plant Germplasm System., 2016).

Characterization of cultivated cactus pear genotypes has displayed wide variation in important compositional traits such as their phenolic, carbohydrate, pigment and soluble/insoluble fibre contents (Parish & Felker, 1997; Pimienta- Barrios, 1994). Previous workers have reported on significant genotypic, environmental, and cultural effects that contribute to compositional variation in cactus pear fruits. On the contrary, work on the on-tree ripening behavior of cactus pears and the effect of harvest maturity on fruit quality and physicochemical composition remains scarce, owing perhaps to the termination of physiological maturation of cactus pear fruit at harvest and the absence of acute climacteric rise in respiration and ethylene production during postharvest ripening (Cantwell, 1999). Physicochemical assessment of fruit quality is thus commonly limited to a snap-shot of fruit composition at harvest, and what may be construed as genotypic differences in quality attributes may in fact reflect differential ripening behavior affecting harvest maturity. As previously reported, sound prediction of harvest maturity is particularly critical for non-climacteric fruit, whose physiological maturation terminates at harvest and their quality tends to decline thereafter (Soteriou, Kyriacou, Siomos, & Gerasopoulos, 2014).

The overall objective of our work has been to characterize the ripening behavior of two prominent *Opuntia ficus-indica* cultivars

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widely cultivated in Cyprus. Physicochemical and physiological changes with maturity were profiled in relation to skin color development, which was referenced as a non-destructive index of harvest maturity. Fruits of orange-fleshed cultivar 'Ntopia' and red-fleshed cultivar 'Hercules' were harvested at five stages of maturity based on skin coloration ranging from mature green to fully colored, overripe fruit. Physical components of quality examined included seed and pulp relative weights, pulp reflectance colorimetry and fruit mechanical texture analysis. Phytochemical components of quality examined entailed pulp pH, titratable acidity, soluble solids and carbohydrates (glucose, fructose, sucrose), phenolic compounds, ascorbate, betalains (betacyanins and betaxanthins), and the pectins and neutral sugars of the fractionated alcohol-insoluble cell wall material. Fruit physiological attributes examined included respiratory activity and ethylene production rate. Analysis of the above components aimed to highlight potential differences in cultivar ripening behavior that are critical for the determination of optimum harvest maturity.

## 2. Materials and methods

### 2.1. Plant material and sample preparation

Cactus pears were obtained from a commercial plantation located at Frenaros, Cyprus (N: 35°2′27″; E: 33°55′9″), comprised of ten-year old plants spaced at 5 m × 5 m and trained to an open vase system. Fertilizer application rates were 90 N-30P-30 K kg $ha^{-1}$. Supplemental irrigation of about 200 mm was delivered during the dry season (June–October) by drip irrigation. Weeds were controlled mechanically. Pest and disease control was minimal as no problems were encountered. All fruit samples were harvested in August, 2014, and their harvest maturity was classified in five stages according to skin coloration: S1: mature green; S2: breaker; S3: skin half- colored; S4: skin coloration near complete; S5: full, deep coloration – over-mature (Supplementary illustration). All samples were transferred for assessment to the Postharvest Technology Laboratory of the Agricultural Research Institute, Nicosia and sample preparation was completed within the day of harvest.

### 2.2. Fruit morphometric, textural and optical assessment

Fruits were manually peeled by severing their tips with a knife, then longitudinally incising the epidermis and releasing the pulp. A homogenate of the pulp was prepared under low speed using a Vita Prep 3 (Vita-Mix Corp., Cleveland, Ohio) blender and filtered through a 1250 μm aperture stainless steel sieve to separate the pulp from the seeds. Whole fruit and net pulp weights were determined to ±0.01 g on an electronic balance (XT 4200C; Precisa Gravimetrics, Dietikon, Switzerland). Seed weight was determined to ±0.001 g on an analytical balance (XT120A; Precisa Gravimetrics, Dietikon, Switzerland). Fruit juice was obtained by filtering the pulp homogenate through double cheesecloth. Juice color was determined using a Minolta CR-410 Chroma Meter (Minolta, Osaka, Japan) with a diffusion illumination 0° viewing angle geometry and the color space XYZ, Yxy, L*a*b*, Hunter, L*C*h, Munsel as the default. Measurements were performed in the CIELAB color space (McGuire, 1992), and recorded parameters were lightness (L*), color components a*(+red/−green) and b*(+yellow/−blue), chroma [$C^* = (a^{*2} + b^{*2})^{1/2}$] and hue angle [h° = arctan (b/a)]. Fruit firmness was measured using a TA.XT plus Texture Analyser (Stable Micro Systems, Surrey, UK) equipped with a 50 kg load cell. Firmness was determined by a compression test, corresponding to 7% strain, applied once on each fruit using a 75-mm flat probe at a test speed of 2 mm $s^{-1}$.

### 2.3. Titratable acidity, soluble solids and carbohydrates content

The pH of the juice was measured with a pH-electrode (Seven-Multi; Mettler-Toledo GmbH, Schwerzenbach, Switzerland). An aliquot of 20 mL juice was titrated with 0.1 M NaOH to a pH endpoint of 8.2 on an automatic titrator (794 Basic Trinitro; Metrohm Ltd, Herisau, Switzerland) to determine its titratable acidity, which was expressed as % w/v content of citric acid. The soluble solids content (SSC) at 20 °C of the filtered juice was measured on a digital refractometer (RFM870; Bellingham-Stanley Ltd, Kent, UK). Part of the juice was transferred to 50 mL falcon tubes, instantly frozen in liquid nitrogen, and then stored at −80 °C for analyses. Analysis of non-structural carbohydrates (glucose, fructose, and sucrose) was performed by liquid chromatography as previously described (Soteriou et al., 2014), using an Agilent HPLC system (Agilent Technologies, Santa Clara, CA, USA) equipped with 1200 Series quaternary pump and a 1260 Series refractive index detector commanded by ChemStation software. In order to eliminate pigment interference, samples were prepared by solid phase extraction using 0.7 ml Sep-Pak® C18 cartridges (WAT020515, Waters, Milford, MA, USA) conditioned with 2.0 ml each of methanol, water and 50% acetonitrile, then 2.0 mL of air to remove the excess acetonitrile. Samples were diluted in 50% acetonitrile in 1:1 ratio. Two ml sample was applied onto the C18 cartridge, the first 1 ml was discarded and the following 1 ml was collected and 10 μl injected onto the HPLC. Separation was performed on a 4.6 × 250 mm carbohydrate column at 35 °C (Waters, Milford, MA, USA) using an acetonitrile:water (75:25) mobile phase at a flow rate of 1.4 ml $min^{-1}$. Quantification was performed against fructose, glucose and sucrose external standard calibrating curves with a coefficient of determination ($R^2$) greater than 0.9999. Recovery trials performed under the same operating conditions were in all cases on the order of 100%.

### 2.4. Total phenols, ascorbic acid, betacyanins and betaxanthins

The total phenols content of the juice was determined according to the method of Singleton, Orthofer, and Lamuela-Raventos (1999). Briefly, 5 ml juice was added to 20 ml acidified methanol (50% MeOH:10% HCl), vortexed for 1 min, then kept at 4° C in the dark for 24 h. Following centrifugation at 5000 × g for 10 min at 2 °C, the supernatant was transferred to a 25 ml amber volumetric flask, brought to full volume with additional extraction solvent and used for the quantification of total phenols. In 15 ml Falcon tubes containing 2.0 ml, of 10% aqueous Folin-Ciocalteau's reagent were added 200 μl of gallic acid standard or extracted sample, and 800 μl of $Na_2CO_3$ 75 g $l^{-1}$ aqueous solution. The mixture was vortexed and incubated at 40° C for 30 min of orbital shaking at 180 rpm in the dark. The tubes were then cooled on ice slurry before quantification of duplicated samples at 765 nm. Quantification was performed on a Jasco V-550 UV–vis spectrophotometer (Jasco Corp., Tokyo, Japan) equipped with Spectra Manager™ II Software against linear calibration with external gallic acid standards over the range of 50–500 mg $l^{-1}$, yielding regression coefficients ($R^2$) greater than 0.99. The total phenols content of the juice was expressed in μg $ml^{-1}$. The total ascorbic acid content of the pulp was determined according to the method of Klein and Perry (1992), based on the reduction of 2,6 dichloroindolphenol by pulp samples diluted 1:10 (w/v) with 1% metaphosphoric acid. Quantification was performed at 515 nm against linear calibration with external L-ascorbic acid standards over the range of 10–100 mg $l^{-1}$, yielding regression coefficients ($R^2$) greater than 0.99. Ascorbic acid content of the pulp was expressed in μg $g^{-1}$ f.w. Photometric determination of betalains was performed after the method of Stintzing et al. (2005). Dilution of aqueous pigment extracts with McIlvaine buffer (pH 6.5, citrate-phosphate) was adjusted to

obtain absorption maxima of $0.9 \leqslant A \geqslant 1.0$. Juice betacyanin content was quantified in betanin equivalents at $\lambda = 538$ nm using the molecular weight of betanin (550 g $mol^{-1}$) and a molar extinction coefficient $\epsilon = 60{,}000$ L $mol^{-1}$ $cm^{-1}$ in $H_2O$. The juice betaxanthin content was determined in indicaxanthin equivalents at $\lambda = 480$ nm using the molecular weight of betaxanthin (308 g $mol^{-1}$) and a molar extinction coefficient $\epsilon = 48{,}000$ L $mol^{-1}$ $cm^{-1}$ in $H_2O$. All measurements were performed in duplicate and results for betacyanins and betaxanthins were expressed in μl/mL while the sum of the two pigments was taken as the total betalain content.

### 2.5. Cell wall pectins and neutral sugars

Cell wall material (CWM) was isolated based on the methods of Selvendran (1975) and Huysamer, Greve, and Labavitch (1997) adapted to cactus pear. Fresh pulp samples (seeds removed) of approximately 25 g obtained from the central region of each fruit were homogenized in 100 mL 95% ethanol using a Lab Gen 125 homogenizer (Cole Parmer, USA) and then reflux-boiled at 82 °C and 40 rpm for 45 min on a Büchi R-215 vacuum controlled rotovapor (Büchi Labortechnik AG, Flawil, Switzerland). The insoluble residue was collected on 20 μm NY20 nylon net filters (Merck Millipore Darmstadt, Germany) by vacuum filtration and washed repeatedly with ethanol and finally acetone to yield a colorless mass which was oven-dried overnight at 35 °C. The dried alcohol-insoluble residue, termed CWM, was then sequentially fractionated into water-soluble (WSF), carbonate-soluble (NSF) and alkali-soluble (KSF) fractions according to the modified method of Jiménez et al. (2001). About 50 mg of CWM sample was sequentially suspended in 15 mL of ultrapure water, 50 mM $Na_2CO_3$, or 4 M KOH; with each suspension kept in orbital shaking at 180 rpm at room temperature for 2.5 h before vacuum filtered through 20 μm NY20 nylon net filters. Each fraction filtrate was diluted to a final volume of 20 ml and used for determination of pectins and neutral sugars. The pectin content of each fraction was quantified as an approximation from uronic acid measurements according to the phenylphenol spectrophotometric assay of Blumenkrantz and Asboe-Hansen (1973) and it did not account for rhamnose, galactose and arabinose components. Galacturonic acid (0–100 μg/ml) was used as a calibration standard and results were expressed in galacturonic acid equivalents per g CWM. Neutral sugars of non-cellulosic origin were analyzed by the anthrone colorimetric method described by Dische (1962), using glucose (0–100 mg/l) as a calibration standard.

### 2.6. Respiration and ethylene production rates

Respiration and ethylene production rates were measured in a static system (Song, Kim, & Yam, 1992). Samples of 4–6 weighed fruits were sealed in 1.5 L glass jars equipped with ball valve controlled outflows bearing 11 mm Thermogreen™ rubber septa (Supelco, Bellefonte, PA, USA). Jars were kept for 60 min at 22 ± 0.5 °C in a Sanyo MIR153 (Panasonic, Osaka, Japan) incubator and headspace samples were then taken with a hypodermic syringe. Respiration rate, expressed in ml $CO_2$ $kg^{-1}$ $h^{-1}$ production, was measured on a portable gas analyzer equipped with an infrared sensor (CheckMateII; PBI Dansensor A/S, Rønnedevej, Denmark). Ethylene ($C_2H_4$) concentration was analysed on a 7890A Agilent Gas Chromatograph (Agilent Technologies, Santa Clara, CA, USA) equipped with a flame ionization detector and an Agilent stainless steel column (3 mm id × 2 m) packed with Porapak Q 80/100 mesh. Helium was used as a carrier gas under constant inlet pressure of 22 psi. Inlet and oven temperatures were maintained at 80 °C and detector temperature at 155 °C. Ethylene was detected at 1.6 min and total runtime was 2.5 min. Calibration was performed with the use of external (1 and 5 ppm) standards. Ethylene production rates were expressed in μl $kg^{-1}$ $h^{-1}$.

### 2.7. Statistical analysis

A completely randomized experimental design was applied with each cultivar-maturity combination replicated six times, with ten fruits per replicate. Separate sets of fruits were sampled for (a) assessment of physical components; (b) determination of pulp SSC, pH, titratable acidity (TA), fructose, glucose and sucrose contents; (c) determination of phenolic, ascorbate, and betalains contents, and (d) determination of pectins and neutral sugars in the water-soluble, carbonate-soluble, and alkali-soluble fractions of the alcohol-insoluble cell wall residue. Respiratory activity and ethylene production rates were determined on six replicate samples of 4–6 weighed fruits each per cultivar for each maturity stage. Data were subjected to analysis of variance (ANOVA). Provided the presence of significant effect and the absence of interaction, mean comparisons were performed according to Duncan's Multiple Range Test. Regression analyses were further performed for the evolution of each component of fruit quality and composition with maturity. All statistical analyses were executed using SAS 9.1. statistical package (SAS Institute, Cary, NC, USA).

## 3. Results and discussion

### 3.1. Morphometric, textural and optical characteristics

Fruit growth, expressed as mean fruit weight, was a genotypic trait more influenced by cultivar rather than the stage of maturity (Table 1). The mean fruit weight over stages S1-S5 of cultivar 'Hercules' (116.4 g) was higher by 19.9% than that of 'Ntopia' (97.1 g). Progressive fruit growth with ripening resulted in an overall (S1-S5) weight increase of 40.6% and 26.4% for 'Ntopia' and 'Hercules', respectively. Incremental fruit growth was greatest between mature-green stage (S1) and the appearance of epidermal color (S2), accounting for 41.1% and 44.3% of overall fruit growth in 'Ntopia' and 'Hercules', respectively. Regression analysis revealed quadratic increase in fruit weight for both cultivars, with more notable "fruit filling" between stages S1-S3 for 'Ntopia' (Fig. 1A). Maximum fruit weight, recorded at S5, classifies both 'Ntopia' (110.5 g) and 'Hercules' (127.8 g) among small-fruited genotypes (Parish & Felker, 1997; Pimienta-Barrios, 1994; Stintzing et al., 2005).

Seed weight was not influenced by fruit ripeness as seed development and fruit physiological maturity were complete by S1, although commercial fruit maturation was just commencing (Table 1). The 100-seed weight of 'Ntopia' was about half (56.3%) that of 'Hercules' and that was also reflected in the lower seed-to-pulp ratio of 'Ntopia' that renders its fruit fleshier and more appealing to consumers. Seed-to-pulp ratio dropped sharply between S1-S2 as seed weight was unaltered while pulp accumulated; no change however was observed with subsequent ripening. In the commercially mature stages S3-S5 the mean seed-to-pulp weight ratio was 5.1% for 'Ntopia' and 7.7% for 'Hercules', which classifies them, relative to other characterized cultivars, as ones of intermediate and high seediness, respectively (Parish & Felker, 1997; Pimienta-Barrios, 1994). Cultivar differentiation was not observed in pulp relative net weight which increased progressively between S1-S3, then remained stable between S3-S5 at a mean value of 53.6%, which is proximate to values reported for other varieties (Table 1; Kuti, 2004).

Fruit firmness was significantly reduced only at S4 and S5 when fruit had attained full maturity (Table 1). Mean firmness of 'Ntopia'

**Table 1**
Effects of cultivar and harvest maturity on cactus pear fruit weight, 100-seed weight, pulp weight seed weight, fruit resistance to compression and juice color components L*, C*, a* and Hue.

| Source of Variance | Fruit weight (kg) | 100Seed weight (g) | Pulp/ Gross (%) | Seed/ Pulp (%) | Firmness (kg-force) | L* (0 −1 0 0) | Chroma ($\sqrt{(a^{*2} + b^{*2})}$) | Hue (0–360°) | a* (0–60) | b* (0–60) |
|---|---|---|---|---|---|---|---|---|---|---|
| Cultivar (C) | *** | *** | | *** | *** | *** | *** | *** | | *** |
| Maturity (M) | ** | | *** | *** | *** | *** | | *** | | ** |
| C × M | | | | | | | | * | ** | |
| *Means* | | | | | | | | | | |
| Cultivar | | | | | | | | | | |
| Ntopia | 97.1 b | 1.137 b | 47.1 | 6.42 b | 2.945 b | 34.80 a | 25.75 a | 46.24 | 15.78 | 18.12 a |
| Hercules | 116.4 a | 2.018 a | 47.6 | 8.96 a | 5.970 a | 28.05 b | 17.49 b | 12.70 | 16.74 | 3.86 b |
| Maturity | | | | | | | | | | |
| S1 | 89.8 c | 1.523 | 31.0 c | 11.76 a | 4.920 a | 38.15 a | 25.82 | 51.81 | 13.14 | 18.77 a |
| S2 | 102.3 bc | 1.564 | 46.8 b | 7.49 b | 4.961 a | 31.12 b | 21.97 | 30.88 | 16.93 | 11.51 b |
| S3 | 109.0 ab | 1.627 | 53.7 a | 6.18 b | 4.755 a | 29.57 b | 20.15 | 22.63 | 17.88 | 8.55 b |
| S4 | 114.9 ab | 1.615 | 55.8 a | 6.37 b | 3.719 b | 29.67 b | 20.97 | 21.76 | 17.19 | 8.81 b |
| S5 | 118.0 a | 1.492 | 51.2 ab | 6.30 b | 3.773 b | 28.89 b | 19.18 | 20.73 | 16.44 | 7.37 b |
| C × M | | | | | | | | | | |
| Ntopia S1 | 78.6 | 1.054 | 29.1 | 9.86 | 2.915 | 42.24 | 29.46 | 79.76 a | 4.91 b | 28.59 |
| Ntopia S2 | 91.7 | 1.170 | 45.0 | 6.98 | 3.024 | 34.87 | 25.3 | 49.86 b | 15.79 a | 19.10 |
| Ntopia S3 | 98.5 | 1.197 | 53.8 | 5.12 | 3.416 | 32.85 | 24.42 | 36.98 bc | 19.65 a | 14.90 |
| Ntopia S4 | 106.5 | 1.159 | 56.4 | 5.41 | 2.951 | 33.23 | 25.74 | 36.16 bc | 18.55 a | 15.76 |
| Ntopia S5 | 110.5 | 1.152 | 52.9 | 4.74 | 2.474 | 32.03 | 24.40 | 34.02 bc | 19.16 a | 13.91 |
| Hercules S1 | 101.1 | 1.992 | 32.1 | 13.66 | 6.524 | 34.74 | 22.79 | 28.51 c | 19.99 a | 10.58 |
| Hercules S2 | 112.9 | 1.958 | 48.6 | 8.01 | 6.670 | 27.37 | 18.62 | 11.91 d | 18.06 a | 3.85 |
| Hercules S3 | 119.4 | 2.057 | 53.6 | 7.25 | 6.188 | 26.28 | 15.87 | 8.29 d | 16.12 a | 2.19 |
| Hercules S4 | 123.4 | 2.120 | 55.3 | 7.33 | 4.705 | 26.11 | 16.20 | 7.37 d | 15.83 a | 1.87 |
| Hercules S5 | 127.8 | 2.020 | 48.8 | 8.39 | 5.371 | 25.76 | 13.95 | 7.44 d | 13.71 a | 0.83 |

*Significant effect at the 0.05 level; **Significant effect at the 0.01 level; ***Significant effect at the 0.001 level. Data represent six replicates of ten fruits for each treatment. Mean comparisons were performed according to Duncan's Multiple Range Test. Means per effect within each column followed by different letters denote significant ($P < 0.05$) differences.

was about half of that demonstrated by 'Hercules', and regression analysis further indicated that while 'Hercules' firmness decreased linearly with maturity, 'Ntopia' was best fitted to a quadratic regression with a firmness peak near S3 (Fig. 1B). Fruit firmness correlated significantly to hundred-seed weight ($r = 0.89$, $p < 0.01$) and the seed-to-pulp weight ratio ($r = 0.73$, $p < 0.05$), which provides a context for interpreting cultivar differences in firmness on the basis of variation in seed weight and progressive fruit growth with maturity (Figs. 1 and 2). Osmotic changes in cell turgor may also influence fruit firmness, while the aptness of the method of mechanical analysis applied is critical for capturing targeted textural changes. For the purposes of the current study, resistance to uniaxial compression was adopted as a measure of fruit firmness that simulates the hand feeling on the fruit; it is the method most appropriate for berries, such as cactus pear, and also avoids direct interference with the seeds, as would be the case with pulp puncture tests (Bourne, 2002).

Variation in juice color was determined by both cultivar and maturity (Table 1). Darker juice, indicated by lower lightness (L*) values, was found in 'Hercules'; in both cultivars significant transition to lower L* was observed only from S1 to S2. Overall juice chroma (C*) was not affected by fruit maturity, while 'Ntopia' exhibited higher chroma than 'Hercules' throughout ripening, owing mostly to higher scores of component b* that denotes yellowness. Juice color transition with maturity was more aptly expressed in hue angle values which characterize the particular hue which color intensity values (C*, a*, b*) refer to. Hue angle values were subject to cultivar-maturity interaction. Higher values designating more yellow-to-orange hue were observed in 'Ntopia' throughout ripening, as opposed to lower values in 'Hercules' designating a more reddish hue (Fig.1C); change in hue with maturity was prolonged in 'Ntopia' and coincided with skin color development, whereas in 'Hercules' hue transition was completed by S2, hence it preceded the appearance of skin color.

### 3.2. Juice acidity, soluble solids and carbohydrates content

Cactus pear is an outstandingly neutral fruit with a range of juice pH 5.5–7.5, hence a fruit of rather insipid acidity (Albano et al., 2015; Cantwell, 1999; Parish & Felker, 1997; Pimienta-Barrios, 1994). However, the balance between sweetness and acidity profoundly affects fruit flavor perception and may be influenced by the presence of even limited acidity (Soteriou et al., 2014). Lack of acidity and abundance of soluble sugars in its juice moreover renders the fruit vulnerable to postharvest microbial attack (Piga et al., 2003; Sáenz, 2013). As the current study demonstrates, the titratable acidity (TA) and the pH of the juice were influenced by both cultivar and maturity, although significant change during ripening of both cultivars was observed only in transition from S1 to S2, which was manifested as a sharp drop in TA with a corresponding rise in pH (Table 2). Cultivar Ntopia demonstrated higher TA and lower pH than 'Hercules' at all stages of maturity, although this difference was maximal at S1 and diminished thereafter. The more perceptible presence of acidity in 'Ntopia' facilitated a more balanced sugar-acid ratio (TA/soluble solids) may partly explain its greater appeal to consumers' taste.

Fruit sweetness is conventionally expressed in terms of the soluble solids content (SSC) of the juice, measured on the basis of the refractive index determined at 20 °C. In most types of fruit the SSC correlates highly with the concentration of soluble carbohydrates. In the current study the SSC correlated significantly with the concentrations of fructose ($r = 0.88$, $p < 0.001$), glucose ($r = 0.88$, $p < 0.001$) and total sugars ($r = 0.90$, $p < 0.001$) in the fruit juice, but not with sucrose. A cultivar-maturity interaction was observed as the two cultivars demonstrated distinct patterns of SSC accumulation with maturity (Table 2; Fig. 1D). In 'Ntopia' accumulation was slower but continued to over-maturity (S5) reaching a maximum of 14.6% and surpassed the maximum of 'Hercules' (13.8%) attained between S3-S4 before complete skin coloration (Fig. 1D).

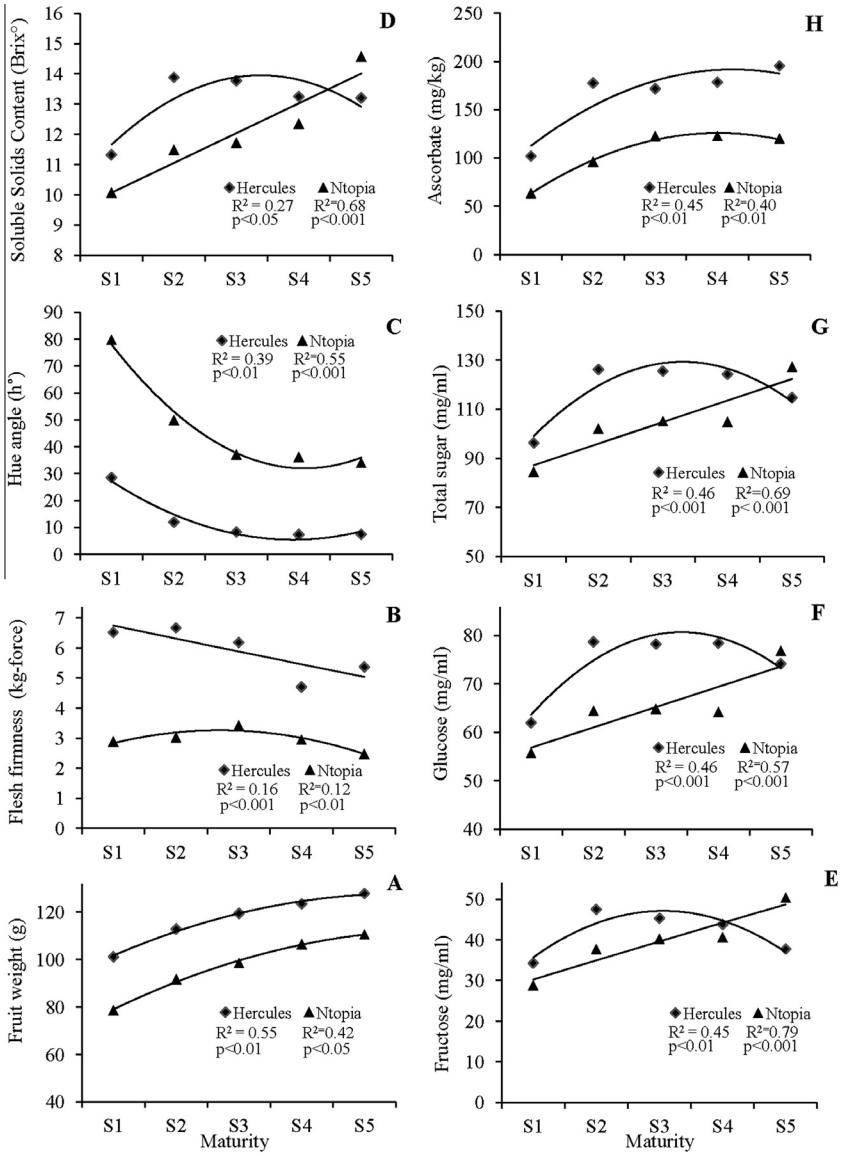


**Fig. 1.** Regression of fruit fresh weight (A), flesh firmness (B), juice hue angle-h° (C) juice soluble solids (D), juice fructose (E), juice glucose (F), juice total sugars content (G), and pulp ascorbate content (H) for cactus pear cultivars 'Ntopia' and 'Hercules' with progressive maturity (S1: mature green; S2: breaker; S3: skin half- colored; S4: skin coloration near complete; S5: full, deep coloration – over-mature). Data points represent means of six replicates consisting of ten sample fruits each.

The SSC range (11–16%) reported for other cactus pear varieties, classifies 'Hercules' and 'Ntopia' as sweet and highly sweet, respectively (Hernádez-Pérez, Carrillo-López, Guevara-Lara, Cruz-Hernádez, & López, 2005; Parish & Felker, 1997; Pimienta-Barrios, 1994). Predominant soluble carbohydrates in the fruit juice were reducing sugars fructose and glucose, while in 'Hercules' alone sucrose was also present in very low concentrations (Table 2). In both cultivars fructose-to-glucose ratio ranged from 1:2 to a near 2:3 at maximum sugar content (Table2; Fig. 1E and F). Predominance of glucose over fructose (3:1) has also been reported for a yellow cultivar by Abdel-Hameed, Nagaty, Salman, & Bazaid (2014) but a near 1:1 ratio for a red cultivar, with sucrose detectible in neither. Cultivar patterns for the evolution of hexoses and total sugars in the current study resembled those obtained for the SSC: a slower but continuous increase with progressive maturation in 'Ntopia' and a clear and symmetric peak shortly after S3 in 'Hercules' (Fig. 1E–G). Interestingly, the sugar peak in 'Hercules' coincides with the appearance of sucrose in this cultivar's sugar profile; but unlike more conventional fruit ripening patterns, where sucrose accumulation takes place at the expense of the two hexoses (Kyriacou, Soteriou, Rouphael, Siomos, & Gerasopoulos, 2015), in 'Hercules' sucrose appears midway through ripening and its presence remains insufficiently low and stable to explain the expenditure of fructose and glucose. It is clear nevertheless that the two cultivars profiled in the current study present differential ripening behavior with respect to sugar content which should be taken into account when deciding on optimum harvest maturity. 'Ntopia' may benefit from harvest performed near full skin coloration (S4–S5) whereas 'Hercules' should optimally be harvested when skin surface is half red (S3) as harvesting at a later stage is likely to deliver fruit depleted of sweetness.

### 3.3. Ascorbic acid, total phenols, betacyanins and betaxanthins

The concentration of ascorbate in the juice of 'Ntopia' and 'Hercules' during ripening averaged 108.6 and 163.1 μg/g pulp, respectively (Table 3). Concentrations in the range of 50–190 μg/g have

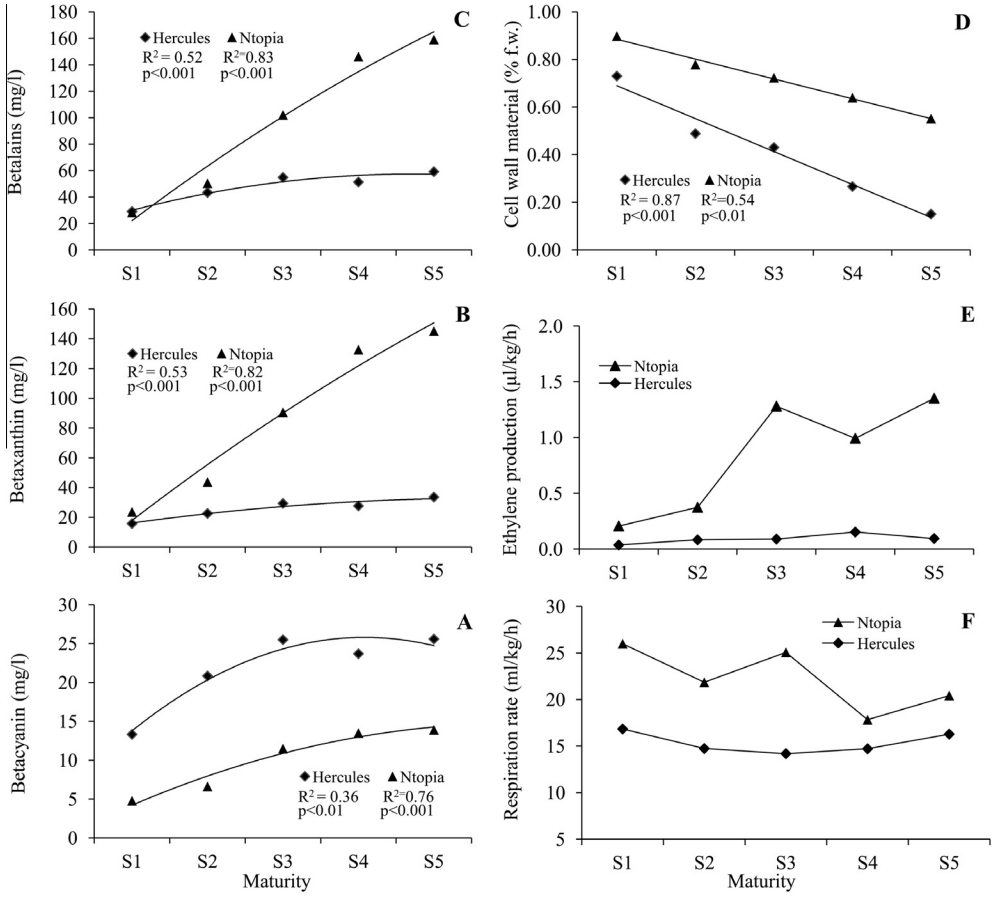


**Fig. 2.** Regression of juice betacyanin (A), betaxanthin (B), and betalain (C), pulp cell wall material (CWM) (D), and changes in ethylene production (E), and respiration rates (F) for cactus pear cultivars 'Ntopia' and 'Hercules' with progressive maturity (S1: mature green; S2: breaker; S3: skin half- colored; S4: skin coloration near complete; S5: full, deep coloration – over-mature). Data points for variables A-D represent means of six replicates consisting of ten sample fruits each. Data points for variables E and F represent means of six replicates each consisting of 4–6 fruits.

**Table 2**
Analysis of variance and mean comparisons for fruit content in soluble solids (SSC), titratable acidity (TA), glucose, fructose, sucrose and total soluble carbohydrates obtained from an orange ('Ntopia') and a red-colored ('Hercules') cultivar at five progressive harvest maturity stages.

| Source of variance | SSC (%) | TA ($g/dm^3$) | pH | Fructose (mg/ml) | Glucose (mg/ml) | Sucrose (mg/ml) | Total (mg/ml) |
|---|---|---|---|---|---|---|---|
| Cultivar (C) | ** | * | *** | | *** | *** | *** |
| Maturity (M) | *** | *** | *** | *** | *** | * | *** |
| C × M | ** | | | *** | ** | * | *** |
| Cultivar | | | | | | | |
| Ntopia | 12.1 | 0.12 a | 5.61 b | 39.6 | 64.7 | 0.00 | 104.3 |
| Hercules | 13.1 | 0.08 b | 6.13 a | 41.7 | 74.3 | 1.4 | 117.4 |
| Maturity | | | | | | | |
| S1 | 10.75 | 0.25 a | 4.52 b | 32.2 | 59.6 | 0.0 | 91.8 |
| S2 | 12.69 | 0.07 b | 6.13 a | 41.7 | 70.2 | 0.0 | 111.9 |
| S3 | 12.75 | 0.06 b | 6.15 a | 41.6 | 69.9 | 1.0 | 112.5 |
| S4 | 12.80 | 0.06 b | 6.20 a | 42.2 | 71.3 | 1.1 | 114.5 |
| S5 | 13.89 | 0.07 b | 6.26 a | 44.1 | 75.5 | 1.4 | 121.0 |
| Cultivar × Maturity | | | | | | | |
| Ntopia S1 | 10.07 d | 0.295 | 3.86 | 28.8 g | 55.7 b | 0.0 b | 84.5 d |
| Ntopia S2 | 11.49 cd | 0.090 | 5.90 | 35.9 ef | 61.7 b | 0.0 b | 97.5 cd |
| Ntopia S3 | 11.72 c | 0.071 | 5.95 | 37.9 def | 61.6 b | 0.0 b | 99.6 c |
| Ntopia S4 | 12.35 bc | 0.072 | 5.98 | 40.6 cde | 64.2 b | 0.0 b | 104.8 bc |
| Ntopia S5 | 14.57 a | 0.089 | 6.08 | 50.4 a | 76.8 a | 0.0 b | 127.2 a |
| Hercules S1 | 11.32 cd | 0.212 | 5.07 | 34.3 fg | 62.0 b | 0.0 b | 96.3 cd |
| Hercules S2 | 13.88 a | 0.045 | 6.35 | 47.5 ab | 78.7 a | 0.0 b | 126.2 a |
| Hercules S3 | 13.77 ab | 0.042 | 6.35 | 45.3 abc | 78.2 a | 2.0 a | 125.5 a |
| Hercules S4 | 13.25 ab | 0.047 | 6.43 | 43.8 bcd | 78.4 a | 2.1 a | 124.3 a |
| Hercules S5 | 13.21 ab | 0.048 | 6.43 | 37.8 def | 74.2 a | 2.7 a | 114.7 ab |

*Significant effect at the 0.05 level; **Significant effect at the 0.01 level; ***Significant effect at the 0.001 level. Data represent six replicates of ten fruits for each treatment. Mean comparisons were performed according to Duncan's Multiple Range Test. Means per effect within each column followed by different letters denote significant ($P < 0.05$) differences.

**Table 3**
Analysis of variance and mean comparisons for antioxidant capacity (DPPH), total phenolic, betacyanin, betaxanthin and total betalains content of juice and ascorbate content of pulp obtained from an orange ('Ntopia') and a red-colored cultivar ('Hercules') at five progressive fruit harvest maturity stages.

| Source of Variance | Phenolics (mg/l GAE) | Ascorbate (mg/kg) | Betacyanins (mg/l) | Betaxanthins (mg/l) | Betalains (mg/l) |
|---|---|---|---|---|---|
| Cultivar (C) | ** | *** | *** | *** | *** |
| Maturity (M) | | *** | ** | *** | *** |
| C × M | | * | * | *** | *** |
| *Means* | | | | | |
| Cultivar | | | | | |
| Ntopia | 466.0 a | 108.6 b | 10.5 b | 95.7 a | 103.2 a |
| Hercules | 333.2 b | 163.1 a | 21.6 a | 26.3 b | 49.7 b |
| Maturity | | | | | |
| S1 | 369.8 | 87.5 b | 10.1 b | 18.7 d | 28.8 d |
| S2 | 395.8 | 137.0 a | 13.7 b | 33.2 d | 46.9 c |
| S3 | 401.7 | 147.1 a | 18.5 a | 60.0 c | 78.5 b |
| S4 | 395.7 | 150.6 a | 18.6 a | 80.2 b | 98.7 a |
| S5 | 427.2 | 153.0 a | 19.1 a | 95.6 a | 114.5 a |
| C × M | | | | | |
| Ntopia S1 | 413.7 | 63.5 c | 4.8 d | 23.7 cd | 28.4 d |
| Ntopia S2 | 466.8 | 96.0 bc | 6.6 cd | 43.8 c | 50.4 cd |
| Ntopia S3 | 481.8 | 122.5 b | 11.5 bcd | 90.6 b | 102.1 b |
| Ntopia S4 | 465.8 | 122.7 b | 13.5 bc | 132.7 a | 146.2 a |
| Ntopia S5 | 493.3 | 120.1 b | 13.9 b | 145.2 a | 159.1 a |
| Hercules S1 | 333.2 | 101.9 bc | 13.3 bc | 15.8 d | 29.1 d |
| Hercules S2 | 324.9 | 177.7 a | 20.8 a | 22.6 cd | 43.4 cd |
| Hercules S3 | 321.5 | 171.7 a | 25.5 a | 29.4 cd | 54.8 cd |
| Hercules S4 | 325.5 | 178.4 a | 23.7 a | 27.6 cd | 51.3 cd |
| Hercules S5 | 361.1 | 195.3 a | 25.6 a | 33.6 cd | 59.2 c |

*Significant effect at the 0.05 level; **Significant effect at the 0.01 level; ***Significant effect at the 0.001 level. Data represent six replicates of ten fruits for each treatment. Mean comparisons were performed according to Duncan's Multiple Range Test. Means per effect within each column followed by different letters denote significant ($P < 0.05$) differences.

been reported in yellow, red and orange clones of *O. ficus-indica* and as high as 458 μg/g in green-skinned clones (Kuti, 2004; Martínez et al., 2014; Pimienta-Barrios, 1994); significantly higher concentrations (768 μg/g) have been encountered in fruits of *Opuntia xoconostle* (Sáenz & Sepúlveda, 2001). Notwithstanding cultivar variation, the fruits of *O. ficus-indica* may be characterized as a good source of vitamin c, comparable to tomato (160 μg/g), higher than peaches (66 μg/g) but lower than oranges (485 μg/g), as referred in the literature (USDA, 2015). Mean comparisons revealed that in both cultivars studied, the most significant increase in ascorbate content was incurred during transition from green stage S1 to the appearance of skin color at S2; incremental increase during this transition was lower in 'Ntopia' (51.2%) than in 'Hercules' (74.4%). Regression analysis also depicted a slower increase with maturity in 'Ntopia' although both cultivars reached maximum ascorbate content near S4 (Fig. 1H). Hence maximum ascorbate content in cactus pear appears to coincide with full pulp maturity and near complete skin coloration.

Betalains are aromatic indole derivatives not related chemically to flavonoids nor synthesized via the phenylpropanoid pathway (Raven, Evert, & Eichhorn, 2004). They constitute an important bioactive component of cactus pear fruit with antioxidant and possibly antimicrobial action (El-Mostafa et al., 2014; Stintzing et al., 2005). The betacyanins, betaxanthins and total betalains of cactus pear fruit in the current study were influenced by both cultivar and maturity but the two factors also interacted (Table 3). The mean betacyanin content of the fruit juice, responsible for red and purple coloration of the pulp, was almost double in red-fleshed 'Hercules' (21.6 mg/l) than in orange-fleshed 'Ntopia' (10.5 mg/l). On the contrary, the mean betaxanthin content, responsible for the orange and yellow coloration, was almost four times higher in orange-fleshed 'Ntopia' (95.7 mg/l) than in 'Hercules' (26.3 mg/l), which also resulted in double the betalains content in 'Ntopia' (103.2 mg/l) than in 'Hercules' (49.7 mg/l). Betacyanin, betaxanthin and total betalains contents of the juice increased with fruit maturity in accordance to pulp color development (Figs. 2A–C; Supplemental Material). Regression analysis revealed a slower increase with progressive maturity in the betacyanin content of 'Ntopia' ($R^2 = 0.75$, $p < 0.001$) than of 'Hercules' ($R^2 = 0.36$, $p < 0.01$). Moreover, the betacyanin content of 'Ntopia' increased throughout ripening whereas in 'Hercules' it peaked near full maturity (S4) and before full skin coloration (Fig. 2A). This underlines the disparity between the early pulp pigmentation and late skin coloration characteristic of 'Hercules', which renders external color a rather poor commercial maturity index for this cultivar. Regression analysis also depicted an increase in betaxanthin content throughout ripening for 'Ntopia' ($R^2 = 0.82$, $p < 0.001$), whereas in 'Hercules' increase ($R^2 = 0.53$, $p < 0.001$) was minimal and restricted to the early stages of maturity (Fig. 2B). A similar pattern to that of betaxanthins was observed also for betalains, wherein a slow climax was reached by S4 in 'Hercules' ($R^2 = 0.52$, $p < 0.001$) as opposed to a steep quadratic ($R^2 = 0.83$, $p < 0.001$) increase persistent to over-maturity (S5) in 'Ntopia' (Fig. 2C). Owing to the presence of phenolic hydroxyls in their molecular constitution, betacyanins have been more correlated to the phenolic content of cactus pear juice than betaxanthins (Stintzing et al., 2005). The low proportional content of betacyanins to betaxanthins in 'Ntopia', the absence of significant betacyanin increase beyond S2 in 'Hercules', and the contribution of non-betalain fractions to the phenolic content may explain the absence of significant maturity effect on the phenolic content of the two cultivars examined (Table 3). Mean (S1–S5) phenolic content was however higher by 39.9% in 'Ntopia' (466.0 μg/mL) than in 'Hercules' (333.2 μg/mL) and this difference was stable throughout ripening, indicating that the phenolic content in cactus pear fruit is formed early on during ripening (Table 3). Similar phenolic content has been reported at harvest for tree-ripened commercially mature fruit of Italian yellow-orange cv. Gialla by Allegra et al. (2015) although subsequent storage resulted in depletion of phenolic content. Both cultivars however, highlight the rich phenolic content of cactus pear pulp and juice, which imparts significant anti-inflamatory, cardioregulatory, and anti-neurodegenerative proper-

ties (El-Mostafa et al., 2014). Cultivar variation in the phenolic content of cactus pear fruit is further influenced by environmental or cultural factors, such as supplemental irrigation. Accordingly, Albano et al. (2015) reported significantly higher phenolic contents of 892 and 698 μg/g in non-irrigated purple and orange genotypes, respectively, from Apulia, Italy.

### 3.4. Cell wall material, solubilized pectins and neutral sugars

In both cultivars the CWM diminished linearly with progressive maturity, at a lower however rate in 'Ntopia' (Table 4; Fig. 2D). Cultivar 'Ntopia' yielded a higher mean CWM than 'Hercules' (Table 4). The lower CWM of 'Hercules' pulp (0.409% f.w.), in comparison to 'Ntopia' (0.718% f.w.), is in contrast to its nearly double fruit firmness which reflects rather its higher seed-to-pulp ratio (Tables 1 and 4). The alcohol-insoluble residue (CWM) of fresh cactus pear pulp was found comparable to that of ripe kiwi, tomato and watermelon (Redgwell et al., 1997).

The water-soluble fraction (WSF) of CWM, which contains solubilized polyuronides of high molecular weight attached to the cell wall through ionic bonds (Brummell & Labavitch, 1997), incurred a significant cultivar effect and accounted for 35.6% and 45.2% of total pectins in 'Ntopia' and 'Hercules', respectively (Table 4). However, the effect of maturity on this pectin fraction was non-significant, which indicates that textural changes in cactus pear during ripening may not relate to polyuronide solubilization and consequent cell wall hydration and swelling, as reported for strawberry, watermelon, tomato and certain apple cultivars (Brummell, 2006; Redgwell et al., 1997). Moreover, the carbonate-soluble (NSF) pectin fraction, containing de-esterified low molecular weight pectins attached to the cell wall by covalent bonds, and the alkali-soluble fraction (KSF), containing extracted matrix glycans tightly bound to the wall by hydrogen bonds (Brummell & Harpster, 2001), were influenced by neither cultivar nor maturity (Table 4). Despite the lower distribution of WSF pectins in 'Ntopia' and the absence of cultivar differentiation in the NSF and KSF fractions, the total pectin content of 'Ntopia' (159.0 mg/g CWM) expressed in uronic acid equivalents, was significantly higher than 'Hercules' (114.8 mg/g CWM). This underlines the significant contribution of pectins to the cell wall matrix of cactus pear fruit, particularly in 'Ntopia', but also highlights the substantial pectinic content present in the pulp besides the peel of this fruit (Habibi, Heyraud, Mahrouz, & Vignon, 2004).

Cell wall neutral sugars, mainly galactose, arabinose and xylose, are essential components of matrix glycans (hemicelluloses) which possess extensive branching and interweaving with cellulose microfibrils and interlock the pectin and cellulose network (Brummell, 2006; Zykwinska, Ralet, Garnier, & Thibault, 2005). Progressive loss of WSF neutral sugars, mainly from arabinan and galactan side chains of matrix glycans, has been associated with changes in cell wall porosity that may affect pectin solubilization and fruit texture (Brummell, 2006). The WSF neutral sugars content of CWM in cactus pear was affected by both cultivar and maturity, and accounted for 48.0% of total neutral sugars in 'Hercules' and 33.3% in 'Ntopia' (Table 4). Cultivar-maturity interaction was attributed to a more prominent reduction of WSF sugars from S1 to S2 in 'Ntopia' rather than 'Hercules' (data not shown). Maturity effect was absent in the NSF fraction despite cultivar differences in its percentage content. Strong bonding of glycans to the cell wall matrix by hydrogen bonds makes changes in neutral sugars with maturity mostly visible in the alkali-soluble fraction of the CWM (Brummell & Harpster, 2001). Release of neutral sugars from the matrix is usually manifested in the late stages of ripening and coincides with extensive fruit softening (Rose, Hadfield, Labavitch, & Bennett, 1998). In the case of cactus pear a sharp release of neutral sugars was observed from S1 to S2 (Table 4). This indicates that significant remodeling of the cell wall structure takes place at the early stages of ripening, right after the mature-green stage. This may affect the pulp texture of the fruit though not directly reflected in fruit compressibility tests due to the mechanical supportive role of the heavily interspersed seeds. The current work highlights a unique behavior of cactus pear with respect to cell wall metabolic events and concomitant textural changes during ripening: it demonstrates no pectin solubilization, as seen in crisp fruit such as the watermelon, but also extensive loss of glycan bonding and loss of neutral sugars, as seen in ripe soft-textured fruits like melon (Brummell, 2006); however, unlike cactus pear the melon develops a soft melting texture with maturity as cell-to-cell adhesion is reduced by breakdown of pectin polymers in the middle lamella, and pectin solubilization also leads to progressive hydration, swelling and softening of the cell wall (Brummell & Harpster, 2001; Crookes & Grierson, 1983; Redgwell et al., 1997).

Irrespective of its linkage to ripening-related fruit softening events, the pectin content of cactus pear fruit, particularly as readily available soluble dietary fiber, enhances its functional importance for the human diet (Bensadón, Hervert-Hernández, Sáyago-Ayerdi, & Goñi, 2010). Dietary consumption of water-soluble pectins has been associated with improved status of intestinal microbiota (Schneeman, 2001), while pectin has been shown to lower the relative percentages of free and esterified cholesterol in low

**Table 4**
Analysis of variance and mean comparisons for cactus pear fruit content in total cell wall material (TCWM), and water soluble (WSF), sodium carbonate soluble (NSF) and 4 N KOH soluble (KSF) fractions of pectins and neutral sugars obtained from an orange and a red colored cultivar at five progressive harvest maturity stages.

| Source of Variance | TCWM | % Total pectins | | | % Total neutral sugars | | |
|---|---|---|---|---|---|---|---|
| | (% FW) | WSF | NSF | KSF | WSF | NSF | KSF |
| Cultivar (C) | *** | * | | | *** | ** | |
| Maturity (M) | *** | | | | ** | | ** |
| C × M | | | | | *** | | |
| Cultivar | | | | | | | |
| Ntopia | 0.718 a | 35.6 b | 38.7 | 25.7 | 33.3 b | 35.2 a | 31.4 |
| Hercules | 0.409 b | 45.2 a | 32.4 | 22.4 | 48.0 a | 25.6 b | 26.4 |
| Maturity | | | | | | | |
| S1 | 0.814 a | 46.5 | 39.1 | 24.4 | 49.8 a | 35.9 | 14.3 b |
| S2 | 0.634 b | 39.6 | 34.8 | 25.5 | 36.6 b | 29.8 | 33.6 a |
| S3 | 0.544 bc | 40.4 | 34.4 | 26.0 | 42.5 ab | 26.9 | 30.5 a |
| S4 | 0.453 cd | 37.7 | 34.8 | 27.5 | 35.5 b | 30.7 | 33.8 a |
| S5 | 0.351 d | 37.5 | 33.6 | 28.1 | 39.4 b | 26.8 | 33.8 a |

*Significant effect at the 0.05 level; **Significant effect at the 0.01 level; ***Significant effect at the 0.001 level; n.s. non-significant (P greater than 0.05). Data represent six replicates of ten fruits for each treatment. Mean comparisons were performed according to Duncan's Multiple Range Test. Means per effect within each column followed by different letters denote significant (P < 0.05) differences.

density lipoprotein was lower in animals fed on diet rich in (*Opuntia* spp.) pectin (Fernandez, Lin, Trejo, & McNamara, 1992). The mean WSF pectin content of the cultivars profiled in the current study was 40.4 mg galacturonic acid equivalents/ g CWM, compared to 23.1 and 69.0 mg/ g in ripe watermelon and melon, respectively (Karakurt & Huber, 2002; Rose et al., 1998).

### 3.5. Ethylene production and respiratory activity

Seasonal variation in the postharvest respiration rates of cactus pear fruit has recently been demonstrated (Allegra et al., 2015), as well as certain physicochemical changes associated with postharvest ripening (Hernádez-Pérez et al., 2005). However, the physiological maturation of cactus pear fruit is considered terminated at harvest and the maturation process, either on-tree or postharvest, is not characterized by climacteric rise in respiration and ethylene production (Cantwell, 1999). The current study verifies the non-climacteric nature of this fruit but also highlights important cultivar variation in respiration and ethylene production rates which may prove valuable for its postharvest handling (Fig. 2E and F). The respiration and ethylene production rates demonstrated throughout ripening classify the cactus pear as a fruit of low respiratory activity and low ethylene production. Cultivar differences however were significant in terms of mean respiration rate ('Ntopia': 22.2 ml $CO_2$ $kg^{-1}$ $h^{-1}$; 'Hercules': 15.3 ml $CO_2$ $kg^{-1}$ $h^{-1}$) but they tended to diminish with progressive maturity (Fig. 2F). 'Ntopia' also had a higher mean ethylene production rate (0.841 μl $kg^{-1}$ $h^{-1}$) and a different ethylene production profile with maturity than 'Hercules' (0.091 μl $kg^{-1}$ $h^{-1}$). 'Hercules' showed a very low and stable ethylene production rate throughout ripening as opposed to 'Ntopia' which demonstrated an ethylene spike between S2–S3 (Fig. 2E). The observed differences between the two cultivars likely relate to their overall differences in metabolic activity and compositional changes associated with ripening, especially for soluble carbohydrates and betalains, as discussed above. These differences may also lay the foreground for cultivar variation in postharvest performance, which deserves further investigation.

## 4. Conclusion

The two cultivars studied presented differential sugar accumulation behavior during ripening with significant implications for their harvest maturity: 'Ntopia' should be harvested near full skin coloration and 'Hercules' when fruit skin is half-colored. Disparity between pulp betacyanin accumulation and delayed skin coloration renders skin color a dubious harvest maturity index for red-fleshed cultivar 'Hercules'. The current work highlights the unique behavior of cactus pear with respect to cell wall metabolic events and textural changes during ripening: no pectin solubilization but weakening of glycan bonding and extensive loss of neutral sugars; while further complication in interpreting fruit compressibility is the high correlation of seed weight and seed-to-pulp ratio to uniaxial fruit compression analysis.

## Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.foodchem.2016.05.113.

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