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Research Article

Characterization of 'Wonderful' pomegranate in the state of Chihuahua, México

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Abstract

The consumption of fruits rich in antioxidants has increased in recent years, a clear example of this is the pomegranate (*Punica granatum* L.). The cultivation of the pomegranate shows great expectations due to its profitability and its adaptability to development in arid zones. In this research, 'Wonderful' pomegranate fruits from Coyame Chihuahua, México were used. Quality parameters and bioactive compounds were evaluated to characterize the pomegranate produced in the state of Chihuahua, six different lots from local producers were used. The results showed significance between the different treatments, lot 5 (L5) presented the best quality characteristics such as: weight, diameter, lower percentage of shell and cartilage, as well as one of the highest percentages of arils, in addition, it had a high Total Soluble Solids (TSS) content, a high Titratable Acidity (TA) and a low sugar-acidity ratio, however, lot 3 (L3) showed the highest antioxidant capacity. In general, the qualities and attributes of the Chihuahua pomegranates, obtained higher values in most of the evaluated variables in comparison with other reported results. On the other hand, as a result of this research, the implementation of the color index in the peel is proposed as a tool for the prediction of the maturity index of the pomegranate. This study contributes to the producers of this fruit tree because there is little information on the production and characterization of the pomegranate.

Keywords: antioxidant capacity; color index; *Punica granatum*; total phenols; quality

Introduction

The consumption of fruits rich in antioxidants has increased in recent years, an example of which is pomegranate (*Punica granatum* L.), which is one of the oldest edible fruits that has been widely cultivated in tropical and subtropical countries. There are more than 1,000 cultivars of *P. granatum*, originating in the Middle East and extending throughout the Mediterranean, to eastern China and India, as well as in the southwestern United States, California, and México (Çam *et al*., 2009).

The increase in demand for pomegranate is due to the benefits it brings to human health. Several studies have shown the therapeutic effect of pomegranate peel, arils and flowers, since they have contributed as

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protection against hepatic oxidative stress, kidney damage, and anticancer activity. It is reported that pomegranate peel extract contains punicalagin and acid ellagic cells capable of inhibiting fatty acid synthase and adipogenesis, so they could have a potential effect in the prevention and treatment of obesity (Ambigaipalan *et al*., 2016). Moreover, pomegranate skin, seeds, and juice are reported to contain considerable amounts of phenolic compounds such as flavonoids, ellagitannins, mainly punicalagins, ellagic acid, and punicalins (Akhavan *et al*., 2015; Derakhshan *et al*., 2018), which is related to antioxidant activity. On the other hand, the quality parameters of the fruits play an important role in the concentration of bioactive compounds present in pomegranate (Mirdehghan *et al*., 2006). To determine the quality of the pomegranate it is necessary to take into account the size, the color of the shell as well as the absence of visual defects in it such as sunburn, cracks, cuts and bruises. Other characteristics that must be taken into account in the pomegranate quality are the color of the aril, sugar content and acidity (García-Pastor *et al*., 2020). It is important to mention that the quality of the fruit and the concentration of bioactive compounds will be affected by the cultivar, the growing region, the climate, the maturity, the age of the trees, the storage circumstances, the cultural practice and the irrigation (Çam *et al*., 2009; Tarantino *et al*., 2020).

Data on total area and world production are currently not reported due to the rapid increase in cultivation in recent years. In 2014, an approximate production of 3 million tons (t) was estimated and in 2017 the estimated production was 3.8 million tons (Karapetsi *et al*., 2021). In 2020, it was reported that Mexico has a planted area of 1,262 ha, of which 1,146.25 ha are harvested for a total production of 8,769.36 t. The state of Chihuahua has a planting extension for the cultivation of pomegranate of 45 ha, of which 40 ha are harvested with a production of 615 tons in 2020 (SIAP, 2020).

It is important to mention that fruit growing in arid and semi-arid zones should be oriented towards the use of plant materials that are less demanding of water and more resistant to stress, which, together with deficit irrigation, will allow significant water savings and the profitable production of high-quality fruits (Prieto *et al.,* 2017). Pomegranate cultivation shows great expectations due to its profitability and its adaptability to development in arid zones with few water requirements, being developed and produced in conditions in which other fruit trees would not do so profitably (Moreno, 2010). Therefore, the pomegranate is about to become an option for sustainable agriculture in the face of water scarcity and the effects of climate change throughout the world. Regarding pomegranate production in the state of Chihuahua in the Coyame region, it has shown great adaptability, allowing adequate production.

Based on the importance and demand of pomegranate cultivation, it is of the utmost importance to publicize the quality of pomegranates produced in the north of México, so the objective of this study was to characterize the quality parameters and bioactive compounds of the 'Wonderful' pomegranate produced in the state of Chihuahua.

Materials and Methods

Sample collection and preparation

The variety used in this investigation was 'Wonderful' (*P. granatum* L.). The samples were collected completely randomly on October 11, 2019, in six lots from different producers, in the Coyame area in the state of Chihuahua, the location coordinates are: Latitude: 28.6353, Longitude: 106.089 28° 38′ 7″ North, 106° 5′ 20″ West. Agronomic management is given in a rustic way, with irrigation every 15 to 21 days and pruning, the latter generally to obtain propagation material. To identify the lots, they were named as follows: L1, L2, L3, L4, L5, and L6.

Experimental design

The experimental design was carried out in completely randomized blocks, there were six treatments (lots of producers), eight repetitions (eight trees) at four points within each orchard, north, south, east and west, in which five fruits were collected for each point cardinal. For bioactive compounds, a composite sample was performed per replicate, each with three analytical replicates.

Quality parameters

Fruits were left at room temperature for seven days and subsequently, the following quality parameters were measured:

Fruit weight (W)

Fruits were weighed individually with a digital scale with a capacity of 5 kg and a precision of ± 0.1 g, the results were expressed in grams (g).

Fruit diameter

The fruit size was obtained from the equatorial (ED) and polar diameter (PD), measuring the largest part of the fruit from both poles, making the readings with a graduated vernier in millimeters, with a precision of ± 0.01 mm.

Diameter-length ratio (D/L)

It is a ratio between the equatorial diameter and the polar diameter, where the polar diameter represents the length.

Percentage peel, cartilage, arils

It is a ratio between the total weight of the fruit and the individual weight of each of the parts of the fruit, later converted into a percentage giving a total of 100%.

The density of juice, juice, bagasse, and juiciness percentage

30 grams of composite sample were taken per repetition, and they were processed in a juice extractor (Turmix, TUO5, USA), the juice was measured in a 50 ml graduated cylinder and left to stand until there was phase separation, to proceed to quantification of the amount of juice and bagasse. For the density of juice which was reported in g mL^{-1} and the percentage of juiciness obtained from the extract, the methodology proposed by Oviedo-Mireles *et al.* (2021) was used. For the calculations of juice (g mL^{-1}) and bagasse (g), the following formulas were used:

$$Juice = (density\ of\ juice) * \left(\frac{\%\ of\ juiciness}{100}\right) \quad (1)$$

$$Bagasse = density\ of\ juice - juice \quad (2)$$

Peel and juice color

For the color of the peel, two faces of the fruit were taken to perform the reading and later the average of both was taken, in the case of the juice 40 mL of juice were taken in a beaker and the reading was taken with a Minolta Chromatometer (CR-300, Minolta, Japan). Color parameters were expressed as tristimulus colorimetric measurements, L*, a*, b*, C and H°. Negative L* indicates darkness and positive L* indicates lightness, negative a* indicates the green color, and high positive a* indicates a red color, a high positive b* indicates a more yellow color, and negative b* indicates a blue color. The chroma (C*) value, calculated as $C^* = (a^{*2} + b^{*2})^{1/2}$, indicates the intensity or saturation of the color. Hue angle (H°), a parameter that has shown to be effective in predicting the visual appearance of color, was calculated using the formula $H° = \tan^{-1}(b^*/a^*)$,

where 0° or 360 ° = red-purple, 90° = yellow, 180° = green, and 270° = blue (Solomon *et al*., 2006). The juice color index (CI) was calculated according to Tzulker *et al*. (2007) with the following equation:

$$CI = \frac{180 - H^{\circ}}{L^{*} + C*} \quad (3)$$

Total Soluble Solids (TSS)

To determine the amount of sugar in the fruit expressed as total soluble solids (TSS), a Red Rooster 90681 refractometer scale from 0 to 32 °Brix was used. Approximately 0.5 ml of the juice was taken, which was placed on the surface of the refractometer and the reading was taken (Zhang and Whiting, 2011).

Titratable Acidity (TA)

To determine the titratable acidity, expressed as a percentage of ascorbic acid, 10 mL of juice per sample were taken, six drops of 1% phenolphthalein indicator were added (0.5 g of phenolphthalein plus 70 mL of ethyl alcohol, calibrated to 100 mL with distilled water) and titrated with 0.1 N NaOH (2.15 g of NaOH, 97% purity, calibrated to 500 mL) until a wine color was obtained; the volume used was converted to the equivalent of ascorbic acid in percent using the following formula:

$$Titulable\ acidity = \frac{\left(\frac{0.1 * mL}{10}\right) * 96}{10} \quad (4)$$

Sugar/Acidity ratio (TSS/TA)

It was determined from the total soluble solids for each part of acid content, expressed as a part of sugar for one part of acid (Flores *et al*., 2018). For this process the following formula was used:

$$\text{Ratio}\,\frac{\text{Sugar}}{\text{Acidity}} = \frac{\text{TSS}}{\text{TA}} \quad (5)$$

Bioactive compounds

For the determination of the bioactive compounds, a sample composed of 5 fruits was made, and a total of 2 g of arils were taken and macerated manually in 20 mL of 80% methanol. The samples were left to stand for 24 h and then the supernatant was taken for the determination of total phenolic compounds and total antioxidants.

Total Phenols (TP)

Total phenol content was determined by the Folin-Ciocalteu method (Singleton and Rossi, 1965) using gallic acid (GA) as standard. For the determination, 750 µL of 20% Na_2CO_3, 1375 µL of distilled water, 250 µL of the 50% Folin-Ciocalteu phenolic reagent, and 250 µL of the sample supernatant were taken. The mixture was then incubated at room temperature in the dark for 60 minutes. Absorbance was measured at 725 nm on a DR 5000 Hach spectrophotometer. The results were expressed as mg of gallic acid per 100 g of fresh weight (mg GA 100 g^{-1}). A calibration curve was drawn in triplicate, using reagent grade gallic acid, the value of the equation was Y= 7.4196x- 0.0154, with an r^2 of 0.9967.

Antioxidant capacity (AC)

The DPPH radical (1,1-diphenyl-1,2-picrylhydrazyl) is a stable compound that has an intense violet coloration and absorbs radiation at 517 nm so that its concentration can be determined by spectrophotometric methods (Kim *et al*., 2002). The reaction was carried out by mixing 2950 µL of DPPH radical solution with 0.50 µL of the sample extract. The mixture was kept at room temperature and protected from light for 30 minutes. Subsequently, the absorbance at 517 nm was measured using a UV/Vis spectrophotometer. The results were expressed as mg of ascorbic acid per 100 g of fresh weight (mg AA 100 g^{-1}). The blank used was

80% methanol and a calibration curve was made. Using a high purity reagent grade ascorbic acid standard, and the calibration was in triplicate, the value of the equation was Y= -24.261x + 0.9212, with an r^2 of 0.9951.

Statistical analysis

Statistical analysis was performed in completely randomized blocks. The data obtained were subjected to an analysis of variance for the proposed experimental design and the separation of means with the least significant difference ($p \leq 0.0.5$). For this process, the statistical package SAS (SAS Institute Inc., SAS/STAT Software: Usage and Reference, Version 6, First Edition, Cary, NC: SAS Institute Inc., 1989) was used.

Results and Discussion

The results obtained for the physical characteristics of the fruit are shown in Table 1, presenting significant differences in the different parameters evaluated.

Table 1. Physical characteristics of 'Wonderful' pomegranate

Treatment	W	ED	PD	D/L	% Peel	% Cartilage	% Arils
	<.0001[W]	0.0003	<.0001	0.4660	<.0001	<.0001	0.0211
L1	345.3 a[x]	88.2 a	74.9 ab	1.175 a	43.7 abc	3.5 b	52.8 abc
L2	312.8 a	83.2 ab	72.6 abc	1.146 a	45.4 ab	2.0 d	55.6 bc
L3	231.4 b	81.3 b	71.4 bc	1.150 a	43.1 bc	3.5 b	53.5 abc
L4	262.4 b	76.5 c	65.6 d	1.381 a	47.0 a	2.6 cd	50.4 c
L5	308.2 a	87.0 a	76.1 a	1.144 a	41.3 c	3.0 bc	55.7 ab
L6	261.3 b	80.0 bc	69.8 c	1.150 a	37.1 d	6.7 a	56.2 a
MSD[Y]	42.5	5.0	4.0	0.278	3.4	0.7	3.5
μ	286.9	82.8	71.7	1.191	42.93	3.54	53.53
CV%	14.58	5.92	5.45	22.96	7.83	20.88	6.48
R^2	0.6574	0.6069	0.6389	0.2264	0.5615	0.8581	0.3342

ED, equatorial diameter mm; PD, pole diameter mm; D/L ratio equatorial diameter over polar diameter; % Peel, peel percentage; % cartilage, percentage of cartilage; % Arils, percentage of arils. probability analysis of variance= w, Pr > 0.05 not significant, 0.05 ≤ Pr ≤ 0.01, significant, Pr < 0.01 highly significant; [X]different letters are statistically different (Pr < 0.05), [Y]Minimum Significant Difference. μ general media, CV% coefficient of variation, R^2 coefficient of determination.

For the weight variable, the L1 pomegranates showed the heaviest fruit. Wetzstein *et al.* (2006) reported an average weight of 345 g for the 'Wonderful' variety in California, which coincides with the registered weights of the L1 treatment. Sarkhosh *et al.* (2009) mention an average of 271.08 g for Iranian varieties, which is close to our results. In the case of the equatorial diameter (ED), L1 recorded the largest value, with 88.2 mm, and the smallest value was presented by L4 with 76.5 mm. For the polar diameter (PD) L5 had the largest value with 76.1 mm and the smallest value for L4 with 65.6 mm. Fawole and Opara (2013) report an equatorial diameter of 84.42 mm and a polar diameter of 74.81 mm in pomegranates of the 'Ruby' variety, meanwhile, Tehranifar *et al.* (2010) reported values of 64.98-86.88 mm for the equatorial diameter and 69.49-81.56 mm of polar diameter. If results are compared with the ones reported, L1 showed higher ED, while L5 higher PD. Al-Maiman and Ahmad (2002) show in their experiment how the weight and both diameters increase at the same time from an immature fruit to one at full maturity, this can be attributed to the fact that the size of the fruit is related to the number and size of the cells present in the mesocarp. In addition, in pomegranate the size of the fruit is influenced by the quality of the flower and the pollination process, since each aril is the result of a fertilized ovule (Wetzstein *et al*., 2011). In the case of the diameter/length ratio (D/L) it indicates the shape of the fruits, the closer this value is to 1.0, the rounder its shape will be, in this case, the fruits evaluated did not

have significant differences between the treatments and showed oval tendencies. These values are similar to those reported by Tehranifar *et al*. (2010) where twenty varieties of Iranian pomegranates were compared. For the percentage of arils, the L6 pomegranates had the highest percentage of arils with 56.2%, and, therefore, a lower percentage of shell of 37.1%, despite being one of the smallest fruits. The pomegranates with the highest percentage of shell were for L4 with 26.68% compared to L6. On the other hand, L5 pomegranates were the second treatment with the lowest peel content and the highest percentage of arils, however, they had lower cartilage content compared to L6. The percentage of peel and arils in the Iranian pomegranate experiment ranged from 32.28-59.82% and 37.59-65%, respectively (Tehranifar *et al*., 2010). In this case, the percentage of shell in the results shown were lower than those reported, while the percentage of arils remained within the range of Iranian pomegranates. In turn, a study in pomegranates from Jalisco, México showed an average of 46.42% of the inedible part for the pomegranate fruit and 51.73% for the edible part (Castañeda-Saucedo *et al*., 2012), while for the pomegranates from Coyame obtained an average of 46.47% of the inedible part and 53.53% for the arils. However, Fawole and Opara, (2013a) mention that the content of arils represents approximately 52% of the total weight of the fruit, so only L4 fruits are below the estimated average.

Table 2. Quality parameters of the pomegranate juice of the 'Wonderful'

Treatment	Density of juice	% Juiciness	Juice	Bagasse	TSS	TA	TSS/TA
	0.0784[w]	0.0020	0.0554	<.0879	<.0001	<.0001	0.0001
L1	1.8 b[x]	78.1 bc	1.43 c	0.41 ab	18.2 ab	0.203 c	102.0 a
L2	2.1 ab	74.2 c	1.58 bc	0.54 a	18.6 a	0.200 c	103.6 a
L3	1.9 b	82.7 ab	1.60 bc	0.34 b	17.4 cd	0.253 b	70.3 bc
L4	2.3 a	85.7 a	1.99 a	0.36 b	16.9 d	0.324 a	53.5 c
L5	2.0 b	76.1c	1.52 c	0.48 ab	18.2 ab	0.295 a	62.1 c
L6	2.1 ab	84.8 a	1.80 ab	0.33 b	18.0 bc	0.206 c	86.9 ab
DMS[y]	0.3	6.3	0.26	0.17	0.6	0.041	22.5
μ	2.06	80.26	1.65	0.41	17.88	0.2465	79.73
CV%	16.40	7.76	15.34	40.13	3.15	16.40	27.80
R^2	0.3016	0.4527	0.4694	0.3152	0.6368	0.7131	0.5961

Juice density (g mL^{-1}), juice (g mL^{-1}), bagasse (g), juiciness percentage (%), TSS (°Brix), TA (% ascorbic acid), TSS/TA (ratio TSS/TA). probability analysis of variance= w, Pr > 0.05 not significant, 0.05 ≤ Pr ≤ 0.01, significant, Pr < 0.01 highly significant; [x]different letters are statistically different (Pr < 0.05), [y]Minimum Significant Difference. μ general media, CV% coefficient of variation, R^2 coefficient of determination.

Table 2 shows the qualities of pomegranate juice. For the first parameter, juice density, the fruits of L4 had the densest juice with 44.44% more than L1, which had the lowest density, this refers to the grams of fruit used to obtain a milliliter of juice, therefore, L1 used 1.8 g to obtain 1 ml of juice, of which 1.43 were liquid and 0.41 bagasse. In a study carried out on pomegranates of the 'Taifi', it is mentioned that of the total edible portion of the fruit, 63.58% represents the juice content, while 36.21% is seed waste (Maiman and Ahmad, 2002). Castaneda-Saucedo *et al*. (2012) report in their study that the percentage of the seed varies from 9.38-23%, so the density of the juice may depend on the size of the seeds of each fruit and therefore the percentage of juiciness. In this case, it could be speculated that the arils and seeds of treatment L4 were smaller than the rest of the treatments, since it required a higher content of grams for a milliliter of juice, in addition, it had the highest percentage of juice with 85.7% and with one of the smallest amounts of bagasse. The percentage of juice for the cultivar 'Wonderful' grown in Condobolin, Australia had an average yield of 37% of the total weight of the fruit (Fawole and Opara, 2013), while the cultivation of this variety in Israel had a range of 18-40%, which justifies it due to the differences in climatic conditions that explain the great variation (Fawole and Opara, 2013a). Likewise, in a study of 14 pomegranate genotypes in Jalisco, México, a maximum mean of 39.7% was reported as a proportion of juice (Tapia-Campos *et al*., 2016), even so, L2 fruits being the pomegranates with

the lowest juice content are well above the data mentioned before. Regarding TSS, TA and sugar-acidity ratio (TSS/TA), the fruits of L2 were the sweetest fruits, and therefore with the lowest acidity content. Poyrazoglu *et al*. (2002) mention a range of soluble solids from 16 to 19 °Brix for pomegranate crops in Turkey, while Castañeda-Saucedo *et al*. (2012) and Tapia-Campos *et al.* (2016), report a maximum of 17 °Brix for pomegranates produced in Jalisco, in turn an increase from 10.30 °Brix in immature fruits to 19.56 °Brix in fully ripe fruit is reported (Zarei *et al*., 2011). Zaouay *et al*., (2012) mention a general minimum threshold for TSS of 12% required for commercial use of pomegranate, so all samples were above the threshold. In the case of California, TA of the 'Wonderful' harvest starts at 1.9% (Karapetsi *et al*., 2021), this agrees with what was reported by Fawole and Opara (2013) who consider a TA value of 1.8% as the standard of more satisfactory maturity. This parameter tends to decrease as the days after flowering increase, while the TSS increase, which denotes that these parameters are related to the physiological development of the fruit (Khodabakhshian *et al*., 2017). In this study, AT ranged between 0.200-0.324% Similar results to those obtained by Zaouay *et al*., (2012) for sweet cultivars ranging from 0.1-0.4%, in this case, Mayuoni-Kirshinbaum and Porat (2014), emphasize the importance of acidity levels in the juice, since acidity influences the perception of pomegranate flavor, especially since it affects the stage of maturity and the time of harvest of the fruit pomegranates harvested early are more acidic and astringent than those harvested at optimum ripeness. Al-Maiman and Ahmad (2002) mention that ascorbic acid content decreases significantly with advancing maturity and total and individual sugars reach maximum levels, which can be attributed to starch hydrolysis (Kulkarni and Aradhya, 2005), so it could be elucidated that the samples were in advanced maturity. In the case of the TSS/TA ratio, it provides the maturity index, and is commonly used to define the "flavor" of the pomegranate fruit during development (Fawole and Opara, 2013a), the values obtained from the TSS/TA ratio in this study were 53.5-103.6, in Tunisia similar values are reported with a maximum of 104.9 in different pomegranate genotypes (Zaouay *et al*., 2012). Melgarejo and Salazar (2006) share with us a maturity index classification for Spanish cultivars where sour varieties have values of 5–7, 17–24 for bittersweet and 31–98 for sweet cultivars. Holland *et al*. (2009) reported that the 'Wonderful' is bittersweet, when comparing the results with these indices, it is observed that the samples had a similar maturity index to the sweet varieties. According to Boroychov-Neori *et al*. (2009) the TSS/TA ratio increases as the pomegranate matures, so in this case, it can be seen that the fruits were in an advanced state of maturity.

For the color parameters of the peel and juice, Tables 3 and 4 are shown, respectively. In the case of L*, this parameter tends to decrease during fruit ripening, indicating that the skin or fruit juice tends to darken during this process (Shwartz *et al*., 2009). The increase in the green-red coordinate, a*, is related to the increase in biosynthesis and the accumulation of anthocyanin pigments, responsible for the intense red color of ripe pomegranate fruits. Studies on the color of the 'Wonderful' variety have shown that the pigmentation increases during the ripening process (Shwartz *et al*., 2009), which continues to increase in intensity even when the TSS content reaches the maximum (Fawole and Opara, 2013a), this characteristic garnet color of the pomegranate is presents with high values of a* and C* and low values of b* and H° (Legua *et al*., 2016). In the case of the CI, it has been implemented to provide an objective criterion of the maturity index through color (Shwartz *et al*., 2009), its increase or decrease during the progression of maturity may be due to the increase in the content of anthocyanins and the decrease in phenols, since it is reported that phenols are probably consumed in the biosynthesis of the flavylium ring during the formation of the anthocyanin pigment, which causes a reduction in its content. In addition, an additional increase in TSS and a slight decrease in the content of anthocyanin pigments are reported due to the progress in the maturation of the pomegranate. This onset of anthocyanin discoloration is associated with decreased acidity, which may be the cause of internal aril decomposition in overripe pomegranate fruits (Kulkarni and Aradhya, 2005). In short, as pomegranate fruits ripen, the color coordinates evolve (Manera *et al*., 2013).

Table 3. Color of 'Wonderful' pomegranate peel

Treatment	Color			C*	H°	CI
	L*	a*	b*			
	0.0003[w]	0.0571	0.0151	0.0377	0.0159	0.0592
L1	36.8 c[x]	34.0 ab	9.5 c	35.4 b	15.3 c	4.6 a
L2	39.6 b	33.1 b	9.9 c	34.6 b	16.5 bc	4.7 a
L3	39.6 b	32.8 b	11.8 ab	34.9 b	19.9 a	4.5 ab
L4	43.3 a	34.8 ab	12.3 ab	36.6 ab	19.5 ab	4.2 ab
L5	41.0 ab	36.4 a	13.4 a	38.8 a	20.1 a	4.0 b
L6	39.4 b	32.4 b	10.8 bc	34.2 b	18.2 abc	4.6 a
DMS[Y]	2.46	2.77	2.36	3.07	3.14	0.48
μ	39.98	33.94	11.29	35.83	18.24	4.43
CV%	6.07	8.05	20.56	8.44	16.97	10.65
R^2	0.5098	0.3143	0.4044	0.3420	0.4103	0.3425

L*(lightness/darkness), a*(green/red), b* (yellow/blue), C* (Chroma), H° (Hue angle) and CI (Color index). [probability] analysis of variance= w, Pr > 0.05 not significant, 0.05 ≤ Pr ≤ 0.01, significant, Pr < 0.01 highly significant; [X]different letters are statistically different (Pr < 0.05), [Y]Minimum Significant Difference. μ general media, CV% coefficient of variation, R^2 coefficient of determination.

For L*, a mean of 39.98 was obtained, in this case, the fruits of the L4 treatment showed the highest reading, which was 43.3. This could indicate, in addition to the TSS/TA ratio, that these fruits were the most immature. For variables a* and C*, pomegranates from lot L5 showed the reddest fruits with 36.4 and 38.8 respectively. Shwartz *et al*. (2009), report values of a* of 34.4-39.4 and for C* of 45.1-46.5 in 'Wonderful' fruits that were harvested on dates similar to those of this work, while Opara *et al*. (2009) mention ranges of 21.48-39.16 for a* and 41.13-50.32 for C* in different pomegranate varieties. For the b* parameters it can be observed that the peel pigmentation tended to have a slight yellow pigmentation, which could be due to the heterogeneous color of the peel, while the H° values indicate that the peel color ranged from red to purple. Finally, the CI has a mean of 4.43 while Shwartz *et al*. (2009), report a maximum value of 2.3 in its most advanced stage of maturity, the differences in the color of the peel can be attributed to the area and climate in which the pomegranates were developed, as mentioned by Tarantino *et al*. (2020). However, based on the data obtained in this investigation, it is recommended to harvest when the CI is 4.0 in the peel, as in the case of lot L5, since it has better quality characteristics such as: weight, diameter, lower percentage of peel and cartilage, as well as one of the highest percentages of arils, in addition, it had a high content of TSS, a high TA and a low sugar-acidity ratio, which is an important index of maturity. However, if what is sought is a higher antioxidant capacity it would be better to harvest the pomegranates when the CI is 4.5 as shown by lot L3.

Table 4. Color of 'Wonderful' pomegranate juice

Treatment	Color			C*	H°	CI
	L*	a*	b*			
	0.1870[w]	0.0036	0.0002	0.0037	0.0009	0.0020
L1	28.9 ab	3.3 ab	-0.77 c	3.4 ab	-13.7 bc	45.2 bc
L2	29.2a	3.8 a	0.65 b	3.9 a	-9.8 a	39.1 c
L3	28.8 ab	3.1 bc	-0.74 bc	3.2 bc	-14.5 bc	47.6 ab
L4	28.8 ab	2.5 c	-0.75 c	2.7 c	-17.1 c	54.6 a
L5	28.6 b	3.5 ab	-0.54 a	3.5 ab	-8.7 a	41.6 bc
L6	29.1 a	3.6 ab	-0.72 bc	3.6 ab	-12.3 ab	42.8 bc
DMS[Y]	0.4	0.6	0.10	0.6	3.9	7.2
μ	28.89	3.30	-0.6956	3.38	-12.67	45.17
CV%	1.50	18.33	-14.23	17.03	-29.97	15.62
R^2	0.2631	0.4179	0.5393	0.4167	0.4721	0.4349

L*(lightness/darkness), a*(green/red), b* (yellow/blue), C* (Chroma), H° (Hue angle) and CI (Color index). [probability] analysis of variance= w, Pr > 0.05 not significant, 0.05 ≤ Pr ≤ 0.01, significant, Pr < 0.01 highly significant; [X]different letters are statistically different (Pr < 0.05), [Y]Minimum Significant Difference. μ general media, CV% coefficient of variation, R^2 coefficient of determination.

For the color of the juice, significant differences were observed in the color coordinates between treatments (Table 4). For L* there was a mean of 28.89, while Schwartz *et al*. (2009) reports a maximum value of 27.6 for pomegranate juice, L2 presented the reddest juice according to the parameters of a* with 3.8 and C* with 3.9, values that were similar to those reported by Shwartz *et al*. (2009) in completely ripe fruits a* 3.5 and C* 3.7, while the variable b* mostly had negative results, which indicates that the juices tended more to a blue than yellow pigmentation. These results together with the signs negatives of the hue angle (H°), suggested that the juices were dark, and could be purple. The CI showed values well above those reported by Shwartz *et al.* (2009), therefore, it is mentioned that the color of the juice is widely related to the conditions in which the crop develops (Shulman *et al*., 1984).

Table 5. Bioactive compounds of the pomegranate 'Wonderful'

Treatment	Total phenols	Antioxidant capacity
W	0.0122	<.0001
L1	196.1 b	57.8 a
L2	208.6 b	58.2 a
L3	246.3 a	60.0 a
L4	191.7 a	48.2 b
L5	209.9 b	39.5 c
L6	206.3 b	45.2 bc
DMS[Y]	29.8	6.6
μ	209.8	51.5
CV%	13.99	12.72
R^2	0.4535	0.6871

Total phenols (mg GA 100 g^{-1}), antioxidant capacity (mg AA 100 g^{-1}). probability analysis of variance= w, Pr > 0.05 not significant, 0.05 ≤ Pr ≤ 0.01, significant, Pr < 0.01 highly significant; [X]different letters are statistically different (Pr < 0.05), [Y]Minimum Significant Difference. μ general media, CV% coefficient of variation, R^2 coefficient of determination.

On the other hand, the fruits from L3 had the highest content of total phenols (TP) with 28.48% more than the fruits from L4. Reza *et al*. (2011) mention a range of 11.62-21.03 mg gallic acid equivalents per gram

of extract (mg GAE g^{-1}), while Li *et al*. (2006) reported 24.4 mg equivalents of tannic acid per gram. The results obtained, including those of L4, are higher than those reported and these differences can be attributed to the different extraction methods, the varieties as well as the area where they were grown. However, Fredes *et al*. (2014) report a range of 3.8-4.0 g GAE kg^{-1} in the 'Wonderful' variety. These data are closer to the data obtained for Coyame pomegranates. In turn, it has been reported that in pomegranates, as well as in many other crops, the level of antioxidant activity can be attributed to the total phenolic level (Gil *et al*., 2000), where hydrolysable tannins represent 92% of its antioxidant activity. The group of hydrolysable tannins contains punicalagin isomers, which were suggested to be responsible for about half of the total antioxidant capacity of pomegranate juice, in addition to ellagic acid, gallic acid, and punicalin (Tzulker *et al*., 2007).

Regarding the antioxidant capacity (AC), lots L1, L2 and L3 showed the highest levels with 57.8, 58.2 and 60 mg AA 100 g^{-1} respectively, while the rest of the treatments had values below of the mean obtained, which was 51.5 mg AA 100 g^{-1}. Mirdehghan *et al*. (2006) report values of 73.54 mg equivalents of ascorbic acid per 100 g in the variety Mollar de Elche, in varieties produced in California a maximum value of 5.79 mmol equivalents of Trolox per g is reported (Ambigaipalan *et al*., 2016). These differences between the reported values and those obtained in this research can be attributed, as in the phenol content, to the extraction conditions, analytical methodologies, and cultivation areas as mentioned by Raffo *et al*. (2006). Although the antioxidant capacity depends mainly on the content of phenols, the attractiveness of the fruit to consumers is mainly related to physical parameters and sensory quality, in addition to the benefits to human health that its consumption provides, which vary depending on the cultivation and climatic conditions during pomegranate ripening (Borochov-Neori *et al*., 2009).

Conclusions

The quality parameters and bioactive compounds had significant differences between lots, in addition, the results obtained show that Chihuahuan pomegranates have qualities to compete in the national and international market, since most of the variables evaluated presented, better results compared to published studies in different parts of the world, with the advantage that agronomic management in the Coyame area is minimal. On the other hand, it is important to mention the implementation of the color index in the peel as a tool for the prediction of the maturity index of the pomegranate in a non-destructive way, being a support for the producers at the time of harvest. Despite the importance that this crop is acquiring in Mexico, there are very few studies on its production and characterization in the country, so it is recommended to continue with works that provide support to the producers of this crop.

Authors' Contributions

Conceptualization: JMSP; Methodology: NGTB, LCNM; Validation: JMSP, NGTB; Formal analysis: JMSP; Investigation: NGTB, LCNM; Data curation: JMSP; Funding acquisition: JMSP, ES; Project administration: JMSP; Writing: NGTB, LCNM; Review and editing: NGTB, JMSP, ES, LCNM; All authors read and approved the final manuscript.

Ethical approval (for researches involving animals or humans)

Not applicable.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

References

Akhavan H, Barzegar M, Weidlich H, Zimmermann BF (2015). Phenolic compounds and antioxidant activity of juices from ten Iranian pomegranate cultivars depend on extraction. Journal of Chemistry 11-14. *https://doi.org/10.1155/2015/907101*

Al-Maiman SA, Ahmad D (2002). Changes in physical and chemical properties during pomegranate (*Punica granatum* L.) fruit maturation. Food Chemistry 76(4):437-441. *https://doi.org/10.1016/S0308-8146(01)00301-6*

Ambigaipalan P, De Camargo AC, Shahidi F (2016). Phenolic compounds of pomegranate by products (outer skin, mesocarp, divider membrane) and their antioxidant activities. Journal of Agricultural and Food Chemistry 64(34):6584-6604. *https://doi.org/10.1021/acs.jafc.6b02950*

Borochov-Neori H, Judeinstein S, Tripler E, Harari M, Greenberg A, Shomer I, Holland D (2009). Seasonal and cultivar variations in antioxidant and sensory quality of pomegranate (*Punica granatum* L.) fruit. Journal of Food Composition and Analysis 22(3):189-195. *https://doi.org/10.1016/j.jfca.2008.10.011*

Çam M, Hişil Y, Durmaz G (2009). Classification of eight pomegranate juices based on antioxidant capacity measured by four methods. Food Chemistry 112(3):721-726. *https://doi.org/10.1016/j.foodchem.2008.06.009*

Derakhshan Z, Ferrante M, Tadi M, Ansari F, Heydari A, Hosseini MS, Conti GO, Sadrabad EK (2018). Antioxidant activity and total phenolic content of ethanolic extract of pomegranate peels, juice and seeds. Food and Chemical Toxicology 114:108-111. *https://doi.org/10.1016/j.fct.2018.02.023*

Fadavi A, Barzegar M, Azizi MH, Bayat M (2005). Note. Physicochemical composition of ten pomegranate cultivars (*Punica granatum* L.) grown in Iran. Food Science and Technology International 11(2):113-119. *https://doi.org/10.1177/1082013205052765*

Fawole Olaniyi A, Opara UL (2013). Changes in physical properties, chemical and elemental composition and antioxidant capacity of pomegranate (cv. Ruby) fruit at five maturity stages. Scientia Horticulturae 150:37-46. *https://doi.org/10.1016/j.scienta.2012.10.026*

Fawole Olaniyi A, Opara UL (2013a). Developmental changes in maturity indices of pomegranate fruit: A descriptive review. Scientia Horticulturae 159:152-161. *https://doi.org/10.1016/j.scienta.2013.05.016*

Fawole Olaniyi A, Opara UL (2013b). Harvest discrimination of pomegranate fruit: Postharvest quality changes and relationships between instrumental and sensory attributes during shelf life. Journal of Food Science 78(8). *https://doi.org/10.1111/1750-3841.12176*

Fawole Olaniyi A, Opara UL, Theron KI (2012). Chemical and phytochemical properties and antioxidant activities of three pomegranate cultivars grown in South Africa. Food and Bioprocess Technology 5(7):2934-2940. *https://doi.org/10.1007/s11947-011-0533-7*

Flores CMA, Soto PJM, Salas SNA, Chávez SE, Piña RFJ (2018). Effect of $CaCO_3$ industrial byproduct on quality atributes, phenolic content and antioxidant capacity of apple Cvs Golden Delicious and Top Red. Nova Scientia 10(1):64-82.

Fredes C, Montenegro G, Zoffoli JP, Santander F, Robert P (2014). Comparison of the total phenolic content, total anthocyanin content and antioxidant activity of polyphenol-rich fruits grown in Chile. Ciencia e Investigacion Agraria 41(1):49-59. *https://doi.org/10.4067/S0718-16202014000100005*

García-Pastor ME, Zapata PJ, Castillo S, Martínez-Romero D, Guillén F, Valero D, Serran M (2020). The effects of salicylic acid and its derivatives on increasing pomegranate fruit quality and bioactive compounds at harvest and during storage. Frontiers in Plant Science 11(July): 1-14. *https://doi.org/10.3389/fpls.2020.00668*

Gil MI, Tomás-Barberán FA, Hess-Pierce B, Holcroft DM, Kader AA (2000). Antioxidant activity of pomegranate juice and its relationship with phenolic composition and processing. Journal of Agricultural and Food Chemistry 48(10):4581-4589.

Hegazi NM, El-Shamy S, Fahmy H, Farag MA (2021). Pomegranate juice as a super-food: A comprehensive review of its extraction, analysis, and quality assessment approaches. Journal of Food Composition and Analysis 97:103773. *https://doi.org/10.1016/j.jfca.2020.103773*

Holland D, Hatib K, Bar-Ya'akov I. (2009). Pomegranate: botany, horticulture, breeding. Horticultural Reviews 35:127-191.

Kandylis P, Kokkinomagoulos E (2020). Food applications and potential health benefits of pomegranate and its derivatives. Foods 9(2):122. *https://doi.org/10.3390/foods9020122*

Karapetsi L, Pantelidis G, Pratsinakis ED, Drogoudi P, Madesis P (2021). Fruit quality traits and genotypic characterization in a pomegranate *ex situ* (*Punica granatum* L.) collection in Greece. Agriculture 11(6):1-22. *https://doi.org/10.3390/agriculture11060482*

Karimi M, Sadeghi R, Kokini J (2017). Pomegranate as a promising opportunity in medicine and nanotechnology. Trends in Food Science and Technology 69:59-73. *https://doi.org/10.1016/j.tifs.2017.08.019*

Khodabakhshian R, Emadi B, Khojastehpour M, Golzarian MR, Sazgarnia A (2017). Non-destructive evaluation of maturity and quality parameters of pomegranate fruit by visible/near infrared spectroscopy. International Journal of Food Properties 20(1):41-52. *https://doi.org/10.1080/10942912.2015.1126725*

Kulkarni AP, Aradhya SM (2005). Chemical changes and antioxidant activity in pomegranate arils during fruit development. Food Chemistry 93(2):319-324. *https://doi.org/10.1016/j.foodchem.2004.09.029*

Legua P, Forner-Giner MÁ, Nuncio-Jáuregui N, Hernández F (2016). Polyphenolic compounds, anthocyanins and antioxidant activity of nineteen pomegranate fruits: A rich source of bioactive compounds. Journal of Functional Foods 23:628-636. *https://doi.org/10.1016/j.jff.2016.01.043*

Li Y, Guo C, Yang J, Wei J, Xu J, Cheng S (2006). Evaluation of antioxidant properties of pomegranate peel extract in comparison with pomegranate pulp extract. Food Chemistry 96(2):254–260. *https://doi.org/10.1016/j.foodchem.2005.02.033*

Manera FJ, Legua P, Melgarejo P, Brotons JM, Hernández FCA, Martínez JJ (2013). Determination of a colour index for fruit of pomegranate varietal group 'Mollar de Elche'. Scientia Horticulturae 150:360-364. *https://doi.org/10.1016/j.scienta.2012.11.036*

Mayuoni-Kirshinbaum L & Porat R (2014). The flavor of pomegranate fruit: A review. Journal of the Science of Food and Agriculture 94(1):21-27. *https://doi.org/10.1002/jsfa.6311*

Melgarejo P, Salazar DM (2006). Seed characterisation of five new pomegranate (*Punica granatum* L.) varieties. Scientia Horticulturae 110:241-246. *https://doi.org/10.1016/j.scienta.2006.07.018*

Mirdehghan SH, Rahemi M, Serrano M, Guillén F, Martínez-Romero D, Valero D (2006). Prestorage heat treatment to maintain nutritive and functional properties during postharvest cold storage of pomegranate. Journal of Agricultural and Food Chemistry 54(22): 8495-8500. *https://doi.org/10.1021/jf0615146*

Moreno PM (2010). El Granado. I Jornada Nacional Sobre El Granado: Producción, Economía, Industralización, Alimentación y Salud.

Poyrazoğlu E, Gökmen V, Artık N (2002). Organic acids and phenolic compounds in pomegranates (Punica granatum L.) grown in Turkey. Journal of Food Composition and Analysis 15(5):567-575. *https://doi.org/10.1006/jfca.2002.1071*

Prieto JRE, Plascencia DD, Baquera PLO, Flores PFM, Rosas YP (2017). Granado (*Punica granatum* L.) la nueva alternativa en la reconversión sustentable de cultivos en Chihuahua. Índice 4.

Raffo A, La Malfa G, Fogliano V, Madani G, Quaglia G (2006). Seasonal variations in antioxidant components of cherry tomatoes (*Lycopersicon esculentum* cv. Naomi F1). Journal of Food Composition and Analysis 19:11-19.

Reza M, Ardekani S, Hajimahmoodi M, Reza Oveisi M, Sadeghi N, Jannat B, ... Moridi T (2011). Comparative antioxidant activity and total flavonoid content of Persian pomegranate (*Punica granatum* L.) cultivars. Iranian Journal of Pharmaceutical Research 10(3):519-524.

Shulman Y, Fainberstein L, Lavee S (1984). Pomegranate fruit development and maturation. Journal of Horticultural Science 59(2):265-274. *https://doi.org/10.1080/00221589.1984.11515196*

Shwartz E, Glazer I, Bar-Ya'akov I, Matityahu I, Bar-Ilan I, Holland D, Amir R (2009). Changes in chemical constituents during the maturation and ripening of two commercially important pomegranate accessions. Food Chemistry 115(3):965-973. *https://doi.org/10.1016/j.foodchem.2009.01.036*

SIAP (2020). Servicio de Información Agroalimentaria y Pesquera. *https://www.gob.mx/siap/prensa/atlas-agroalimentario-2020*

Singleton VL, Rossi JA (1965). Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. American Journal Enology and Viticulturae 16:144-158

Solomon A, Golubowicz S, Yablowicz Z, Grossman S, Bergman M, Gottlieb HE, Altman A... Flaishman MA (2006). Antioxidant activities and anthocyanin content of fresh fruits of common fig (*Ficus carica* L.). Journal of Agricultural and Food Chemistry 54(20):7717-7723. *https://doi.org/10.1021/jf060497h*

Tarantino A, Difonzo G, Lopriore G, Disciglio G, Paradiso VM, Gambacorta G, Caponio F (2020). Bioactive compounds and quality evaluation of 'Wonderful' pomegranate fruit and juice as affected by deficit irrigation. Journal of the Science of Food and Agriculture 100(15):5539-5545. *https://doi.org/10.1002/jsfa.10606*

Tehranifar A, Zarei M, Nemati Z, Esfandiyari B, Reza M (2010). Scientia Horticulturae Investigation of physico-chemical properties and antioxidant activity of twenty Iranian pomegranate (*Punica granatum* L.) cultivars. Scientia Horticulturae 126(2):180-185. *https://doi.org/10.1016/j.scienta.2010.07.001*

Tzulker R, Glazer I, Bar-Ilan I, Holland D, Aviram M, Amir R (2007). Antioxidant activity, polyphenol content, and related compounds in different fruit juices and homogenates prepared from 29 different pomegranate accessions. Journal of Agricultural and Food Chemistry 55(23):9559-9570. *https://doi.org/10.1021/jf071413n*

Viuda-Martos M, Ruiz-Navajas Y, Fernández-López J, Sendra E, Sayas-Barberá E, Pérez-Álvarez, JA (2011). Antioxidant properties of pomegranate (*Punica granatum* L.) bagasses obtained as co-product in the juice extraction. Food Research International 44(5):1217-1223. *https://doi.org/10.1016/j.foodres.2010.10.057*

Wetzstein HY, Zhang Z, Ravid N, Wetzstein ME (2011). Characterization of attributes related to fruit size in pomegranate. HortScience 46(6):908-912. *https://doi.org/10.21273/hortsci.46.6.908*

Wetzstein HY, Zhang Z, Ravid N, Wetzstein ME, Drogoudi PD, Tsipouridis C, ... Bashir-Sadr Z (2006). Composition of pomegranate juice. Food Chemistry 95(2):121-129. *https://doi.org/10.21273/hortsci.46.6.908*

Zarei M, Azizi M, Bashir-Sadr Z (2011). Evaluation of physicochemical characteristics of pomegranate (*Punica granatum* L.) fruit during ripening. Fruits 66(2):121-129. *https://doi.org/10.1051/fruits/2011021*

Zhang C, Whiting MD (2011). Improving 'Bing' sweet cherry fruit quality with plant growth regulators. Scientia Horticulturae 127(3):341-346. *https://doi.org/10.1016/j.scienta.2010.11.006*