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Changes in the bioactive compounds and antioxidant activity in red-fleshed dragon fruit during its development

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ABSTRACT

Red-fleshed dragon fruit (*Hylocereus polyrhizus*) is a promising food with a functional appeal. This study investigated the changes in the bioactive compounds and antioxidant activity of the red-fleshed dragon fruit at eight development stages. In general, the levels of total phenolic compounds tested using Folin-Ciocalteu, and Fast Blue BB reagents, betacyanin, betaxanthin, anthocyanins, and antioxidant activity by TEAC, FRAP, and ß-carotene bleaching increased over the fruit development stages, whereas vitamin C content significantly decreased. Six phenolic compounds were identified, including catechin, vanillin, gallic acid, caffeic acid, chlorogenic acid, and ferulic acid. Catechin was the majority compound, followed by vanillin. All these compounds decreased during fruit development; chlorogenic and ferulic acids were only detected 30 days after anthesis. Based on the results, the suitable harvest period of red-fleshed dragon fruit is between 36 and 38 days after anthesis.

1. Introduction

In light of significant global population growth and the consumer's awareness of the health-promoting effects of fruits, the demand for antioxidant-rich fruits has been remarkably increased in domestic and international markets (Arivalagan et al., 2021; Molla et al., 2021). Popularly known as pitaya or pitahaya, the red-fleshed dragon fruit (*Hylocereus polyrhizus*), a member of the Cactaceae family, is a non-climacteric exotic fruit species native to central Mexico and South America (Jiang et al., 2020). Over the last years, the cultivation and consumption of the red-fleshed dragon fruit have increased in many tropical and sub-tropical regions of the world, including Brazil, owing to its unique flavor, pleasant taste, exotic appearance, promising nutritional and functional properties (Magalhães et al., 2019; Thaiudom et al., 2021). It provides a broad spectrum of vitamins, lycopene, minerals, and carbohydrates, particularly reducing sugars, including glucose and fructose (Joshi and Prabhakar, 2020; Le Bellec et al., 2006). The functional appeal of the red-fleshed dragon fruit has been attributed to the presence of soluble and insoluble fiber and several phytochemicals, including carotenoids, phenolic compounds, and the purple-red pigments, betalains, present in the peel and also in the pulp, which are considered to play an essential rule in antioxidant activity (Choo and Yong, 2011; Fathordoobady et al., 2016; Leong et al., 2018). In previous *in vitro* and *in vivo* studies, these phytochemicals from *Hylocereus polyrhizus* extracts (pulp and peel) demonstrated a wide range of biological properties such as anti-inflammatory, anti-spasmodic, radioprotective (Kaur et al., 2018), antioxidant, antimicrobial, anti-cancer, and anti-diabetic activities (Joshi and Prabhakar, 2020). Besides, a recent study conducted by Ravichandran et al. (2021), highlighted *Hylocereus polyrhizus* as a promising natural source of anti-glycation and antioxidant compounds that may have the potential of reducing the risk of glycation associated with diabetic and aging disorders. A previous study of our group (Magalhães et al., 2019) reported significant physicochemical changes in the red-fleshed dragon fruit during its development, such as an increase in soluble solids, pH, diameter, and mass, as well as a decrease in firmness, skin thickness, and acidity. Although the chemical composition and the functional potential of *Hylocereus polyrhizus* have been extensively studied with ripe fruits (Al-Mekhlafi et al., 2021; Fathordoobady et al., 2021; Ho et al., 2020; Madane et al., 2020; Ravichandran et al., 2021), studies focusing on the changes in the bioactive compounds of *Hylocereus polyrhizus* during development are still lacking. Several authors emphasized that the biosynthesis, contents, and the

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bioactivities of the bioactive compounds might be influenced by various factors, such as locations, agricultural practices, rainfall, air temperature, species, soil characteristics, edaphoclimatic conditions, plant tissues, cultivar, and developmental stages (Gominho et al., 2018; Li et al., 2020; Zayed et al., 2020; Zuo et al., 2012). Despite the existence of many factors that influence the profile of bioactive compounds of fruits, a deeper understanding of the changes in these compounds during the development of the red-fleshed dragon fruit may be essential to determine the best moment for harvesting and consumption of high-quality red-fleshed dragon fruit with good nutritional value and the highest functional appeal. Therefore, this study aimed to evaluate the bioactive compounds of the red-fleshed dragon fruit at different development stages to provide more details about the most suitable harvest period.

2. Material and methods

2.1. Fruit material and experimental site

Field research was carried out in an experimental orchard at Federal University of Lavras (UFLA), Lavras, Minas Gerais, Brazil (21° 14′ 06″ S 45° 00′ 00″ W; 918 m of altitude) between March and April 2016. The climate is high altitude subtropical with dry winters and rainy summers. The total precipitation during the evaluation period was 140 mm, whereas the mean values for maximum and minimum temperature were about 28.2 and 16.3 °C, respectively. The red-fleshed dragon fruit samples were obtained as detailed in a previous study of our group (Magalhães et al., 2019). A total of 60 red-fleshed dragonfruits (*Hylocereus polyrhizus*) from plants two -years old, selected based on visual uniformity of fully opened flowers in the middle part, were marked in a single day at flowering time. The fruit samples from marked inflorescences were harvested at eight development stages: 28, 30, 32, 34, 36, 38, 40, and 42 days after anthesis (DAA) as shown in Fig. 1 The fruits were collected in the morning and immediately transported to the Laboratory of Fruit and Vegetables, Department of Food Science, Federal University of Lavras. Afterward, the samples were washed with potable tap water, sanitized with sodium hypochlorite 200 mg/L for 15 min, wiped dry with a paper towel, and manually peeled. After peeling, the fruit pulp was frozen in liquid nitrogen and stored at −80 °C until the analysis.

2.2. Treatments and experimental design

The treatments consisted of red-fleshed dragon fruits collected at eight different development stages (28, 30, 32, 34, 36, 38, 40, and 42 DAA) arranged in a completely randomized design with three repetitions. Each experimental plot consisted of three fruits.

2.3. Chemicals and reagents

Folin-Ciocalteu reagent, Fast Blue BB reagent, 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox), potassium persulfate, 2,2′-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS), 2,4,6-tri (2-pyridyl)-striazine (TPTZ), iron (III) chloride hexahydrate, sodium acetate trihydrate ($C_2H_3NaO_2{\cdot}3H_2O$), iron (II) sulfate heptahydrate ($FeSO4{\cdot}7H_2O$), linoleic acid, trans-β-carotene, Tween 40 emulsifier, chloroform, methanol (HPLC grade), commercial phenolic standards of gallic acid, catechin, chlorogenic acid, caffeic acid, vanillin, *p*-coumaric acid, quercetin, and *trans*-cinnamic acid were obtained from Sigma-Aldrich (St. Louis, MO, USA); *o*-coumaric acid, ferulic acid, *m*-coumaric acid, and rutin were purchased from Fluka Chemie (Steinheim, Germany). Ascorbic acid, oxalic acid, gallic acid, acetone, methanol, ethanol, sodium carbonate (Na_2CO_3), sodium hydroxide (NaOH), glacial acetic acid, and hydrochloric acid were purchased from Êxodo (Brazil). All chemicals and reagents were of analytical grade, and the ultra-pure water was obtained from a Milli-Q system (Millipore, Bedford, MA, USA).

2.4. Extraction and determination of vitamin C content

The content of vitamin C, known as the sum of ascorbic acid (AA) and dehydroascorbic acid (DHA), was determined by the colorimetric method established by Strohecker et al. (1967). For extraction, 5 g of red-fleshed dragon fruit pulp were mixed with 45 mL oxalic acid (5%, w/v) and 0.1 g of Kieselgur; the mixture was kept under mechanical agitation 5 min. Subsequently, the extract was filtered using filter paper with 14 μm porosity, and the vitamin C was measured at 520 using a UV/Vis Spectrophotometer (Varian, Cary 50 Probe); the results were reported as milligrams of ascorbic acid per 100 g of fresh mass (mg AA/100 g FM).

2.5. Extraction of phenolic compounds

Total phenolic compounds and antioxidant activity were measured in extracts obtained according to the methodology described by Gonçalves et al. (2019), with minor modifications. Briefly, 2.5 g of each sample were extracted with 10 mL aqueous methanol (50%, v/v), and immediately sonicated in an ultrasonic bath for 30 min. Subsequently, they were centrifuged at 25,400 × g for 15 min under 4 °C, and the supernatant was collected. The residue was re-extracted with an additional 10 mL of aqueous acetone (70%, v/v) under the same conditions. The two obtained supernatants were combined and filtered through filter paper with 14 μm porosity and stored at −80 °C until analysis.

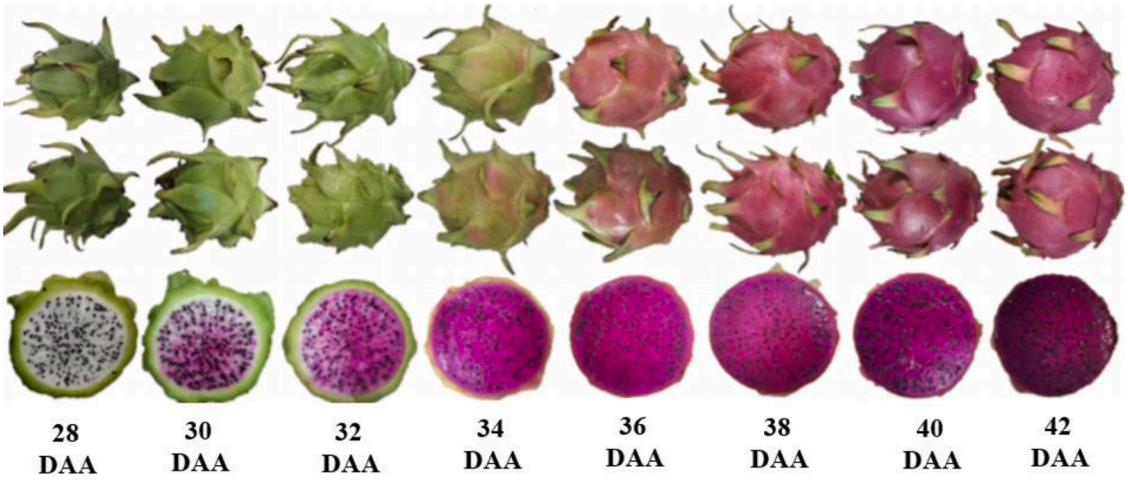

Fig. 1. Image of red-fleshed dragon fruit at eight different development stages. **DAA:** Days after anthesis.

2.6. Determination of total phenolic content (TPC)

The total phenolic content (TPC) of extracts was evaluated by Folin-Ciocalteu (Pinto et al., 2021) and Fast Blue BB (FBBB) (Medina, 2011) colorimetric methods, with some modifications. For Folin-Ciocalteu, 30 μL of extract was mixed with 150 μL of Folin-Ciocalteu reagent (10%, v/v) and 120 μL of sodium carbonate (4%, w/v). The absorbance was read at 720 nm after incubation in darkness for 2 h. For Fast Blue BB, 200 μL of the extract was mixed with 20 μL of Fast Blue BB reagent (0.1%, v/v) and 20 μL of sodium hydroxide (5%, w/v), and the absorbance was measured at 420 nm after 1.5 h of incubation in darkness. All measurements were carried out in triplicate, using a 96-well microplate reader (Biochrom EZ Read 2000). The results were reported as milligram gallic acid equivalents per 100 g of fresh weight of the sample (mg GAE/100 g FM).

2.7. Determination of antioxidant activity

2.7.1. Ferric-reducing antioxidant power (FRAP)

The ferric reducing antioxidant power (FRAP) of extracts was determined based on the reduction of a ferric complex (Fe^{3+}-TPTZ) to the ferrous form (Fe^{2+}-TPTZ) in the presence of antioxidants, according to the method described by Pulido et al. (2000), with minor modifications. FRAP reagent was prepared by mixing 40 mM TPTZ (diluted in 40 mM HCl), 300 mM of acetate buffer (pH 3.6), and 20 mM of $FeCl_3$ in a ratio of 10:1:1 (v/v/v). An aliquot (9 μL) of the extract was mixed with 269 μL of FRAP reagent and 27 μL distilled water in a 96-well microplate, and the mixture was incubated at 37 °C for 30 min. The absorbance was assessed at 595 nm using a microplate reader (Biochrom EZ Read 2000). For calibration, a five-point standard curve (0—2000 uM) was prepared using ferrous sulfate ($FeSO_4 \bullet 7H_2O$) as the reference, and the results were reported as μM ferrous sulfate per gram of fresh mass (μM $FeSO_4$/g FM).

2.7.2. Trolox equivalent antioxidant capacity (TEAC)

The Trolox equivalent antioxidant capacity (TEAC) was determined based on the reduction of $ABTS^{\circ+}$ by antioxidants as reported by Re et al. (1999), with slight modifications. The working solution of $ABTS^{\circ+}$ was generated by reacting 5 mL of ABTS solution (7 mM) with 88 μL of $K_2S_2O_8$ solution (2.45 mM) for 16 h at room temperature in darkness. Before analysis, the resulted stock solution ($ABTS^{\circ+}$) was diluted with ethanol to an absorbance of 0.70 ± 0.05 at 734 nm using a microplate reader (Biochrom EZ Read 2000). The extracts (3 μL) were mixed with 297 μL of $ABTS^{\circ+}$ diluted solution, and after six minutes, the absorbance was read at 734 nm using the microplate reader. A five-point standard curve (100–2000 μM) was built using Trolox as the reference standard, and the results were expressed as μM Trolox Equivalent per gram of fresh mass (μM TE/g FM).

2.7.3. β-carotene bleaching activity

The β-carotene bleaching activity was performed using the previous method with some modifications (Kassim et al., 2013). This method is based on the ability of antioxidants to inhibit the β-carotene discoloration induced by the conjugated diene hydroperoxides originated from the oxidative degradation of linoleic acid (Bouaziz et al., 2015). In brief, 50 μL β-carotene solution (1 mg/mL in chloroform) was mixed with 40 μL linoleic acid and 530 μL Tween 40. After the chloroform has been wholly evaporated at 40 °C using a vacuum pump, distilled aerated water was added with vigorous shaking to form an emulsion. The absorbance was immediately adjusted to values between 0.6 and 0.70 at 470 nm using a microplate reader (Biochrom EZ Read 2000). An aliquot of 270 μL from the emulsion was mixed with 20 μL of extracts in the 96-well microplate followed by incubation at 40 °C in a water bath, for 2 h in darkness. The absorbance was read at 470 nm using a microplate reader at zero time and after 2 h of incubation. The antioxidant activity expressed as a percentage of oxidation inhibition (OI) was determined according to the formula reported below:

$$OI(\%) = 1 - [(Abs_{t=0} - Abs_{t=2}) / (Abs_{c=0} - Abs_{c=2})] \text{ x } 100 \quad (1)$$

Where $Abs_{t=0}$ and $Abs_{t=2}$ is the absorbance of the test samples read at 0 and 2 h, respectively; $Abs_{c=0}$ and $Abs_{c=2}$ is the absorbance of the control (ß-carotene emulsion and water) read at 0 and 2 h, respectively.

2.8. Individual phenolic compounds by HPLC-DAD

2.8.1. Extraction

The individual phenolic compounds were extracted according to the method described by Ramaiya et al. (2013), with slight modifications. Briefly, 2.5 g of the sample were extracted with 20 mL aqueous methanol HPLC grade (70%, v/v) followed by sonication in an ultrasonic for 1 h at room temperature. Then the extracts were centrifuged at 8.832 × g for 15 min at 4 °C, filtered (14 μm), and stored at −80 °C until HPLC analysis.

2.8.2. Identification and quantification

The identification of the individual phenolic compounds was carried out using HPLC-DAD/UV–Vis (Shimadzu Corporation, Kyoto, Japan) consisting of a quaternary pump (LC-20AT) coupled to a DAD detector (SPD-M20A), a vacuum degasser (DGU-20A5), a column oven (CTO-20AC), and an auto-sampler (SIL-20A). The phenolic compounds were separated using Shimadzu Shim-pack ODS GVP (4.6 × 250 mm) C18 column coupled to a Shimadzu-pack ODS GVP (4.6 × 10 mm, 5 μm) C18 guard column under the same conditions described by Gonçalves et al. (2019). The mobile phases were composed of 2% acetic acid (v/v, solvent A) and 70:28:2 methanol/water/acetic acid (v/v/v, solvent B). The flow rate was 1.0 mL min^{-1} with a gradient elution program and run time of 60 min. The injection volume was 20 μL, and the phenolic compounds were detected at 280 nm. Before injection, all samples were filtered (0.45 μm), and the analysis was carried out in triplicate at 15 °C. Phenolic compounds were identified according to their retention times, UV–Vis absorptive spectra, comparing with authentic standards. The results were reported as milligram per 100 g of fresh mass (mg/100g FM).

2.9. Extraction and determination of betalains

Extraction and determination of betalains contents were performed according to a previous method by Qingzhu et al. (2016), with slight modifications. In brief, 0.5 g of fresh red-fleshed dragon fruit pulp previously ground in liquid nitrogen was first mixed with 5 mL aqueous methanol (80%, v/v) followed by an ultrasound extraction for 10 min at room temperature. Subsequently, after 20 min of stirring in darkness at room temperature, the extracts were filtered (14 μm), and the residues were re-extracted one more time under the same conditions described above. The two extracts were combined, and the analysis was performed in triplicate using a microplate reader (Biochrom EZ Read 2000). Betacyanin and betaxanthin contents were calculated according to the formulas given below:

$$\text{Betacyanin (mg / 100 g)} = (A_{538} \text{ x DF x MM x V x 100})/(\varepsilon \text{ x P x l}) \quad (2)$$

$$\text{Betaxanthin (mg / 100 g)} = (A_{483} \text{ x DF x MM x V x 100})/(\varepsilon \text{ x P x l}) \quad (3)$$

Where A_{538} is absorbance for betacyanin at 538 nm; A_{483} is absorbance for betaxanthins; DF is a dilution factor; MM is the molecular mass (550 g/mol for betanin and 308 g/mol for indicaxanthin); V is the pigment solution volume (mL); Ɛ is the molar extinction coefficient (60,000 L/mol.cm for betanin and 48,000 L/mol.cm for indicaxanthin), and l is the length of the cell (1 cm); P is the fresh pigment mass (g).

2.10. Extraction and determination of total anthocyanins

Total anthocyanins contents were extracted and measured using the method described by Fuleki and Francis (1968), with slight modifications. 5 g fresh of red-fleshed dragon fruit pulp was mixed with 25 mL of hydrochloric-acidified (0.1%, v/v) ethanol, vortexed, and incubated for 1 h at room temperature in darkness. Then, the extract was filtered through Whatman filter paper No.4, and the final volume was adjusted to 50 mL with hydrochloric-acidified (0.1%, v/v) ethanol. The analyzes were carried out in triplicate, and the absorbance was measured at 535 nm using a microplate reader (Biochrom EZ Read 2000). The total anthocyanin content (AC) was expressed as milligrams of cyanidin-3-glucoside equivalents per 100 g of sample, according to the formula given below:

$$AC(mg/100g) = [(A \times MM \times DF)/(\varepsilon \times l)] \times 100 \quad (4)$$

Where A is the absorbance; MM is the molecular weight of cyanidin-3-glucose (449.2 g/mol); DF is the dilution factor; Ɛ is the molar absorptivity (26,900 L.mol^{-1}), and l is the length of the cell (1 cm).

2.11. Statistical analysis

The significant differences between treatments (fruit age) were analyzed by one-way analysis of Variance (ANOVA) using the Sisvar software (Ferreira, 2011), followed by polynomial regression analysis for predicting the suitable model by the F-test. The principal component analysis (PCA) was carried out using Minitab 18 software. The statistical significance level was fixed at 5% ($p < 0.05$).

3. Results

3.1. Changes in vitamin C and total phenolic content (TPC)

As depicted in Fig. 2A–C, the effect of developmental stages on vitamin C and TPC was significant ($p < 0.05$). The vitamin C content of red-fleshed dragonfruit significantly decreased in a linear pattern over the developmental stages, varying from 84.07 to 18.92 mg/100 g FM between 28 and 42 days after anthesis (DAA). On the contrary, the TPC, in general, increased from 377.59 mg GAE/100 g FM at 28 DAA to 434.26 GAE/100 g FM at 42 DAA (Folin-Ciocalteu assay) and from 234.85 mg GAE/100 g FM at 28 DAA to 385.31GAE/100 g FM at 42 DAA (Fast Blue BB assay).

3.2. Changes in individual phenolic compounds

As shown in Table 1, developmental stages significantly influenced the individual phenolic compounds ($p < 0.05$). It is important to note that the individual phenolic compounds in this study were evaluated from 30 DAA to 42 DAA. Among eleven phenolic compounds assessed, a total of six, namely, catechin, vanillin, gallic acid, caffeic acid, chlorogenic acid, and ferulic acid, were detected in red-fleshed dragonfruit, among which catechin was the predominant phenolic compound found during the entire development period and ranged from 11.92 ± 0.19 mg/100 g FM at 30 DAA to 8.31 ± 0.12 mg/100 g FM at 42 DAA. The

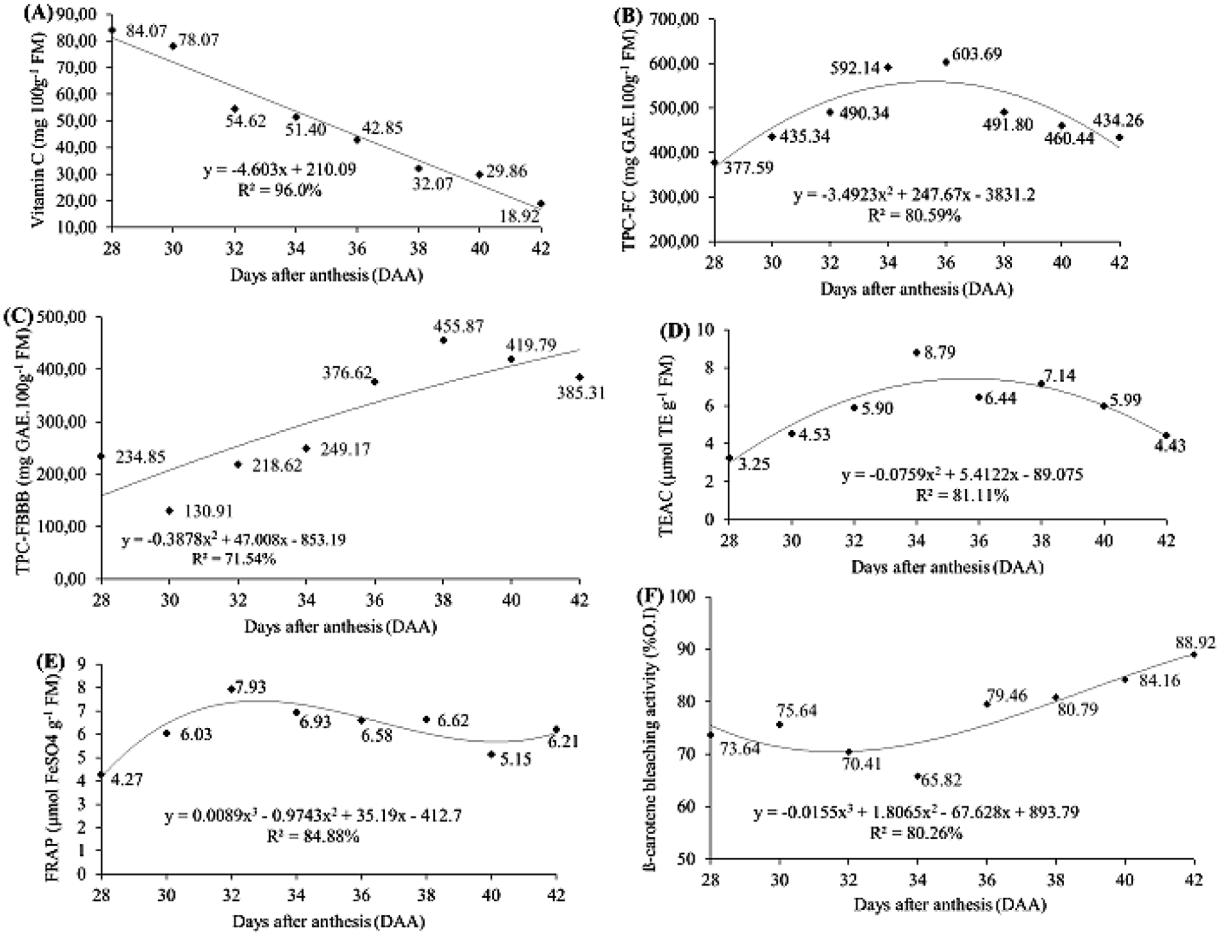

Fig. 2. Changes in (A) vitamin C, (B) TPC-FC, (C) TPC-FBBB, (D) TEAC, (E) FRAP, and (F) β-carotene bleaching activity in red-fleshed dragon fruit during fruit development. Note: TPC-FC: total phenolic compounds measured by Folin-Ciocalteau; TEAC: Trolox equivalent antioxidant capacity; FRAP: ferric-reducing antioxidant power; TPC-FBBB: total phenolic compounds measured by Fast Blue BB.

Table 1
Changes in individual phenolic compounds detected by HPLC in red-fleshed dragon fruit during fruit development.

Phenolic compound	Days after anthesis (DAA) 30	32	34	36	38	40	42
Gallic acid	0.80 ± 0.15	0.19 ± 0.04	0.14 ± 0.06	0.07 ± 0.00	0.07 ± 0.01	0.03 ± 0.03	ND
(+) - Catechin	11.92 ± 0.19	6.92 ± 1.25	3.43 ± 0.74	4.08 ± 2.88	8.49 ± 0.04	11.50 ± 0.06	8.31 ± 0.12
Chlorogenic acid	0.88 ± 0.31	ND	ND	ND	ND	ND	ND
Ferulic acid	0.50 ± 0.03	ND	ND	ND	ND	ND	ND
Caffeic acid	0.56 ± 0.14	0.35 ± 0.00	0.10 ± 0.01	0.09 ± 0.04	0.13 ± 0.01	0.06 ± 0.00	ND
Vanillin	1.12 ± 0.46	1.32 ± 0.00	0.17 ± 0.00	0.19 ± 0.01	0.25 ± 0.09	0.11 ± 0.02	0.11 ± 0.01

Values expressed in mg 100 g^{-1} of fresh mass. Mean value ± standard deviation of three repetitions ($n = 3$); ND: non detected.

catechin levels decreased significantly from 30 DAA to 34 DAA, followed by an increase up to 40 DAA and then decreased to the last evaluation stage. Vanillin was the second most dominant phenolic compound, which ranged from 1.12 ± 0.46 mg/100 g FM at 30 DAA to 0.11 ± 0.01 mg/100 g FM at 42 DAA with an increase until 32 DAA and then slightly fluctuated during the subsequent fruit development stages. The contents of gallic and caffeic acids decreased from 0.80 ± 0.15 and 0.56 ± 0.14 mg/100 g FM at 30 DAA to 0.03 ± 0.03 and 0.06 ± 0.00 mg/100 g FM at 40 DAA, respectively, and were not detected at 42 DAA. On the other hand, chlorogenic and ferulic acids were only detected at 30 DAA, and their contents were 0.88 ± 0.31 and 0.50 ± 0.03 mg/100 g FM, respectively.

3.3. Changes in anthocyanins and betalains

As shown in Fig. 3A–C, anthocyanins and betalains (betacyanins and betaxanthins) significantly increased ($p < 0.05$) during the entire period of development, respectively. The contents of anthocyanins increased from 0.23 mg cy-3-glu/100 g FM at 28 DAA to 16.94 mg cy-3-glu/100 g FM at 42 DAA. On the other hand, the contents of the two groups of betalains (betacyanins and betaxanthins) increased linearly from 7.09 mg/100 g FM at 28 DAA to 250 mg/100 g FM and 4.24 mg/100 g FM at 28 DAA to 59.87 mg/100 g FM at 42 DAA, respectively.

3.4. Changes in antioxidant activity

The antioxidant activity was measured by three methods (TEAC, FRAP, and ß-carotene bleaching). Despite the performed method, the developmental stages had a significant influence ($p < 0.05$) on the antioxidant activity in red-fleshed dragonfruit. The antioxidant activity by TEAC ranged from 3.25 µmol TE/g FM at 28 DAA to 4.43 µmol TE/g FM at 42 DAA with higher levels at 34 DAA, followed by a decrease throughout the fruit development stages (Fig. 2D). The antioxidant activity by FRAP ranged from 4.27 µmol $FeSO_4$/g FM at 28 DAA to 6.21 µmol $FeSO_4$/g FM at 42 DAA with the highest contents at 32 DAA. It then decreased throughout the subsequent fruit development stages (Fig. 2E). Moreover, the antioxidant activity by ß-carotene bleaching ranged from 73.64% at 28 DAA to 88.92% at 42 DAA with the highest values at 42 DAA; there was a fast rise from 28 DAA to 30 DAA, followed by a decrease up to 32 DAA and a new increase in a linear pattern throughout fruit development (Fig. 2F).

3.5. Principal component analysis (PCA)

PCA was performed to summarize the variability of the variables across the fruit development. As shown in Table 2, the first two principal components explained 87.67% of the total variance and gave

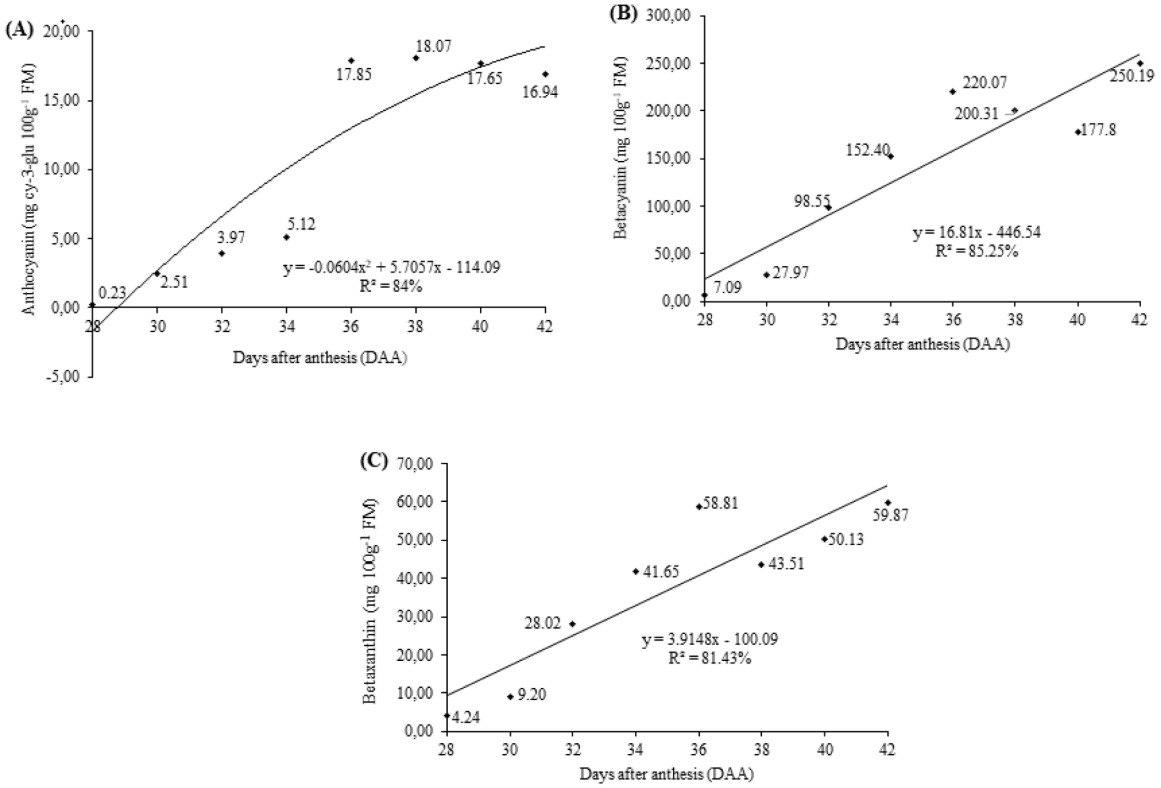

Fig. 3. Changes in (A) anthocyanin (B) Betacyanin, (C) betaxanthin in red-fleshed dragon fruit during fruit development.

Table 2
Contribution of each variable to the PCA factors (PC1 and PC2) - loadings.

Principal component	Eigenvalue	Total variance (%)	Cumulative variance (%)	Variable[a]	Loadings
PC1	5.37	59.67	59.67	Vitamin C	−0.96
				TPC-FBBB	0.89
				Total anthocyanins	0.91
				Betacyanin	0.98
				Betaxanthin	0.97
PC2	2.52	28.0	87.67	TPC-FC	0.81
				TEAC	0.79
				β-carotene bleaching activity	0.98
				FRAP	0.77

[a]TPC-FC: total phenolic compounds measured by Folin-Ciocalteau; TEAC: Trolox equivalent antioxidant capacity; FRAP: ferric-reducing antioxidant power; TPC-FBBB: total phenolic compounds measured by Fast Blue BB.

eigenvalues of 5.37 and 2.52, respectively. The PC1 (59.67%) was mainly contributed by vitamin C, TPC-FBBB, total anthocyanins, betacyanin, and betaxanthin. The PC2 (28%) was positively related to TPC-FC, TEAC, β-carotene bleaching activity, and FRAP. The scores biplot (Fig. 4A) allowed separating the samples according to the development stages into four groups: I (28 and 30 DAA), II (32 DAA), III (34 and 36DAA), and IV (38, 40, and 42 DAA). The two first groups were clearly distinguished from others due to their high contents of vitamin C. The third group was highlighted with high levels of FRAP, betaxanthin, TEAC, and TPC-FC. On the other hand, the fourth group was differentiated from the rest of the groups due to their high contents of betacyanin, anthocyanins, ß-carotene bleaching, and TPC-FBBB.

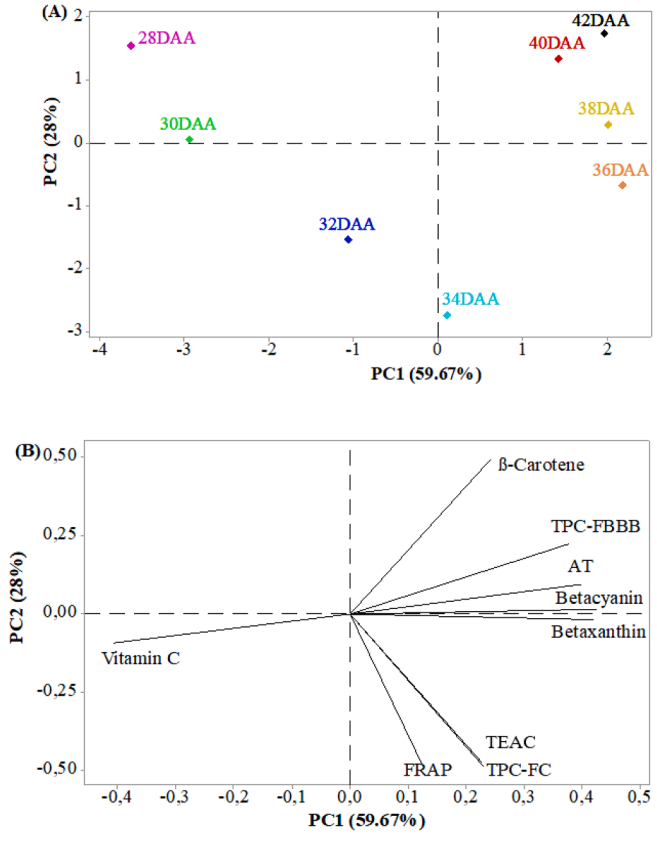

Fig. 4. Principal Component Analysis for the total of nine variables evaluated in red-fleshed dragon fruit during fruit development (A) Scores bi-plot (B) Loadings bi-plot. Note: DAA: Days after anthesis; TPC-FC: total phenolic compounds measured by Folin-Ciocalteau; AT: anthocyanins; TEAC: Trolox equivalent antioxidant capacity; FRAP: ferric-reducing antioxidant power; ß-Carotene: β-carotene bleaching activity; TPC-FBBB: total phenolic compounds measured by Fast Blue BB.

4. Discussion

Red-fleshed dragonfruit is considered a "superfruit" and a good candidate for inclusion in regular human diets due to its extensive range of bioactive compounds (Arivalagan et al., 2021). These compounds are well known for providing a wide range of health-promoting biological activities and functions in preventing several diseases. Bioactive compounds in red-fleshed dragonfruit may vary considerably according to the degree of maturation. For this reason, a comprehensive understanding of this fruit and the influence of that specific factor such as developmental stage has on its chemical composition and, hence, on its bioactive potential, it is useful to guide farmers and professionals who work with red-fleshed dragon fruit about best harvesting time to take advantage of its full potential. This study investigated vitamin C, phenolic compounds, anthocyanins, betalains, and antioxidant activity of red-fleshed dragonfruit at different development stages.

The results showed that vitamin C content decreased during fruit development. Vitamin C is the predominant water-soluble antioxidant found in plant cells and plays a crucial role as a cofactor for many enzymes in both plants and humans (Lemmens et al., 2020). Vitamin C, a potential antioxidant, protects the human body against free radicals and is associated with an inverse relationship with cardiovascular mortality and cancer (Martín-Calvo and Martínez-González, 2017; Wang et al., 2019). Vitamin C content is an important parameter to evaluate the nutritional quality of fruits (Lim et al., 2014). The reduction of vitamin content might be attributed to its oxidative degradation by ascorbic acid oxidase (ascorbinase) enzyme or the action of oxidizing enzymes such as peroxidase (Chitarra and Chitarra, 2005; Siriamornpun and Kaewseejan, 2017). Additionally, the ascorbic acid reduction may also be related to a high-speed synthesis at the initial stage of the fruit development. Afterward, the consumption is greater than synthesis (Nie et al., 2020). Moreover, vitamin C may also be strongly influenced by growing conditions, pedoclimate, crop management, maturation stage, and genotypic/cultivars (Lee and Kader, 2000; Szczepaniak et al., 2019). To our knowledge, no authors have reported the changes in the ascorbic acid content along with the development of *Hylocereus polyrhizus*. Choo and Yong (2011) found levels of vitamin C of 32.65 and 31.05 mg/100 g FM in ripe red and white dragon fruits, respectively, averages close to that found in this study at 38 and 40 days after anthesis (DAA). Other authors have also reported a similar observation of a reduction in vitamin C content in other fruits, such as peach (Guizani et al., 2019; Liu et al., 2015), banana, mango, and papaya (Siriamornpun and Kaewseejan, 2017), acerola cherry (Xu et al., 2020), and carob (*Ceratonia siliqua* L.) (Benchikh et al., 2014).

Phenolic compounds, as secondary metabolites, are the major bioactive compounds widely present in fruits and vegetables, with antioxidant properties and known to provide many health benefits, including prevention of cardiovascular diseases and cancer (Guimarães et al., 2019; Singh et al., 2018). This study found that the TPC measured by Folin-Ciocalteau and Fast Blue BB methods showed a quadratic behavior, increasing and decreasing along the fruit development

process. Despite these variations, when compared between red-fleshed dragonfruit at the early stage of development and later stages, it can be noted that TPC increased over the fruit development. A similar trend was observed during the development of fruit from three dragon fruit cultivars (Hua et al., 2018) and in four fennel fruit populations (England, Spain, Poland, and Iran) (Salami et al., 2017). The increase of the phenolic compounds during the fruit development might be related to the synthesis of new compounds from the phenylpropanoid metabolic pathway under the regulation of phenylalanine ammonia-lyase, a key enzyme of phenolic biosynthesis (Rebey et al., 2019). The synthesis of these compounds changes with the maturation stages of the fruits since the enzyme activity also changes along the stages of fruit development (Elmastaş et al., 2017; Schulz et al., 2015). On the other hand, a slight decrease tendency of TPC may be due to the oxidation of polyphenols by polyphenol oxidase (Nie et al., 2020) and the conversion of soluble phenolics into insoluble phenolics, which are linked to polysaccharides in the cell wall during fruit ripening (Benchikh et al., 2014), Although TPC ranged during the fruit development, in general, our results were higher than the highest values previously reported by Hua et al. (2018) for three pitaya cultivars ("Guanhuahongfen", "Guanhuahong", and "Guanabana") at five maturation stages (282, 184, and 159 mg/100 g, respectively). These variations could be attributed to various factors such as species, cultivar, ripening stage, soil, climate (Alañón et al., 2021; Xu et al., 2020), and extraction conditions (Vongsak et al., 2013). According to Vasco et al. (2008), who classified fruits into three groups according to their phenolic levels: low (<100 mg GAE/100 g), medium (100–500 mg GAE/100 g), and high (>500 mg GAE/100 g), our results suggest that red-fleshed dragonfruit is rich in phenolic compounds, increasing its functional appeal.

This study identified six individual phenolic compounds; catechin and vanillin were the most dominant phenolic compounds during the entire development period. Gallic acid and caffeic acid were detected until 40 DAA, while chlorogenic acid and ferulic acid were detected only at 30 DAA. In line with our results, gallic acid, catechin, ferulic acid, chlorogenic acid, and caffeic acid were also identified in previous studies with ripe dragon fruit (Arivalagan et al., 2021; Esquivel et al., 2007; Li et al., 2019, 2018; Morais et al., 2019). Unlike total phenolic, the six individual phenolic compounds identified decreased along the developmental stages. As mentioned above, our study showed that catechin and vanillin were the predominant phenolic compounds.

In contrast, a recent study performed by Arivalagan et al. (2021), who assessed the biochemical and nutritional characterization of dragon fruit, demonstrated that phenolic acids, such as caffeic, ferulic, and protocatechuic acids were the major compounds in the red-fleshed dragon fruit. According to these authors, the higher antioxidant potential of red-fleshed dragonfruit is closely related to the presence of these compounds. Phenolic acids have been recognized as crucial compounds involved in the prevention of several diseases owing to their ability to scavenge the free radicals generated during the metabolic processes (Forni et al., 2019). On the other hand, catechin has been reported to have many health benefits against diseases caused by oxidative stress, including cancer, diabetes, cardiovascular, and neurodegenerative diseases (Bernatoniene and Kopustinskiene, 2018). Vanillin has potential antimutagenic and antioxidant properties (Sefi et al., 2019).

The anthocyanins pigments are phenolic compounds widely distributed in plant tissue that confer different colors to the fruit, including red, blue, and purple. As an antioxidant, they have been associated with potential human health benefits and play a crucial role during fruit development such as protecting plants against pest and disease attacks, highlight burns, as well as attracting animals to participate in pollination (Kong et al., 2003; Martin et al., 2011; Qu et al., 2021). In the present study, it was found that the total anthocyanins increased during the whole fruit development. Some studies report that anthocyanins are found in small amounts in the early stage of the fruit and increase during maturation (González et al., 2015). The increasing tendency in anthocyanins contents during development was also reported for other fruits such as mulberry (Lee and Hwang, 2017), blackberry (Schulz et al., 2019), strawberry (Ornelas-Paz et al., 2013), jambolan, guabiju, and jaboticaba (Seraglio et al., 2018).

Betalains are plant-derived natural pigments currently having a huge demand for use as natural colorants in the food industry (Gengatharan et al., 2015). Betalains are considered an important quality characteristic of fruits because of their attractive color and provide many health benefits due to their high antioxidant activity against some oxidative stress-related disorders (Sadowska-Bartosz and Bartosz, 2021). According to their chemical structure, they are divided into two main groups: betacyanin which is responsible for the red-violet color, and betaxanthins accountable for the yellow-orange color (Gengatharan et al., 2015). The results showed that the contents of betalains increased with the advancement of maturation. As expected, the levels of betacyanins were higher than that of betaxanthins in all developmental stages, which was also consistent with the findings of García-Cruz et al. (2016), who suggested that the red-purple color of flesh dragon fruit results from betacyanins. Furthermore, it has been reported that betacyanins have higher antioxidant activity than betaxanthins (Azeredo, 2009; Paśko et al., 2021). The increase of betalains contents throughout the development period of red-fleshed dragon fruit was also reported in previous studies (Hua et al., 2018; Wu et al., 2019).

In the present study, it was found that the antioxidant activity measured by the three methods, namely TEAC, FRAP, and ß-carotene bleaching, in general, increased when comparing red-fleshed dragonfruit at the early stage and later stages of development, despite quadratic or cubic behavior observed over development. It was in line with the behavior observed in TPC, anthocyanins, and betalains, suggesting a positive role in the antioxidant activity of the red-fleshed dragon fruit. Increasing evidence shows that the antioxidant activity in red-fleshed dragon fruit is closely related to the phenolic compounds (Luu et al., 2021; Paśko et al., 2021). In addition, Esquivel et al. (2007) demonstrated that betalains are the most important bioactive compounds in *Hylocereus polyrhizus*. On the other hand, it is worth mention that vitamin C, despite being recognized as a potent antioxidant compound, decreased throughout the whole period of fruit development, suggesting that this bioactive compound apparently might not play a pivotal role in the antioxidant activity of the red-fleshed dragon fruit. However, the linear decrease of vitamin C combined with the linear, quadratic and cubic behavior of total phenolic, anthocyanins and betalains contributed to the fluctuations in antioxidant activity between the first and last day of evaluation despite the clear trend of decrease. It is widely recognized that the health-promoting effects of bioactive compounds are closely related to their antioxidant activity; however, the antioxidant activity of fruits is influenced by different mechanisms of their several phytochemicals, and no method can be claimed to provide unequivocal results (Shahidi and Zhong, 2015; Wojdyło et al., 2017).

The PCA agreed with the results previously discussed, which suggest that TPC, betacyanin, betaxanthin, anthocyanins, and antioxidant activity, in general, increased during the fruit development stages, while vitamin C decreased.

Overall, the results of this study demonstrated that the developmental stages did not only influence the quantitative but also the qualitative profile of the individual phenolic compounds present in red-fleshed dragon fruit, which was found to decrease during the entire development period. However, the ripe red-fleshed dragon fruit assembles bioactive compounds with high antioxidant activity and functional appeal. In a previous study, Magalhães et al. (2019) suggested that the red-fleshed dragon fruit from 34 to 42 DAA is suitable for consumption and commercialization. Still, the authors recommend that fruits be harvested 38 DAA or longer to supply near markets when they are fully ripe. Based on the results of our study, to maximize the potential functional benefits of the red-fleshed dragon fruit, the suitable harvesting period seems to be 36 and 38 DAA. Thus, this information can contribute to promote the valorization of red-fleshed dragon fruit in different industrial applications and increase the economic value of this

fruit; nevertheless, further studies about the influence of species, cultivar, growing locations, and soil may be useful for achieving more details about the concentration of bioactive compounds.

5. Conclusion

This study demonstrated that the bioactive compounds and antioxidant activities of red-fleshed dragon fruit were significantly affected by the fruit development stages. In general, the contents of total phenolic compounds, betacyanin, betaxanthin, and anthocyanins increased with the advancement of the fruit development, suggesting that these compounds might have increased antioxidant activity. However, the content of vitamin C and individual phenolic compounds decreased throughout the fruit development stages. Although the contents of bioactive compounds were affected by developmental stages, their levels at ripe stages were high, suggesting that red-fleshed dragon fruit is a promising source of natural antioxidants. Finally, between 36 and 38 DAA can be considered as a suitable time for harvesting.

CRediT authorship contribution statement

Elídio Zaidine Maurício Zitha: Conceptualization, Investigation, Methodology, Data curation, Formal analysis, Writing – original draft. **Deniete Soares Magalhães:** Conceptualization, Investigation, Methodology. **Rafael Carvalho do Lago:** Conceptualization, Investigation, Formal analysis. **Elisângela Elena Nunes Carvalho:** Conceptualization, Resources, Supervision. **Moacir Pasqual:** Conceptualization, Resources, Supervision. **Eduardo Valério de Barros Vilas Boas:** Conceptualization, Funding acquisition, Project administration, Supervision, Validation, Visualization, Writing – original draft.

Declaration of Competing Interest

The authors declare no potential conflicts of interest

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References

Al-Mekhlafi, N.A., Mediani, A., Hadiani, N., Abas, F., Dymerski, T., Lubinska-szczyge, M., Vearasilp, S., Gorinstein, S., 2021. Metabolomic and antioxidant properties of different varieties and origins of dragon fruit. Microchem. J. 160 https://doi.org/10.1016/j.microc.2020.105687.

Alañón, M.E., Pimentel-moral, S., Arráez-román, D., Segura-carretero, A., 2021. HPLC-DAD-Q-ToF-MS pro Fi ling of phenolic compounds from mango (*Mangifera indica* L.) seed kernel of different cultivars and maturation stages as a preliminary approach to determine functional and nutraceutical value. Food Chem. 337, 127764 https://doi.org/10.1016/j.foodchem.2020.127764.

Arivalagan, M., Karunakaran, G., Roy, T.K., Dinsha, M., Sindhu, B.C., Shilpashree, V.M., Satisha, G.C., Shivashankara, K.S., 2021. Biochemical and nutritional characterization of dragon fruit (*Hylocereus* species). Food Chem. 353 https://doi.org/10.1016/j.foodchem.2021.129426.

Azeredo, H.M.C., 2009. Original article betalains : properties, sources, applications, and stability – a review. Int. J. Food Sci. Technol. 44, 2365–2376. https://doi.org/10.1111/j.1365-2621.2007.01668.x.

Benchikh, Y., Louaileche, H., George, B., Merlin, A., 2014. Changes in bioactive phytochemical content and *in vitro* antioxidant activity of carob (*Ceratonia siliqua* L.) as influenced by fruit ripening. Ind. Crop. Prod. 60, 298–303. https://doi.org/10.1016/j.indcrop.2014.05.048.

Bernatoniene, J., Kopustinskiene, D.M., 2018. The role of catechins in cellular responses to oxidative stress. Molecules 23, 1–11. https://doi.org/10.3390/molecules23040965.

Bouaziz, A., Khennouf, S., Zarga, M.A., Abdalla, S., Baghiani, A., Charef, N., 2015. Phytochemical analysis, hypotensive effect, and antioxidant properties of myrtus communis L. growing in Algeria. Asian Pac. J. Trop. Biomed. 5, 19–28. https://doi.org/10.1016/S2221-1691(15)30165-9.

Chitarra, M.I.F., Chitarra, A.B., 2005. Pós-colheita De Frutos e hortaliças: Fisiologia e Manuseio, 2nd Ed. UFLA, Lavras.

Choo, W.S., Yong, W.K., 2011. Antioxidant properties of two species of Hylocereus fruits antioxidant properties of two species of *Hylocereus* fruits. Adv. Appl. Sci. Res. 2, 418–425.

Elmastaş, M., Demir, A., Genç, N., Dölek, Ü., Güneş, M., 2017. Changes in flavonoid and phenolic acid contents in some *Rosa* species during ripening. Food Chem. 235, 154–159. https://doi.org/10.1016/j.foodchem.2017.05.004.

Esquivel, P., Stintzing, F.C., Carle, R., 2007. Phenolic compound profiles and their corresponding antioxidant capacity of purple pitaya (*Hylocereus* sp.) genotypes. Zeitschrift fur Naturforsch.-Sect. C J. Biosci. 62, 636–644. https://doi.org/10.1515/znc-2007-9-1003.

Fathordoobady, F., Jarzębski, M., Pratap-Singh, A., Guo, Y., Abd-Manap, Y., 2021. Encapsulation of betacyanins from the peel of red dragon fruit (*Hylocereus polyrhizus* L.) in alginate microbeads. Food Hydrocoll. 113 https://doi.org/10.1016/j.foodhyd.2020.106535.

Fathordoobady, F., Mirhosseini, H., Selamat, J., Manap, M.Y.A., 2016. Effect of solvent type and ratio on betacyanins and antioxidant activity of extracts from *Hylocereus polyrhizus* flesh and peel by supercritical fluid extraction and solvent extraction. Food Chem. 202, 70–80. https://doi.org/10.1016/j.foodchem.2016.01.121.

Ferreira, D.F., 2011. Sisvar: a computer statistical analysis system. Ciênc. Agrotecnologia 35, 1039–1042. https://doi.org/10.1590/S1413-70542011000600001.

Forni, C., Facchiano, F., Bartoli, M., Pieretti, S., Facchiano, A., D'Arcangelo, D., Norelli, S., Valle, G., Nisini, R., Beninati, S., Tabolacci, C., Jadeja, R.N., 2019. Beneficial role of phytochemicals on oxidative stress and age-related diseases. Biomed. Res. Int. 2019, 16. https://doi.org/10.1155/2019/8748253.

Fuleki, T., Francis, F.J., 1968. Quantitative methods for anthocyanins. 1. Extraction and determination of total anthocyanin in cranberries. J. Food Sci. 33, 72–77. https://doi.org/10.1111/j.1365-2621.1968.tb00887.x.

García-Cruz, L., Valle-Guadarrama, S., Salinas-Moreno, Y., Luna-Morales, C.C., 2016. Postharvest quality, soluble phenols, betalains content, and antioxidant activity of *Stenocereus pruinosus* and *Stenocereus stellatus* fruit. Postharvest Biol. Technol. 111, 69–76. https://doi.org/10.1016/j.postharvbio.2015.07.004.

Gengatharan, A., Dykes, G.A., Choo, W.S., 2015. Betalains : natural plant pigments with potential application in functional foods. LWT Food Sci. Technol. 64, 645–649. https://doi.org/10.1016/j.lwt.2015.06.052.

Gominho, J., Dolores, M., Lourenço, A., Fernández, J., Pereira, H., 2018. *Cynara cardunculus* L. as a biomass and multi-purpose crop: a review of 30 years of research. Biomass Bioenergy 109, 257–275. https://doi.org/10.1016/j.biombioe.2018.01.001.

Gonçalves, G.A.S., Resende, N.S., Carvalho, E.E.N., de Resende, J.V., Vilas Boas, E.V.B., 2019. Effect of processing and frozen storage on the phenolic profile, bioative compounds, antioxidant capacity, and enzymatic activity of mangaba pulp. Curr. Nutr. Food Sci. 15, 48–60. https://doi.org/10.2174/1573401313666171004144858.

González, B., Vogel, H., Razmilic, I., Wolfram, E., 2015. Polyphenol, anthocyanin, and antioxidant content in different parts of maqui fruits (*Aristotelia chilensis*) during ripening and conservation treatments after harvest. Ind. Crops Prod. 76, 158–165. https://doi.org/10.1016/j.indcrop.2015.06.038.

Guimarães, J.T., Keven, E., Ranadheera, C.S., Moraes, J., Raices, R.S.L., Silva, M.C., Ferreira, M.S., Freitas, M.Q., Meireles, M.A.A., Cruz, A.G., 2019. Effect of high-intensity ultrasound on the nutritional profile and volatile compounds of a prebiotic soursop whey beverage. Ultrason. Sonochem. 55, 157–164. https://doi.org/10.1016/j.ultsonch.2019.02.025.

Guizani, M., Maatallah, S., Dabbou, S., Serrano, M., Hajlaoui, H., Helal, A.N., Kilani-Jaziri, S., 2019. Physiological behaviors and fruit quality changes in five peach cultivars during three ripening stages in a semi-arid climate. Acta Physiol. Plant. 41 https://doi.org/10.1007/s11738-019-2950-6.

Ho, P.L., Tran, D.T., Hertog, M.L.A.T.M., Nicolaï, B.M., 2020. Modeling respiration rate of dragon fruit as a function of gas composition and temperature. Sci. Hortic. 263 https://doi.org/10.1016/j.scienta.2019.109138 (Amsterdam).

Hua, Q., Chen, C., Tel, N., Wang, H., Wu, J., Chen, J., Zhang, Z., Zhao, J., Hu, G., Qin, Y., 2018. Metabolomic characterization of pitaya fruit from three red-skinned cultivars with different pulp colors. Plant Physiol. Biochem. 126, 117–125. https://doi.org/10.1016/j.plaphy.2018.02.027.

Jiang, X., Lu, Y., Liu, S.Q., 2020. Effects of pectinase treatment on the physicochemical and oenological properties of red dragon fruit wine fermented with *Torulaspora delbrueckii*. LWT Food Sci. Technol. 132, 109929 https://doi.org/10.1016/j.lwt.2020.109929.

Joshi, M., Prabhakar, B., 2020. Phytoconstituents and pharmaco-therapeutic benefits of pitaya: a wonder fruit. J. Food Biochem. 44, e13260. https://doi.org/10.1111/jfbc.13260.

Kassim, N.K., Rahmani, M., Ismail, A., Aspollah, M., Cheng, G., Ee, L., Nasir, N., Awang, K., 2013. Antioxidant activity-guided separation of coumarins and lignan from *Melicope glabra* (Rutaceae). Food Chem. 139, 87–92. https://doi.org/10.1016/j.foodchem.2013.01.108.

Kaur, G., Thawkar, B., Dubey, S., Jadhav, P., 2018. Pharmacological potentials of betalains. J. Complement Integr. Med. 15 https://doi.org/10.1515/jcim-2017-0063.

Kong, J.M., Chia, L.S., Goh, N.K., Chia, T.F., Brouillard, R., 2003. Analysis and biological activities of anthocyanins. Phytochemistry 64, 923–933. https://doi.org/10.1016/S0031-9422(03)00438-2.

Le Bellec, F., Vaillant, F., Imbert, E., 2006. Pitahaya (Hylocereus spp.): a new fruit crop, a market with a future. Fruits 61, 237–250. https://doi.org/10.1051/fruits:2006021.

Lee, S.K., Kader, A.A., 2000. Preharvest and postharvest factors influencing vitamin C content of horticultural crops. Postharvest Biol. Technol. 20, 207–220. https://doi.org/10.1016/S0925-5214(00)00133-2.

Lee, Y., Hwang, K.T., 2017. Changes in physicochemical properties of mulberry fruits (*Morus alba* L.) during ripening. Sci. Hortic. 217, 189–196. https://doi.org/10.1016/j.scienta.2017.01.042 (Amsterdam).

Lemmens, E., Alós, E., Rymenants, M., Storme, N.De, Keulemans, W.(J), 2020. Dynamics of ascorbic acid content in apple (*Malus* x *domestica*) during fruit development and storage. Plant Physiol. Biochem. 151, 47–59. https://doi.org/10.1016/j.plaphy.2020.03.006.

Leong, H.Y., Show, P.L., Lim, M.H., Ooi, C.W., Ling, T.C., 2018. Natural red pigments from plants and their health benefits : a review. Food Rev. Int. 34, 463–482. https://doi.org/10.1080/87559129.2017.1326935.

Li, X., Li, M., Ji, N., Jin, P., Zhang, J., Zheng, Y., Zhang, X., Li, F., 2019. Cold plasma treatment induces phenolic accumulation and enhances antioxidant activity in fresh-cut pitaya (*Hylocereus undatus*) fruit. LWT Food Sci. Technol. 115 https://doi.org/10.1016/j.lwt.2019.108447.

Li, X., Li, M., Wang, L., Wang, J., Jin, P., Zheng, Y., 2018. Methyl jasmonate primes defense responses against wounding stress and enhances phenolic accumulation in fresh-cut pitaya fruit. Postharvest Biol. Technol. 145, 101–107. https://doi.org/10.1016/j.postharvbio.2018.07.001.

Li, Y., Kong, D., Fu, Y., Sussman, M.R., Wu, H., 2020. The effect of developmental and environmental factors on secondary metabolites in medicinal plants. Plant Physiol. Biochem. 148, 80–89. https://doi.org/10.1016/j.plaphy.2020.01.006.

Lim, Y.J., Oh, C., Park, Y., Kim, D., Kim, U., Cho, Y., Eom, S.H., 2014. Physiological components of kiwifruits with *in vitro* antioxidant and acetylcholinesterase inhibitory activities. Food Sci. Biotechnol. 23, 943–949. https://doi.org/10.1007/s10068-014-0127-z.

Liu, H., Cao, J., Jiang, W., 2015. Changes in phenolics and antioxidant property of peach fruit during ripening and responses to 1-methylcyclopropene. Postharvest Biol. Technol. 108, 111–118. https://doi.org/10.1016/j.postharvbio.2015.06.012.

Luu, T., Le, T., Huynh, N., Quintela-alonso, P., 2021. Dragon fruit : a review of health benefits and nutrients and its sustainable development under climate changes in Vietnam. Czech J. Food Sci. 39, 71–94. https://doi.org/10.17221/139/2020-CJFS.

Madane, P., Das, A.K., Nanda, P.K., Bandyopadhyay, S., Jagtap, P., Shewalkar, A., Maity, B., 2020. Dragon fruit (*Hylocereus undatus*) peel as antioxidant dietary fibre on quality and lipid oxidation of chicken nuggets. J. Food Sci. Technol. 57, 1449–1461. https://doi.org/10.1007/s13197-019-04180-z.

Magalhães, D.S., da Silva, D.M., Ramos, J.D., Salles Pio, L.A., Pasqual, M., Vilas Boas, E. V.B., Galvão, E.C., de Melo, E.T., 2019. Changes in the physical and physico-chemical characteristics of red-pulp dragon fruit during its development. Sci. Hortic. 253, 180–186. https://doi.org/10.1016/j.scienta.2019.04.050 (Amsterdam).

Martín-Calvo, N., Martínez-González, M.Á., 2017. Vitamin C intake is inversely associated with cardiovascular mortality in a cohort of spanish graduates: the SUN project. Nutrients 9, 954. https://doi.org/10.3390/nu9090954.

Martin, C., Butelli, E., Petroni, K., Tonelli, C., 2011. How can research on plants contribute to promoting human health? Plant Cell 23, 1685–1699. https://doi.org/10.1105/tpc.111.083279.

Medina, M.B., 2011. Determination of the total phenolics in juices and superfruits by a novel chemical method. J. Funct. Foods 3, 79–87. https://doi.org/10.1016/j.jff.2011.02.007.

Molla, M.M., Kamal, M.M., Sabuz, A.A., Chowdhury, M.G.F., Khan, M.H.H., Khatun, A., Miaruddin, M., Zashimuddin, M., Islam, M.M., 2021. Chemical composition, bioactive compounds, antioxidants potential and mycotoxin of minor exotic *archidendron pauciflorum* fruit with the focus to Bangladesh. Biocatal. Agric. Biotechnol. 34 https://doi.org/10.1016/j.bcab.2021.102039.

Morais, S.G.G., Borges, G.S.C., Lima, M.S., Martín-Belloso, O., Magnani, M., 2019. Effects of probiotics on the content and bioaccessibility of phenolic compounds in red pitaya pulp. Food Res. Int. 126 https://doi.org/10.1016/j.foodres.2019.108681.

Nie, X.R., Li, H.Y., Wei, S.Y., Han, Q.H., Zhao, L., Zhang, Q., Li, S.Q., Qin, W., Wu, D.T., 2020. Changes of phenolic compounds, antioxidant capacities, and inhibitory effects on digestive enzymes of kiwifruits (*Actinidia chinensis*) during maturation. J. Food Meas. Charact. 14, 1765–1774. https://doi.org/10.1007/s11694-020-00424-1.

Ornelas-Paz, J., de, J., Yahia, E.M., Ramírez-Bustamante, N., Pérez-Martínez, J.D., Escalante-Minakata, P., Ibarra-Junquera, V., Acosta-Muñiz, C., Guerrero-Prieto, V., Ochoa-Reyes, E., 2013. Physical attributes and chemical composition of organic strawberry fruit (*Fragaria* x *ananassa* Duch, Cv. Albion) at six stages of ripening. Food Chem. 138, 372–381. https://doi.org/10.1016/j.foodchem.2012.11.006.

Paśko, P., Galanty, A., Zagrodzki, P., Ku, Y.G., Luksirikul, P., Weisz, M., Gorinstein, S., 2021. Bioactivity and cytotoxicity of different species of pitaya fruits – a comparative study with advanced chemometric analysis. Food Biosci. 40 https://doi.org/10.1016/j.fbio.2021.100888.

Pinto, D., Vieira, E.F., Peixoto, A.F., Freire, C., Freitas, V., Costa, P., Delerue-matos, C., Rodrigues, F., 2021. Optimizing the extraction of phenolic antioxidants from chestnut shells by subcritical water extraction using response surface methodology. Food Chem. 334 https://doi.org/10.1016/j.foodchem.2020.127521.

Pulido, R., Bravo, L., Saura-calixto, F., 2000. Antioxidant activity of dietary polyphenols as determined by a modified ferric reducing /antioxidant power assay. J. Agric. Food Chem. 48, 3396–3402. https://doi.org/10.1021/jf9913458.

Qingzhu, H., Chengjie, C., Zhe, C., Pengkun, C., Yuewen, M., Jingyu, W., Jian, Z., Guibing, H., Jietang, Z., Yonghua, Q., 2016. Transcriptomic analysis reveals key genes related to betalain biosynthesis in pulp coloration of *Hylocereus polyrhizus*. Front. Plant Sci. 6 https://doi.org/10.3389/fpls.2015.01179.

Qu, S., Li, M., Wang, G., Yu, W., Zhu, S., 2021. Transcriptomic, proteomic and LC-MS analyzes reveal anthocyanin biosynthesis during litchi pericarp browning. Sci. Hortic. 289 https://doi.org/10.1016/j.scienta.2021.110443 (Amsterdam).

Ramaiya, S.D., Bujang, S., Zakaria, H., King, S., Arif, M., Sahrir, S., 2013. Sugars, ascorbic acid, total phenolic content and total antioxidant activity in passion fruit (*Passiflora*) cultivars. J. Sci. Food Agric. 93, 1198–1205. https://doi.org/10.1002/jsfa.5876.

Ravichandran, G., Lakshmanan, D.K., Murugesan, S., Elangovan, A., Rajasekaran, N.S., Thilagar, S., 2021. Attenuation of protein glycation by functional polyphenolics of dragon fruit (*Hylocereus polyrhizus*); an *in vitro* and in silico evaluation. Food Res. Int. 140 https://doi.org/10.1016/j.foodres.2020.110081.

Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., Rice-Evans, C., 1999. Antioxidant activity applying an improved ABTS radical cation decolorization assay. Free Radic. Biol. Med. 26, 1231–1237. https://doi.org/10.1016/S0891-5849(98)00315-3.

Rebey, I.B., Wannes, W.A., Kaab, S.B, Bourgou, S., Tounsi, M.S., Ksouri, R., Fauconnier, M.L., 2019. Bioactive compounds and antioxidant activity of *Pimpinella anisum* L. accessions at different ripening stages. Sci. Hortic. 246, 453–461. https://doi.org/10.1016/j.scienta.2018.11.016 (Amsterdam).

I. Sadowska-Bartosz, G. Bartosz, 2021. Biological properties and applications of betalains. Molecules 26, 2520. 10.3390/molecules26092520.

Salami, M., Rahimmalek, M., Ehtemam, M.H., 2017. Comprehensive research on essential oil and phenolic variation in different foeniculum vulgare populations during transition from vegetative to reproductive stage. Chem. Biodivers. 14, e1600246 https://doi.org/10.1002/cbdv.201600246.

Schulz, M., Borges, G.S.C., Gonzaga, L.V., Seraglio, S.K.T., Olivo, I.S., Azevedo, M.S., Nehring, P., de Gois, J.S., de Almeida, T.S., Vitali, L., Spudeit, D.A., Micke, G.A., Borges, D.L.G., Fett, R., 2015. Chemical composition, bioactive compounds and antioxidant capacity of juçara fruit (*Euterpe edulis* Martius) during ripening. Food Res. Int. 77, 125–131. https://doi.org/10.1016/j.foodres.2015.08.006.

Schulz, M., Katia, S., Seraglio, T., Della, F., Nehring, P., Camargo, A., Daguer, H., Valdemiro, L., Carolina, A., Costa, O., Fett, R., 2019. Blackberry (*Rubus ulmifolius* Schott): chemical composition, phenolic compounds and antioxidant capacity in two edible stages. Food Res. Int. 122, 627–634. https://doi.org/10.1016/j.foodres.2019.01.034.

Sefi, M., Elwej, A., Chaâbane, M., Bejaoui, S., Marrekchi, R., Jamoussi, K., Gouiaa, N., Boudawara-Sellemi, T., Cafsi, M.El, Zeghal, N., Soudani, N., 2019. Beneficial role of vanillin, a polyphenolic flavoring agent, on maneb-induced oxidative stress, DNA damage, and liver histological changes in Swiss albino mice. Hum. Exp. Toxicol. 38, 619–631. https://doi.org/10.1177/0960327119831067.

Seraglio, S.K.T., Schulz, M., Nehring, P., Betta, F.D, Valese, A.C., Daguer, H., Gonzaga, L. V., Fett, R., Costa, A.C.O., 2018. Nutritional and bioactive potential of myrtaceae fruits during ripening. Food Chem. 239, 649–656. https://doi.org/10.1016/j.foodchem.2017.06.118.

Shahidi, F., Zhong, Y., 2015. Measurement of antioxidant activity. J. Funct. Foods 18, 757–781. https://doi.org/10.1016/j.jff.2015.01.047.

Singh, B., Pal, J., Kaur, A., Singh, N., 2018. Phenolic compounds as beneficial phytochemicals in pomegranate (*Punica granatum* L.) peel : a review. Food Chem. 261, 75–86. https://doi.org/10.1016/j.foodchem.2018.04.039.

Siriamornpun, S., Kaewseejan, N., 2017. Quality, bioactive compounds and antioxidant capacity of selected climacteric fruits with relation to their maturity. Sci. Hortic. 221, 33–42. https://doi.org/10.1016/j.scienta.2017.04.020 (Amsterdam).

Strohecker, R., Zaragoza, F.M., Henning, H.M., 1967. Analisis De vitaminas : Métodos Comprobadose. Paz Montalvo, Madrid.

Szczepaniak, O.M., Kobus, J., Weronika, C., Monika, K., 2019. Functional properties of cornelian cherry (*Cornus mas* L.): a comprehensive review. Eur. Food Res. Technol. 245, 2071–2087. https://doi.org/10.1007/s00217-019-03313-0.

Thaiudom, S., Oonsivilai, R., Thaiwong, N., 2021. Production of colorant powder from dragon fruit (*Hylocerecus polyrhizus*) peel : bioactivity, heavy metal contamination, antimutagenicity, and antioxidation aspects. J. Food Process. Preserv. 45, e15044. https://doi.org/10.1111/jfpp.15044.

Vasco, C., Ruales, J., Kamal-Eldin, A., 2008. Total phenolic compounds and antioxidant capacities of major fruits from Ecuador. Food Chem. 111, 816–823. https://doi.org/10.1016/j.foodchem.2008.04.054.

Vongsak, B., Sithisarn, P., Mangmool, S., Thongpraditchote, S., Wongkrajang, Y., Gritsanapan, W., 2013. Maximizing total phenolics, total flavonoids contents and antioxidant activity of *Moringa oleifera* leaf extract by the appropriate extraction method. Ind. Crops Prod. 44, 566–571. https://doi.org/10.1016/j.indcrop.2012.09.021.

Wang, J., Wang, J., Ye, J., Kranthi, S., Raghavan, V., 2019. Influence of high-intensity ultrasound on bioactive compounds of strawberry juice: profiles of ascorbic acid, phenolics, antioxidant activity and microstructure. Food Control 96, 128–136. https://doi.org/10.1016/j.foodcont.2018.09.007.

Wojdyło, A., Nowicka, P., Oszmian, J., Golis, T., 2017. Phytochemical compounds and biological effects of actinidia fruits. J. Funct. Foods 30, 194–202. https://doi.org/10.1016/j.jff.2017.01.018.

Y. Wu, J. Xu, Y. He, M. Shi, X. Han, W. Li, X. Zhang, X. Wen, 2019. Metabolic profiling of pitaya (*Hylocereus polyrhizus*) during fruit development and maturation. Molecules 24, 1114. 10.3390/molecules24061114.

Xu, M., Shen, C., Zheng, H., Xu, Y., Xue, C., Zhu, B., Hu, J., 2020. Metabolomic analysis of acerola cherry (*Malpighia emarginata*) fruit during ripening development via UPLC-Q-TOF and contribution to the antioxidant activity. Food Res. Int. 130 https://doi.org/10.1016/j.foodres.2019.108915.

Zayed, A., Serag, A., Farag, M.A., 2020. *Cynara cardunculus* L.: outgoing and otential trends of phytochemical, industrial, nutritive and medicinal merits. J. Funct. Foods 69. https://doi.org/10.1016/j.jff.2020.103937.

Zuo, L.L., Wang, Z.Y., Fan, Z.L., Tian, S.Q., Liu, J.R., 2012. Evaluation of antioxidant and antiproliferative properties of three *actinidia* (*Actinidia kolomikta, Actinidia arguta, Actinidia chinensis*) extracts *in vitro*. Int. J. Mol. Sci. 13, 5506–5518. https://doi.org/10.3390/ijms13055506.