

Research Article

Quantification of bioactive compounds and antioxidant ability of forty seven Iranian pomegranate cultivars

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Abstract

Pomegranate is recognized as such a unique fruit, having noticeably high antioxidant ability and health-promoting compounds. The huge diffusion of new cultivars needs the information of all plant features to satisfy the market request. The objective of the present study was to determine the bioactive compounds and antioxidant ability from the peel, leaf, and seed of 47 pomegranate cultivars grown in Iran. Pomegranate peel samples showed a higher level of total phenol, flavonoid, flavonol, soluble sugar, ascorbate, and antioxidant activity than leaf and seed. In peel samples, the highest level of total phenol, flavonol, flavonoid, anthocyanin, soluble sugar, ascorbate, and antioxidant activity was found in White Sweet Tochal, Shirin Shahsavari Sar Yazd, Zard Baghmalek, Atabaki Nayriz, Nabati Shahreza, Hasebe Sar Yazd, and Baghmalek, respectively. The leaves of Germeze Baghmalek, Shirin Ajan, Pust Sefeede Korak, Alake Saveh, Robab Neyriz, Syahe Dane Gome, and Shirin Neyriz had maximum levels of total phenol, flavonol, flavonoid, anthocyanin, soluble sugar, ascorbate, and antioxidant activity, respectively. The strongest level of total phenol, flavonol, flavonoid, anthocyanin, soluble sugar, ascorbate, and antioxidant activity in seed was in Nabati Shahreza, Alake Gharegae, Aarosak Korak, Sabz Baghmalek, Pust Sorkh Sar Yazd, Pust Nazok Saveh, and Shirin Taft, respectively. The correlation values between the bioactive compounds (phenol, flavonol, flavonoid, soluble sugar, and ascorbate) and antioxidant capacity of the peel, leaf, and seed, show that these metabolites are among the constituents contributing to the antioxidant ability of pomegranate. The data of current research can help to choose the pomegranate cultivars for commercial or medicinal perspectives.

Keywords: Antioxidant activity, Bioactive compounds, Leaf; Peel, Pomegranate, Seed

Abbreviations: DPPH 2,2'-Diphenylpicrylhydrazyl radicals, TCA trichloroacetic acid, DNPH dinitrophenyl hydrazine, DW dry weight, FW fresh weight

Introduction

The phytochemicals and/or secondary metabolites have biological functions responsible for plants' medicinal characteristics, also participating in the construction of drugs used by the medicinal industry to treat different diseases (Sreenivasulu and Fernie, 2022). Phenolic compounds are one of the main groups of secondary metabolites, including more than 8000 separate structures and comprising anthocyanins, flavonoids, and hydroxycinnamates. Medicinal and nutritional properties of phenolic compounds are long-recognized and have been the topic of recent investigations. In contrast to other groups of plant metabolites, the health-promoting characteristics of phenolic compounds in the diet were revealed to adjust energy metabolism, decreasing

cholesterol and blood pressure and declining endothelial dysfunction (Jukanti *et al.*, 2020).

The content of secondary metabolites in plants is influenced by environmental, genetic, and agronomic factors. Environmental factors such as rainfall, temperature, solar radiation, and humidity display a strong effect on the content of secondary metabolites in many plants. In *Tithonia diversifolia*, there were found combinations of sesquiterpenes, flavonoids, lactone, and *t*-cinnamic acid, which were directly linked to the quantity of rainfall and alterations in temperature (Sampaio *et al.*, 2016). So, plants can acclimate to the environment through several genetic and biochemical mechanisms, which can be helpful for the plant's survival as well as for obtaining drugs.

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Pomegranate (*Punica granatum* L.) is a fruit native from countries such as Iran, Afghanistan, and China that is consumed all over the world (Ismail *et al.*, 2012). Pomegranate and its derived products are considered suitable sources of bioactive metabolites. The characteristics of pomegranate depend on climatic factors, growing locations, cultivars, and maturity steps. Consumption of pomegranate leads to the cure of ulcers, diabetes, cancer, and cardiovascular diseases (Moga *et al.*, 2021). The properties of pomegranate, particularly the antioxidant, relate to its chemical composition. Pomegranate is rich in polyphenols, comprising phenolic compounds such as gallic acid, flavonoids such as anthocyanins, and tannins such as punicalagin. These metabolites can be found not only in the edible section but also in other sections of fruit that are thrown away after consumption, such as the peel. The non-edible parts of pomegranate fruit, namely lamellas and peels, display significantly more levels of these compounds than the edible parts (Tehraniyar *et al.*, 2010). Pomegranate's by-products include peel and seeds, which contain 50% and 12% of the whole fruit, respectively (Tehraniyar *et al.*, 2010). Due to the growing desire to use natural preservatives in the food industry, utilization of waste parts of pomegranate (peel) with high antioxidant activity could be valuable.

The aim of this work is to evaluate the antioxidant activity of different types of pomegranate cultivated in Iran. For this purpose, the seed, peel, and leaf of pomegranates grown in different areas were investigated in order to compare the antioxidant activity, total phenol, flavonoid, flavonol, soluble sugar, anthocyanin, and ascorbate. Also, we examined the correlation between bioactive compounds and antioxidant capacity. The other goal of this research was to provide helpful health information to consumers regarding the antioxidants of various types of pomegranates. Such information will assist in the cultivar selection for commercial production to meet market requests. Undoubtedly, our study is the first report that compares the antioxidant properties of seed, leaf, and peel in different cultivars of pomegranate. It shall be noted that our populations have not been compared so far.

Materials and methods

Sample preparation: The 47 cultivars of pomegranates cultivated in Iran were studied: Taghalid Malas Tochal (TMT), White Sweet Tochal (WST), Syahe Dane Tochal (SDT), Malas Tochal (MT), Saveh Tochal (ST), Pust Sefeede korak (PSK), Malas Dane Sefeede korak (MDSK), Gol Barik Korak (GBK), Aarosak Korak (AK), Ghapan Ajan (GA), Pust Nazok Ajan (PNA), Syahe Dane Ajan (SDA), Shirin Ajan (SA), Sabz Baghmalek (SB), Germeze Baghmalek (GB), Zard Baghmalek (ZB), Pust Nazok Saveh (PNS), Alake Saveh (AS), Post Koloft Saveh (PKS), Syahe Saveh (SS), Alake Gharegae (AG), Syahe Dane Gome (SDG), Safeed Karale (SK), Salimi (S), Malas Sori (MS), Safeed Shirin Paveh (SSP), Shirin Sharavane (SS),

Zhalkeh (Z), Dane Syahe Taft (DST), Gol Taft (GT), Malas Eslamiyeh Taft (MET), Eslamiyeh Taft (ET), Zagh Taft (ZT), Shirin Taft (ST), Hasebe Sar Yazd (HSY), Shirin Shahsavari Sar Yazd (SSSY), Pust Sorkh Sar Yazd (PSSY), Shirin Sar Yazd (SSY), Mohr Mahe Sar Yazd (MMSY), Shirin Neyriz (SN), Robab Neyriz (RN), Atabaki Nayriz (AN), Kaleh Sage Nayriz (KSN), Nabati Shahreza (NS), Diabete Shahreza (DS), Makhmale Shahreza (MS) and Dane Mashke Shahreza (DMS). The leaves and ripen fruits were collected from eight provinces of Iran (i.e., Tehran, Esfahan, Semnan, Yazd, Khuzestan, Arak, Kermanshah and Fars) in October, 2019. The seed and peel of the fruits were separated manually and stored in -20°C. The leaves were dried in an oven at 60°C for 48 h. Dried samples were powdered and stored at room temperature until extraction.

Assessment of total phenol and flavonol content:

In order to prepare the methanolic extract, 0.1 g of tissue was homogenized in 5 mL of 80% methanol and then centrifuged at 5000 rpm for 20 minutes. For the total phenol content measurement, 0.1 mL methanolic extract was mixed with 2.5 mL Folin–Ciocalteu reagent 10%. The mixtures were neutralized by 7% sodium bicarbonate, and then absorbance was recorded at 765 nm. Gallic acid was used as a standard for the calibration curve (Conde *et al.*, 1995).

Akkol *et al.* (2008) method was used for flavonol content measurement. In this method, 0.5 mL of 2% aluminum chloride and 1.5 mL of 5% sodium acetate were added to 0.5 mL of methanolic extract, and absorbance was recorded at 445 nm after 2.5 h, and rutin was used as a standard.

Determination of flavonoid and anthocyanin: The content of flavonoid was determined by the Chang *et al.* (2002) method. In this method 0.1 g of tissue was homogenized in 5 mL of methanol 80%. Methanolic extract (0.5 mL) was mixed with 1.5 mL of 80% methanol, 0.1 mL of aluminium chloride (10%), 0.1 mL of potassium acetate (1 M), and 2.8 mL of distilled water, and the absorbance was assayed at 415 nm after 30 min. The calibration curve was plotted by different concentrations of quercetin.

The content of anthocyanin was measured by 0.3% HCl in methanol at 25°C using the extinction coefficient ($33000 \text{ cm}^2 \text{ mol}^{-1}$) at 550 nm (Wagner, 1979).

Measurement of total soluble sugar: The soluble sugar content was determined by the Dubois *et al.* (1956) method. Tissues (0.1 g) were homogenized in 3 mL distilled water and then were centrifuged at 5000 rpm for 20 minutes. The extract (500 μL) was added with 450 μL distilled water, 500 μL phenol 5%, and 2.5 mL sulfuric acid 97%, and then the absorbance was measured at 485 nm after 30 min.

Determination of ascorbate content: In order to measure ascorbate content, 0.1 g of tissue was homogenized in TCA (6%). The extract (4 mL) was added with 2% dinitrophenyl hydrazine (2 mL) and 1 drop of 10% thiourea solution. The mixture was boiled

at 100°C for 15 min, and then 5 mL of sulfuric acid (80%) was added. The absorbance was recorded at 530 nm. The standard curve was prepared using different concentrations of ascorbic acid (Mukherjee and Choudhuri, 1983).

Assessment of antioxidant activity: Antioxidant activity of pomegranate extracts (leaf, peel, and seed) was determined with DPPH (1, 1-diphenyl-2-picrylhydrazyl) radical scavenging activity (Abe *et al.*, 1998). Tissues (0.1 g) were homogenized in 1 mL of 96% ethanol and then were centrifuged at 10000 rpm for 5 min. Ethanolic extract (20 µL) was mixed with 800 µL of DPPH (0.5 mM in ethanol), and the absorbance was measured at 517 nm after 30 min in darkness. Using the following equation, the free radical scavenging activity was calculated:

Inhibition of DPPH radical (%) = [(Absorbance of control - Absorbance of sample) / (Absorbance of control)] × 100

Statistical analysis: The current research was carried out as factorial based on a completely randomized design with 3 replications, and the data were analyzed using one-way variance analysis (ANOVA) in SPS software. Means were compared by Duncan's test at a probability of 0.05.

HCA (hierarchical cluster analysis) was used for evaluating correlation between each pair of variables and performed by online CIMminer software.

Results and discussion

Total phenol and flavonol content: The results for total phenol content measurement of the pomegranate from the different cultivars were displayed in table 1. A significant variation in total phenol content was found among the 47 cultivars studied, and the values ranged from 1931.9 to 12740.6 µg g⁻¹ FW in peel, 53.86 to 3821.79 µg g⁻¹ DW in leaf, and 10.81 to 145.05 µg g⁻¹ FW in seed. According to our results, the total phenol content of the peel was higher than the seed and leaf in all cultivars. Also, the content of total phenol in the leaf was significantly higher than that in the seed. In cultivars analyzed, the highest content of total phenol in peel, leaf, and seed was in WST, GB, and NS, respectively. The peel, leaf, and seed of MS, ST, and SSY had the lowest content of total phenol as compared to other cultivars, respectively.

Changes in the total flavonol content of the peel, leaf, and seed for 47 pomegranate cultivars were shown in Table 1. The peel of all cultivars showed more content of flavonol as compared to seed and leaf. The flavonol content of the leaf was higher than that of the seed. Among peel samples, SSSY (12117.86 µg g⁻¹ FW) and SK (12111.9 µg g⁻¹ FW) showed the highest flavonol level, and PSSY (532.02 µg g⁻¹ FW) displayed the lowest flavonol level. The flavonol content in leaves varied from 86.43 µg g⁻¹ DW in ZT to 1175.76 µg g⁻¹ DW in SA. The flavonol content of extracts varied within 3.21–40.11 µg g⁻¹ FW in seeds. In seeds, the highest and lowest flavonol content were detected in

AG and AK, respectively.

According to our results, the total phenol content of peels was higher than that of seeds and leaves. As confirmed data of the present study, the presence of phenolic content in pomegranate parts has been described to be maximum in peel (Pande and Akoh, 2009). The phenolic content of pomegranate is strongly affected by the cultivar, climate conditions, and agronomical practices (Cicero *et al.*, 2018). In other investigations, the total phenol content of different pomegranate cultivars exhibited significant differences (Mousavinejad *et al.*, 2009). The phenol content in some cultivars studied, such as peel of WST, leaf of GB, and seed of NS in the present work, was higher than in previous studies. The level of total phenol in each pomegranate can be a good symptom of its health benefits. Therefore, the utilization of these cultivars in the food and pharmaceutical industries is recommended. Clinical research has shown that the consumption of phenols in pomegranate improves biochemical parameters, comprising biomarkers of oxidative stress, chronic diseases related to over-generations of free radicals, and protection against the onset of age-related diseases (Kojadinovic *et al.*, 2017). All pomegranate phenolic compounds show antioxidant activity concomitant with the indirect inhibition of inflammatory signs. In this context, pomegranates could be considered such as a good natural drug (Lansky and Newman, 2007).

Flavonoid and anthocyanin content: As shown in table 2, a great difference in terms of flavonoid content was observed among the pomegranate cultivars, and the changes were statistically significant (P<0.05). The hierarchy for the contents observed was peel > leaf > seed. Russo *et al.* (2018) demonstrated that pomegranate peel samples had a higher content of flavonoid than other parts, which is consistent with current findings. Flavonoid content of extracts varied between 148.76–670.63 µg g⁻¹ FW in peel. This range was within 26.33–235.64 µg g⁻¹ DW in leaf extracts and 2.63–9.08 µg g⁻¹ FW in seed extracts. These values are higher than those found by Sabraoui *et al.* (2020). Statistical mean values revealed that the peel extract had approximately 73.85-fold higher total flavonoid than that of seed extract, and leaf extract had 25.95-fold more total flavonoid than that of seed extract. Among the cultivars analyzed, the ZB, PSK, and AK had the highest flavonoid content in peel, leaf, and seed, respectively.

Currently, the interest in possible health benefits of flavonoids has enhanced due to their strong antibacterial, antioxidant, and other various biological impacts. Flavonoids can inhibit hurt caused by free radicals, and they participate in the antioxidant mechanism by interacting with switch metal ions, particularly copper and/or iron (Prochazkova *et al.*, 2011). This difference in flavonoid content could be related to the type of cultivar, method of extraction, and environmental conditions. It was proposed that the high

Table 1. Total phenol and flavonol content in different parts of pomegranate cultivars

| Cultivar s | Total phenol content | | | Flavonol content | | |
|---------------|------------------------------------|------------------------------------|------------------------------------|------------------------------------|------------------------------------|------------------------------------|
| | Leaf ($\mu\text{g g}^{-1}$ DW) | Peel ($\mu\text{g g}^{-1}$ FW) | Seed ($\mu\text{g g}^{-1}$ FW) | Leaf ($\mu\text{g g}^{-1}$ DW) | Peel ($\mu\text{g g}^{-1}$ FW) | Seed ($\mu\text{g g}^{-1}$ FW) |
| TMT | 1220.23 \pm 13.2 ^{gh} | 7436.1 \pm 53.9 ^c | 93.44 \pm 4.9 ^{ef} | 278.20 \pm 2.9 ^q | 7623.2 \pm 29.9 ^{ab} | 18.77 \pm 0.17 ^{gj} |
| SST | 544.89 \pm 11.3 ^k | 12740.6 \pm 58.6 ^a | 82.26 \pm 3.1 ^g | 244.48 \pm 1.2 ^r | 5147.8 \pm 25.2 ^{bc} | 9.29 \pm 0.16 ^{qu} |
| SDT | 906.99 \pm 27.9 ⁱ | 5618.6 \pm 42.8 ^{ef} | 36.61 \pm 1.5 ^m | 470.78 \pm 2.9 ^l | 3609.16 \pm 16 ^{bc} | 13.83 \pm 54 ^{jr} |
| MT | 1180.03 \pm 17.2 ^h | 6112.6 \pm 92.5 ^d | 47.96 \pm 1.6 ^k | 638.09 \pm 1.6 ^h | 4670.9 \pm 42.9 ^{bc} | 10.66 \pm 46 ^{nt} |
| ST | 53.86 \pm 3 ⁿ | 3213.6 \pm 35.8 ^h | 64.89 \pm 3.8 ^{hi} | 598.84 \pm 4.4 ⁱ | 4334.6 \pm 14 ^{bc} | 15.66 \pm 0.11 ^{ip} |
| PSK | 2214.05 \pm 11.6 ^d | 7223.4 \pm 47.2 ^c | 73.39 \pm 1.7 ^h | 539.05 \pm 6.1 ^j | 2396.7 \pm 21.8 ^{bc} | 5.79 \pm 0.44 ^{sw} |
| MDSK | 1311.06 \pm 7.2 ^{fg} | 2276.2 \pm 43.3 ^j | 83.35 \pm 3.7 ^g | 342.08 \pm 3.4 ^p | 960.03 \pm 26.3 ^c | 12.16 \pm 0.36 ^{l-r} |
| MDSK | 1549.8 \pm 22.6 ^{ef} | 3411.82 \pm 45.5 ^h | 80.92 \pm 9 ^g | 367.56 \pm 2.4 ^{mn} | 4073.83 \pm 20.8 ^{bc} | 15.91 \pm 0.31 ^{ju} |
| GBK | 447.56 \pm 17.3 ^l | 6440.1 \pm 19.2 ^d | 98.08 \pm 1.8 ^e | 142.52 \pm 2.3 ^s | 4522.03 \pm 18.6 ^{bc} | 3.21 \pm 0.17 ^w |
| GA | 932.3 \pm 13.6 ⁱ | 8790.9 \pm 46.3 ^b | 122.63 \pm 5.5 ^d | 802.86 \pm 4.4 ^f | 4876.53 \pm 14.3 ^{bc} | 19.65 \pm 0.40 ^{fj} |
| PNA | 2024.69 \pm 13.2 ^d | 3271.8 \pm 9.9 ^h | 78.29 \pm 0.7 ^{gh} | 1035.78 \pm 4.8 ^b | 3394.53 \pm 18 ^{bc} | 7.82 \pm 0.26 ^{rw} |
| SDA | 2059.03 \pm 16.1 ^d | 5830.8 \pm 30.6 ^e | 88.54 \pm 0.6 ^f | 469.18 \pm 3.2 ^l | 4285.3 \pm 24.7 ^{bc} | 24.97 \pm 0.44 ^{cf} |
| SA | 1627.53 \pm 17.5 ^e | 5192.3 \pm 43.3 ^f | 80.11 \pm 1.3 ^g | 1175.76 \pm 5.7 ^a | 4996.5 \pm 22.6 ^{bc} | 8.37 \pm 0.59 ^{ou} |
| SB | 1128.4 \pm 12.6 ^h | 2573.48 \pm 31.7 ⁱ | 77.58 \pm 1 ^{gh} | 717.63 \pm 5.9 ^g | 2788.96 \pm 9.5 ^{bc} | 9.74 \pm .57 ^{pu} |
| GB | 3821.79 \pm 11.7 ^a | 4101.7 \pm 41.4 ^{fg} | 91 \pm 1.4 ^{ef} | 1003.12 \pm 4.6 ^c | 1154.13 \pm 18.5 ^{bc} | 11.9 \pm 0.26 ^{mr} |
| ZB | 3140.16 \pm 15.2 ^b | 8831.3 \pm 37.8 ^b | 79.31 \pm 3.1 ^g | 141.43 \pm 3 ^s | 5614.66 \pm 35.2 ^{bc} | 4.19 \pm 0.43 ^{uw} |
| PNS | 906.14 \pm 11.23 ⁱ | 3414 \pm 50.3 ^{gh} | 135.83 \pm 3.5 ^b | 859.63 \pm 1.7 ^e | 1383.3 \pm 8.5 ^{bc} | 30.22 \pm 0.53 ^{bc} |
| AS | 678.17 \pm 6 ^j | 4335.8 \pm 22.8 ^{fg} | 84.72 \pm 1 ^g | 352.87 \pm 4.4 ^o | 1240.73 \pm 17.9 ^{bc} | 17.99 \pm 0.26 ^{gl} |
| PKS | 1645.73 \pm 27.5 ^e | 5890.8 \pm 52.4 ^e | 88.60 \pm 1.2 ^f | 494.57 \pm 3.2 ^k | 1894.2 \pm 10.6 ^{bc} | 19.16 \pm 0.65 ^{gj} |
| SS | 1355.4 \pm 65.2 ^g | 3605.4 \pm 41.5 ^g | 75.03 \pm 1.8 ^{gh} | 842.78 \pm 4.6 ^e | 1963.91 \pm 13 ^{bc} | 21.97 \pm 0.17 ^{fh} |
| AG | 224.14 \pm 4.1 ^m | 3959.7 \pm 21.8 ^g | 100.64 \pm 4.1 ^e | 358.09 \pm 1.3 ^{no} | 838.32 \pm 22.5 ^c | 40.11 \pm 0.39 ^a |
| SDG | 73.33 \pm 4.3 ⁿ | 3511.6 \pm 30.6 ^g | 127.28 \pm 1.8 ^c | 537.69 \pm 6.2 ^j | 2982.8 \pm 12.9 ^{bc} | 18.58 \pm 0.26 ^{g-k} |
| SK | 548.43 \pm 21.5 ^k | 4501.8 \pm 46.4 ^f | 67.76 \pm 1.7 ^{hi} | 701.65 \pm 3.5 ^g | 12111.9 \pm 19.3 ^a | 12.06 \pm 0.44 ^{m-r} |
| S | 752.01 \pm 4.40 ^j | 4891.3 \pm 59.1 ^d | 81.13 \pm 2.3 ^g | 113.93 \pm 2.6 ^u | 1929.9 \pm 14.9 ^{bc} | 11.29 \pm 0.60 ^{m-s} |
| MS | 2724.56 \pm 17.5 ^c | 6857.5 \pm 25 ^d | 61.39 \pm 1.6 ⁱ | 304.50 \pm 4.3 ^q | 3860.1 \pm 27.7 ^{bc} | 11.85 \pm 0.60 ^{m-r} |
| SSP | 613.32 \pm 21.4 ^j | 5548.2 \pm 42.7 ^{ef} | 112.36 \pm 2.3 ^d | 149.00 \pm 2.8 ^s | 5364.73 \pm 26.2 ^{bc} | 21.89 \pm 0.39 ^{f-h} |
| SS | 844.29 \pm 3.7 ⁱ | 7998.6 \pm 33.4 ^c | 69.53 \pm 1.3 ^{hi} | 205.28 \pm 3.3 ^r | 4186.46 \pm 18.2 ^{bc} | 4.4 \pm 0.160 ^{u-w} |
| Z | 1039.6 \pm 9.2 ^h | 5631.6 \pm 30 ^{ef} | 57.16 \pm 3.4 ⁱ | 125.29 \pm 2.8 ^t | 4667.77 \pm 31.3 ^{bc} | 12.69 \pm 0.360 ^{m-r} |
| DST | 1273.73 \pm 10.3 ^g | 2860.4 \pm 43.7 ⁱ | 89.14 \pm 1.4 ^f | 664.75 \pm 2.8 ^h | 546.26 \pm 20.1 ^c | 27.63 \pm 0.51 ^{b-e} |
| GT | 1739.03 \pm 3.6 ^{de} | 6244.8 \pm 41.6 ^d | 115.15 \pm 3.4 ^d | 931.17 \pm 4.4 ^d | 4344.7 \pm 22.3 ^{bc} | 17.03 \pm 0.39 ^{h-m} |
| MAT | 2275.38 \pm 12.1 ^f | 5613.1 \pm 42.5 ^e | 59.33 \pm 0.66 ⁱ | 375.59 \pm 3.8 ^m | 2404.46 \pm 10.5 ^{bc} | 4.95 \pm 0.16 ^{t-w} |
| AT | 1495.7 \pm 11 ^d | 3080.4 \pm 44.3 ^h | 51.85 \pm 1.4 ^j | 415.37 \pm 3.4 ^l | 3770.7 \pm 29 ^{bc} | 9.87 \pm 0.34 ^{o-u} |
| ZT | 944.18 \pm 18.2 ⁱ | 2742.93 \pm 11.4 ⁱ | 43.69 \pm 2.7 ^l | 86.43 \pm 2.5 ^v | 2135.2 \pm 19.6 ^{bc} | 12.34 \pm 0.37 ^{l-r} |
| ST | 2047.9 \pm 11.9 ^d | 7744.2 \pm 32.3 ^c | 71.54 \pm 1 ^h | 252.98 \pm 3 ^r | 3421.56 \pm 17.7 ^{bc} | 12.82 \pm 0.39 ^{k-r} |
| HSY | 2365.73 \pm 16.6 ^d | 6257 \pm 18 ^d | 118.8 \pm 1.1 ^d | 169.71 \pm 2.1 ^s | 683.65 \pm 11.9 ^c | 23.11 \pm 0.38 ^{d-f} |
| SSSY | 1465.46 \pm 27.2 ^f | 4660.3 \pm 48.7 ^f | 77.59 \pm 1.6 ^{gh} | 707.10 \pm 4.5 ^g | 12117.86 \pm 32.4 ^a | 21.81 \pm 1.2 ^{f-h} |
| PSSY | 2300.02 \pm 17.4 ^d | 3688.2 \pm 29.4 ^g | 94.53 \pm 0.9 ^{ef} | 368.56 \pm 2.7 ^{mn} | 532.02 \pm 16.4 ^c | 8.27 \pm 1.1 ^{o-w} |
| SSY | 1884.7 \pm 26.5 ^{de} | 6297.9 \pm 41.9 ^d | 10.81 \pm 2.9 ⁿ | 1005.05 \pm 3.4 ^c | 4658.76 \pm 23.5 ^{bc} | 11.06 \pm 0.18 ^{m-s} |
| MMSY | 862.09 \pm 14.1 ⁱ | 4460.1 \pm 34.8 ^f | 101.08 \pm 0.4 ^e | 362.37 \pm 3.8 ^{no} | 826 \pm 22.6 ^c | 15.78 \pm 0.62 ^{j-o} |
| SN | 2149.26 \pm 25.2 ^d | 3800.8 \pm 17.3 ^g | 92.28 \pm 2.3 ^{ef} | 1185.12 \pm 4.4 ^a | 3627.23 \pm 12.7 ^{bc} | 4.73 \pm 0.25 ^{u-w} |
| RN | 1479.13 \pm 14.5 ^f | 4029.4 \pm 58.5 ^g | 138.99 \pm 2.4 ^b | 503.96 \pm 3.5 ^k | 2833.96 \pm 16.9 ^{bc} | 31.31 \pm 0.36 ^b |
| AN | 1557.66 \pm 6.6 ^{ef} | 3036.9 \pm 10.5 ^{gh} | 69.85 \pm 1.5 ^{hi} | 232.91 \pm 3.7 ^r | 2060.33 \pm 26.5 ^{bc} | 15.68 \pm 0.43 ^{j-n} |
| KSN | 1336.26 \pm 17.8 ^{fg} | 3815.2 \pm 38.1 ^g | 98.02 \pm 0.9 ^e | 406.05 \pm 2.3 ^l | 1764.23 \pm 16.2 ^{bc} | 29.44 \pm 0.90 ^{bc} |
| NS | 1541.5 \pm 21.3 ^{ef} | 5394.3 \pm 45.1 ^f | 145.05 \pm 2.5 ^a | 203.97 \pm 3.4 ^r | 4050.26 \pm 20.3 ^{bc} | 14.16 \pm 0.88 ^{j-q} |
| DS | 204.83 \pm 9 ^m | 3110.9 \pm 35.4 ^h | 112.66 \pm 2.5 ^d | 339.12 \pm 2 ^p | 2061.3 \pm 32.3 ^{bc} | 28.39 \pm 0.50 ^{b-d} |
| MS | 1021.25 \pm 18.7 ^h | 1931.92 \pm 17.2 ^k | 76.42 \pm 1.3 ^{gh} | 672.1 \pm 1.9 ^h | 2308.26 \pm 12.5 ^{bc} | 20.37 \pm 0.55 ^{f-i} |
| DMS | 1227.6 \pm 14.2 ^{gh} | 3477.6 \pm 23.2 ^{gh} | 84.75 \pm 0.5 ^g | 537.59 \pm 4.3 ^j | 2657.86 \pm 16.6 ^{bc} | 22.69 \pm 0.34 ^{e-h} |

Values are means \pm SE of three replicates. Different letters indicated significant ($P < 0.05$) differences.

content of bioactive compounds such as flavonoids are available in non-edible parts, which could be applied for different aims in the food industry, such as enrichment or improvement of new crops (Kulkarni *et al.*, 2004).

Anthocyanins are the main source for the violet-blue

and attractive red colors of pomegranate peels and arils, and reveal significant antioxidant activity (Schwartz *et al.*, 2009). As shown in Table 1, a great difference in terms of anthocyanin content was witnessed among the pomegranate cultivars. Pomegranate leaf samples were

Table 2. Flavonoid and anthocyanin content in different parts of pomegranate cultivars

| Cultivars | Flavonoid content | | | Anthocyanin content | | |
|-----------|------------------------------------|------------------------------------|------------------------------------|------------------------------------|------------------------------------|------------------------------------|
| | Leaf ($\mu\text{g g}^{-1}$ DW) | Peel ($\mu\text{g g}^{-1}$ FW) | Seed ($\mu\text{g g}^{-1}$ FW) | Leaf ($\mu\text{g g}^{-1}$ DW) | Peel ($\mu\text{g g}^{-1}$ FW) | Seed ($\mu\text{g g}^{-1}$ FW) |
| TMT | 3.37 \pm 0.13 ^{h-j} | 563.82 \pm 4 ^e | 144.80 \pm 2.5 ^{ef} | 5.50 \pm 0.003 ^g | 1.9 \pm 0.003 ^c | 0.0233 \pm 0 ^k |
| SST | 3.73 \pm 0.24 ^{g-i} | 534.86 \pm 4.4 ^f | 58.52 \pm 1.6 ^l | 5.64 \pm 0.014 ^g | 1.1 \pm 0.003 ^g | 0.068 \pm 0.0007 ^f |
| SDT | 4.08 \pm 0.11 ^{e-j} | 592.59 \pm 3.5 ^d | 146 \pm 3.5 ^{ef} | 7.6 \pm 0.024 ^c | 0.79 \pm 0.003 ^k | 0.0186 \pm 0.0013 ^l |
| MT | 3.09 \pm 0.42 ^{ij} | 649.73 \pm 2.3 ^b | 137.69 \pm 1.6 ^f | 6.7 \pm 0.014 ^e | 1.6 \pm 0.003 ^d | 0.044 \pm 0 ^{hi} |
| ST | 5.899 \pm 0.8 ^{bn} | 567.32 \pm 4.8 ^e | 26.33 \pm 1.5 ⁿ | 6.7 \pm 0 ^e | 0.78 \pm 0.003 ^k | 0.0411 \pm 0.0007 ⁱ |
| PSK | 235.64 \pm 1.9 ^a | 554.78 \pm 4.8 ^{ef} | 3.43 \pm 0.12 ^{g-j} | 3.84 \pm 0.011 ⁱ | 0.85 \pm 0.003 ^j | 0.0357 \pm 0.0007 ^{jk} |
| MDSK | 148.60 \pm 2.3 ^{ef} | 231.82 \pm 2.7 ^k | 3.22 \pm 0.53 ^g | 7.2 \pm 0.024 ^d | 0.91 \pm 0.007 ^j | 0.00543 \pm 0.0007 ⁿ |
| MDSK | 136.79 \pm 3 ^f | 364.61 \pm 1.1 ^j | 4.91 \pm 0.61 ^{c-j} | 5.8 \pm 0.012 ^f | 0.99 \pm 0.006 ⁱ | 0.0543 \pm 0.0007 ^g |
| GBK | 66.09 \pm 2.6 ^k | 264.59 \pm 4 ^m | 9.08 \pm 0.40 ^a | 5.24 \pm 0.010 ^h | 0.98 \pm 0.003 ⁱ | 0.107 \pm 0.0007 ^b |
| GA | 139.66 \pm 5.3 ^f | 609.35 \pm 2.4 ^c | 7.16 \pm 1.2 ^{ad} | 7.16 \pm 0.021 ^d | 0.89 \pm 0.003 ^j | 0.086 \pm 0.0007 ^e |
| PNA | 226.74 \pm 1.8 ^b | 490.91 \pm 4.2 ^g | 4.80 \pm 0.36 ^{c-j} | 7.9 \pm 0.006 ^c | 1.01 \pm 0.003 ^h | 0.0831 \pm 0.0007 ^e |
| SDA | 111.08 \pm 2.7 ^g | 536.34 \pm 3.7 ^f | 3.88 \pm 0.69 ^{f-g} | 7.06 \pm 0.011 ^d | 0.88 \pm 0.003 ^j | 0.0582 \pm 0.0013 ^g |
| SA | 210.23 \pm 2.6 ^c | 603.86 \pm 1 ^{cd} | 2.97 \pm 0.44 ^{i-j} | 5.56 \pm 0.010 ^g | 1.4 \pm 0 ^d | 0.0551 \pm 0.0007 ^g |
| SB | 3.27 \pm 0.35 ^{h-j} | 620.59 \pm 1.8 ^c | 155.71 \pm 4.3 ^e | 6.34 \pm 0.003 ^e | 1.2 \pm 0.003 ^f | 0.201 \pm 0.007 ^a |
| GB | 2.63 \pm 0.13 ^j | 251.56 \pm 2.3 ^m | 120.66 \pm 1.7 ^g | 2.9 \pm 0.003 ^k | 2.1 \pm 0.006 ^b | 0.0434 \pm 0.0007 ^{hi} |
| ZB | 4.17 \pm 0.91 ^{e-j} | 670.63 \pm 4.2 ^a | 101.12 \pm 1.9 ^h | 2.5 \pm 0.017 ^l | 0.86 \pm 0.003 ^j | 0.0815 \pm 0 ^e |
| PNS | 5.32 \pm 0.95 ^{c-j} | 410.83 \pm 3.5 ⁱ | 101.69 \pm 3.3 ^h | 5.85 \pm 0 ^f | 0.67 \pm 0.006 ^l | 0.1009 \pm 0.0007 ^c |
| AS | 4.46 \pm 0.8 ^{d-j} | 489.56 \pm 4.4 ^h | 92.54 \pm 3.2 ⁱ | 8.61 \pm 0.010 ^a | 1.15 \pm 0.007 ^j | 0.0419 \pm 0.0013 ⁱ |
| PKS | 6.78 \pm 0.25 ^{a-f} | 359.84 \pm 2.6 ^j | 149.11 \pm 2.7 ^{ef} | 3.9 \pm 0.015 ⁱ | 1.21 \pm 0 ^f | 0.048 \pm 0 ^h |
| SS | 6.76 \pm 0.28 ^{a-f} | 603.27 \pm 1.1 ^{cd} | 151.82 \pm 3.4 ^e | 8.2 \pm 0.018 ^b | 1.3 \pm 0.007 ^e | 0.0489 \pm 0 ^h |
| AG | 6.12 \pm 0.86 ^{b-i} | 525.47 \pm 3.3 ^f | 35.02 \pm 0.73 ^l | 7.7 \pm 0.013 ^c | 0.62 \pm 0.003 ^m | 0.0473 \pm 0.0007 ^h |
| SDG | 4.18 \pm 0.28 ^{e-j} | 595.89 \pm 2.5 ^{cd} | 32.52 \pm 1.3 ^m | 6.5 \pm 0.011 ^e | 0.47 \pm 0 ^o | 0.045 \pm 0.0007 ^h |
| SK | 3.71 \pm 0.08 ^{g-j} | 383.87 \pm 2.7 ^j | 29.05 \pm 0.90 ^m | 6.4 \pm 0.012 ^e | 1.08 \pm 0.003 ^h | 0.027 \pm 0.0007 ^k |
| S | 4.55 \pm 0.83 ^{d-j} | 463.52 \pm 3.2 ⁱ | 55.32 \pm 3.6 ^l | 7.8 \pm 0.023 ^c | 1.03 \pm 0.003 ^h | 0.0240 \pm 0.0007 ^k |
| MS | 5.49 \pm 0.24 ^{c-j} | 610.59 \pm 4.3 ^d | 173.68 \pm 2.7 ^d | 6.03 \pm 0.007 ^f | 0.72 \pm 0 ^k | 0.0178 \pm 0.0007 ^l |
| SSP | 4.66 \pm 0.97 ^{d-j} | 531.3 \pm 2.6 ^f | 79.51 \pm 2.4 ^j | 6.1 \pm 0.006 ^f | 1.09 \pm 0.003 ^h | 0.067 \pm 0.0013 ^f |
| SS | 3.33 \pm 0.36 ^{hj} | 562.82 \pm 1.4 ^e | 115.62 \pm 3.6 ^g | 7.07 \pm 0.012 ^c | 1.47 \pm 0 ^d | 0.0124 \pm 0.0007 ^m |
| Z | 4.61 \pm 0.56 ^{d-j} | 636.01 \pm 3.5 ^b | 85.63 \pm 3.4 ^{ij} | 7.6 \pm 0.015 ^c | 0.75 \pm 0.003 ^k | 0.022 \pm 0.0007 ^k |
| DST | 5.005 \pm 0.72 ^{c-j} | 181.10 \pm 3.1 ⁿ | 66.04 \pm 1.7 ^k | 2.2 \pm 0.11 ^m | 1.98 \pm 0.003 ^c | 0.052 \pm 0.0007 ^g |
| GT | 3.63 \pm 0.91 ^{g-j} | 560.55 \pm 4.9 ^{ef} | 67.72 \pm 1 ^k | 3.19 \pm 0.015 ^k | 1.13 \pm 0.009 ^g | 0.037 \pm 0 ^j |
| MAT | 3.80 \pm 0.17 ^{g-j} | 494.82 \pm 2.2 ^g | 94.12 \pm 2.1 ⁱ | 3.52 \pm 0.011 ^j | 1.4 \pm 0.003 ^d | 0.044 \pm 0 ^{hi} |
| AT | 3.61 \pm 0.80 ^{g-j} | 260.7 \pm 1.9 ^m | 159.68 \pm 5 ^e | 4.8 \pm 0.029 ^h | 1.34 \pm 0 ^e | 0.039 \pm 0.0013 ^j |
| ZT | 3.05 \pm 0.49 ^{ij} | 331.26 \pm 1.9 ^k | 61.28 \pm 1.8 ^l | 5.5 \pm 0 ^g | 1.14 \pm 0.006 ^g | 0.051 \pm 0 ^g |
| ST | 4.21 \pm 0.29 ^{e-j} | 577.62 \pm 3.7 ^e | 128.23 \pm 2 ^{gf} | 3.8 \pm 0.006 ⁱ | 0.63 \pm 0.006 ^m | 0.0209 \pm 0 ^l |
| HSY | 5.17 \pm 0.12 ^{c-j} | 220.59 \pm 3.1 ⁱ | 85.37 \pm 3.1 ^{ij} | 2.14 \pm 0.011 ⁿ | 1.48 \pm 0.003 ^d | 0.033 \pm 0.0007 ^{jk} |
| SSSY | 4.66 \pm 0.17 ^{d-j} | 642.8 \pm 2.3 ^b | 153.98 \pm 2.6 ^e | 3.7 \pm 0.015 ^{ij} | 1.21 \pm 0.003 ^{fg} | 0.0528 \pm 0.0007 ^g |
| PSSY | 4.45 \pm 0.51 ^{d-j} | 148.76 \pm 4.9 ^o | 134.93 \pm 3.1 ^f | 4.05 \pm 0.010 ⁱ | 1.23 \pm 0.003 ^f | 0.107 \pm 0.0007 ^b |
| SSY | 4.51 \pm 0.66 ^{d-j} | 564.46 \pm 2.5 ^e | 106.86 \pm 1.6 ^{gh} | 2.7 \pm 0.014 ^k | 1.28 \pm 0 ^f | 0.0365 \pm 0.0007 ^j |
| MMSY | 4.73 \pm 0.97 ^{c-j} | 233.23 \pm 1.9 ⁱ | 88.82 \pm 3.6 ⁱ | 3.8 \pm 0.006 ⁱ | 1.27 \pm 0.003 ^f | 0.0706 \pm 0.0007 ^f |
| SN | 5.40 \pm 1 ^{c-j} | 315.82 \pm 3.5 ^l | 152.07 \pm 3.6 ^e | 2.7 \pm 0.020 ^k | 1.28 \pm 0 ^f | 0.0761 \pm 0.0007 ^{ef} |
| RN | 8.54 \pm 1.2 ^{ab} | 444.49 \pm 4.2 ⁱ | 108.41 \pm 1 ^{gh} | 3.3 \pm 0.015 ^j | 1.59 \pm 0 ^d | 0.0388 \pm 0.0007 ^j |
| AN | 4.59 \pm 0.37 ^{d-j} | 654.55 \pm 5.5 ^b | 92.001 \pm 2 ⁱ | 3.4 \pm 0.014 ^j | 2.26 \pm 0.003 ^a | 0.0349 \pm 0 ^{jk} |
| KSN | 4.242 ^{d-j} | 588.82 \pm 2.1 ^d | 130.12 \pm 1.4 ^{fg} | 3.7 \pm 0.20 ^{ij} | 0.98 \pm 0.003 ⁱ | 0.091 \pm 0.0007 ^d |
| NS | 4.89 \pm 0.45 ^{c-j} | 479.96 \pm 2.6 ^h | 78.15 \pm 1.4 ^j | 4.5 \pm 0.012 ^h | 0.75 \pm 0 ^k | 0.026 \pm 0.0007 ^k |
| DS | 6.37 \pm 0.41 ^{a-g} | 178.12 \pm 13.2 ⁿ | 68.43 \pm 3 ^k | 3.8 \pm 0.017 ⁱ | 0.76 \pm 0.003 ^k | 0.0792 \pm 0.0007 ^{ef} |
| MS | 7.57 \pm 0.38 ^{a-c} | 398.79 \pm 6.6 ^j | 80.60 \pm 2.4 ^j | 4.8 \pm 0.015 ^h | 0.55 \pm 0 ⁿ | 0.0194 \pm 0.0007 ^l |
| DMS | 7.01 \pm 0.15 ^{a-e} | 452.97 \pm 4.5 ⁱ | 119.97 \pm 0.53 ^g | 3.7 \pm 0.011 ^{ij} | 1.21 \pm 0.003 ^{fg} | 0.0582 \pm 0.002 ^g |

Values are means \pm SE of three replicates. Different letters indicated significant ($P < 0.05$) differences.

quantitatively the richest in anthocyanin content, whereas pomegranate seed samples were the poorest. For leaf and peel samples, anthocyanin content ranged between 2.14 and 8.61 $\mu\text{g g}^{-1}$ DW and between 0.55 and 2.26 $\mu\text{g g}^{-1}$ FW, respectively. In leaf samples, the AS

showed the highest anthocyanin content, while HSY had the lowest anthocyanin content. The anthocyanin content in AN was the highest, while MS exhibited the lowest anthocyanin content in peel samples. The anthocyanin content in the seeds ranged from 0.005 to

0.201 $\mu\text{g g}^{-1}$ FW, with the lowest and highest values observed for MDSK and SB, respectively.

Anthocyanins of pomegranate originate mostly from the arils and exhibit low bioavailability, and thus their function in pomegranate bioactivity is yet to be well-known. However, consumers are drawn to the pomegranate fruit and its products with the unique strong red color. So, the industry offers red-colored, rich pomegranates to increase marketing. Amounts of anthocyanin content in current work were more than the results reported for ten pomegranate cultivars grown in Iran (Akhavan *et al.*, 2015). These data showed that the levels of anthocyanin varied among various cultivars of pomegranate, and cultivar was the main factor that influenced this parameter (Kalaycıoglu and Erım, 2017). These results are very important because they can select cultivars with high anthocyanin content to extract and utilize them in the medicinal industry or such as food additives.

Total soluble sugar content: Total soluble sugar content of peel, leaf, and seed in each cultivar of pomegranate was exhibited in table 3. The content of soluble sugar in the leaf was significantly lower than that of the peel and seed. In leaf samples, the RN cultivar had the highest (19.51 mg g^{-1} DW) soluble sugar content, whereas its DS was the lowest (3.11 mg g^{-1} DW). In current work, cultivars with more seed-soluble sugar have lower peel-soluble sugar content and vice versa. According to the results, the soluble sugar content varied within 5.9–22.1 mg g^{-1} FW in seed and 6.8–29.7 mg g^{-1} FW in peel. The highest total soluble sugar content of peel and seed was in NS and PSSY, while the peel of SDG and seed of SS had the lowest content.

The taste of pomegranate fruit is determined by the level of organic acids and sugar content. Soluble sugars in fruits of wild and cultivated pomegranates varied from 17.57 to 19.99 $\text{mg}/100\text{ g}$ and 13.13 to 16.55 $\text{mg}/100\text{ g}$ of fruit with a high level of glucose and fructose, respectively (Hasnaoui *et al.*, 2011). The starch hydrolysis is one of the events occurring throughout fruit ripening that store into simple sugars in the early steps of fruit development. Sucrose and starch convert into glucose during fruit ripening. So, the content of total sugars significantly enhanced during fruit ripening. Some reports have presented high differences in soluble sugar content in different pomegranate cultivars from several geographic areas (Mekni *et al.*, 2019). Clearly, differences in this content could be associated with the diversity of agro-climatic conditions, but we believe that cultivar impact has the main impact on the content of sugar. Also, soluble sugars can feed NADPH-producing metabolic pathways, which can be involved in ROS scavenging. Thus, soluble sugars can act such as antioxidants. So, in addition to the taste of pomegranate, they can also be involved in the antioxidant power of pomegranate.

Ascorbate content: Significant differences ($P < 0.05$) were perceived in the ascorbate content of the studied

cultivars (Table 3). The order of the ascorbate content was peel > leaf > seed, with peel showing greater content than their respective leaf and seed. Ascorbate content of peels ranged from 3184.6 $\mu\text{g g}^{-1}$ FW to 128162 $\mu\text{g g}^{-1}$ FW, while it ranged in seed from 177.4 $\mu\text{g g}^{-1}$ FW to 5687.03 $\mu\text{g g}^{-1}$ FW. Ascorbate content in pomegranate peels is approximately 22.53-fold greater than that in seeds. The ascorbate content of leaves was 639.9–41057.3 $\mu\text{g g}^{-1}$ DW. Ascorbate values in the current work were lower than results reported by Peng (2019) and Tehranifar *et al.* (2010). However, Ferrara *et al.* (2011) observed significant changes in ascorbate or vitamin C content among different genotypes of pomegranate, ranging from 89.0 to 236.3 mg/L . Ascorbic acid, normally known as vitamin C, plays an important function in the human body. It is essential for the synthesis of collagen, a protein that has several connective roles in the body. As an antioxidant, it reacts with peroxide and histamine to decrease inflammatory signs (Barrita and Sanchez, 2013). Also, vitamin C content is generally regarded as such a nutritional factor to evaluate the fruit quality. Therefore, cultivars with higher ascorbate content can be more important. In peel, leaf, and seed samples, HSY, SDG, and PNS had the highest content of ascorbate, respectively.

Antioxidant activity: The DPPH radical scavenging method is usually employed to estimate the capacity of antioxidants to scavenge free radicals. In this work, the differences in antioxidant ability among the pomegranate cultivars were statistically significant (Table 4). Among the 47 pomegranate cultivars, the DPPH scavenging activity of peels was 90.27–95.79%, 28.96–92.09% for leaves, and 0.55–8.99% for seeds. Peels exhibited the strongest DPPH free radical scavenging ability. Other studies revealed antioxidant activity of pomegranate peel 2.8-fold more than pomegranate leaf and seed extract (Ismail *et al.*, 2012). While in the current study, the antioxidant activity of peel was 3.11 and 10.65-fold higher, respectively, than those of leaf and seed. Among the analyzed samples, peel extracts from ZB and ZT showed the strongest antioxidant activity, whereas for leaf and seed samples, the highest values were obtained for SN and ST, respectively. The lowest DPPH of pomegranate peels was observed in SS, AS, and PNA. In leaf and seed samples, DS and MMSY had lower antioxidant ability than other cultivars, respectively.

The primary reason for the positive health impact of pomegranate is the unique antioxidant ability of this fruit. In particular, it has been revealed that pomegranate juice has the maximum antioxidant activity among polyphenol-rich beverages. The high antioxidative characteristics of pomegranate have made it as the topic of numerous functional researches and a number of investigations with *in vitro* and *in vivo* models. On the other hand, the diversity of fruit quality within cultivars, with regard to nutraceutical value, would be beneficial to detect and to cultivate cultivars having an upper commercial value (Di Stefano *et al.*,

Table 3. Soluble sugar and ascorbate content in different parts of pomegranate cultivars.

| Cultivars | Soluble sugar content | | | Ascorbate content | | |
|-----------|---------------------------------|---------------------------------|---------------------------------|---------------------------------|---------------------------------|---------------------------------|
| | Leaf (mg g ⁻¹ DW) | Peel (mg g ⁻¹ FW) | Seed (mg g ⁻¹ FW) | Leaf (μg g ⁻¹ DW) | Peel (μg g ⁻¹ FW) | Seed (μg g ⁻¹ FW) |
| TMT | 5.88±0.034 ^{eh} | 16.06±0.14 ^e | 7.85±0.057 ^j | 4510.1±187.9 ⁱ | 41296±843.4 ^f | 3344.6±65.1 ^{ed} |
| SST | 10.15±0.043 ^{be} | 13.4±0.13 ^g | 12.2±0.050 ^f | 2860.7±179.4 ^l | 37838±454.6 ^g | 3050.3±60 ^e |
| SDT | 5.73±0.079 ^{eh} | 11.1±0.13 ^h | 15.2±0.058 ^d | 6444.5±238.6 ^{gh} | 7630.2±351.6 ^l | 894.5±47.1 ^h |
| MT | 6.36±0.051 ^{eh} | 25.5±0.15 ^b | 16.03±0.050 ^{dc} | 10541.1±304.1 ^g | 117342.6±687.2 ^b | 2419.2±45.4 ^f |
| ST | 3.49±0.026 ^{gh} | 27.8±0.20 ^b | 12.7±0.079 ^f | 10969±245.6 ^g | 33712±751.7 ^g | 3791.1±94.9 ^d |
| PSK | 5.08±0.036 ^{eh} | 9.8±0.10 ⁱ | 17.85±0.060 ^c | 16098.6±235.5 ^e | 63150±596.4 ^{cd} | 4227±84.2 ^{cd} |
| MDSK | 7.03±0.052 ^{eh} | 14.9±0.21 ^{ef} | 12.1±0.060 ^f | 5426±147.8 ^h | 4627.3±283.1 ^m | 5086.7±60.9 ^b |
| MDSK | 4.04±0.043 ^{gh} | 14.6±0.21 ^f | 14.32±0.080 ^e | 12653.6±334.1 ^f | 22417.3±424.9 ⁱ | 259.6±38.1 ^k |
| GBK | 4.77±0.062 ^{fh} | 21.1±0.13 ^c | 19.03±0.058 ^{bc} | 4517.5±201.8 ⁱ | 14176.2±417.8 ^j | 4169.06±61.4 ^{cd} |
| GA | 6.25±0.036 ^{eh} | 17.55±0.08 ^d | 15.7±0.072 ^{cd} | 19891.8±309.2 ^d | 17181.3±545 ^j | 2233.4±84.2 ^f |
| PNA | 6.45±0.051 ^{eh} | 14.3±0.20 ^f | 9.1±0.072 ^h | 6430.8±254.4 ^{gh} | 13034.9±534.7 ^j | 1614.4±33.3 ^g |
| SDA | 8.15±0.060 ^{dg} | 12.9±0.14 ^g | 22.07±0.043 ^a | 9544.6±199.4 ^g | 45878±587.3 ^e | 177.4±14.1 ^k |
| SA | 5.68±0.052 ^{eh} | 14.01±0.066 ^f | 13.4±0.052 ^{ef} | 3669.6±127.9 ^j | 29054.6±490.4 ^h | 313.2±33.2 ^k |
| SB | 5.12±0.052 ^{eh} | 16.8±0.12 ^e | 11.2±0.050 ^{fg} | 13865.6±240.4 ^f | 41578.2±1250.7 ^f | 1965.1±47.5 ^{fg} |
| GB | 9.89±0.043 ^{cf} | 23.25±0.14 ^c | 13.9±0.10 ^{ef} | 22631.6±195.3 ^c | 49433±611.6 ^e | 2228.9±45.9 ^f |
| ZB | 6.45±0.045 ^{eh} | 8.8±0.18 ^j | 6.5±0.07 ^k | 1181.1±67.6 ^m | 23019.4±323.4 ⁱ | 1505.4±87.7 ^g |
| PNS | 3.19±0.055 ^h | 10.06±0.12 ^h | 16.9±0.07 ^c | 13063.4±230.5 ^f | 1876±96.2 ⁿ | 5687.03±60.7 ^a |
| AS | 4.418±0.052 ^{fh} | 12.5±0.07 ^g | 15.7±0.06 ^{cd} | 18183.3±265.7 ^d | 65500.6±736.1 ^{cd} | 3643.1±83.4 ^d |
| PKS | 5.42±0.087 ^{eh} | 15.98±0.13 ^e | 8.1±0.050 ⁱ | 21919.3±315.7 ^c | 79562.6±528.6 ^c | 2225.2±41.6 ^f |
| SS | 8.46±0.091 ^{dg} | 15.7±0.06 ^{ef} | 5.9±0.036 ^l | 6905.8±194 ^{gh} | 5115.3±203.3 ^m | 3807.8±43.8 ^d |
| AG | 6.75±0.086 ^{eh} | 17.3±0.13 ^d | 9.85±0.057 ^h | 8349±151.2 ^g | 58178.6±521.3 ^d | 876.6±16.4 ^h |
| SDG | 5.55±0.086 ^{eh} | 6.8±0.10 ^k | 15.1±0.052 ^d | 41057.3±259.2 ^a | 9936±520.7 ^k | 1114.5±31.9 ^h |
| SK | 3.55±0.043 ^{gh} | 10.55±0.08 ^h | 16.7±0.044 ^c | 3560.6±152.6 ^k | 3184.6±174.5 ⁿ | 1201.2±41.7 ^h |
| S | 5.7±0.086 ^{eh} | 13.2±0.11 ^g | 11.4±0.052 ^g | 5367.1±189.2 ^h | 27497.2±648.4 ^h | 4771.5±43.2 ^c |
| MS | 9.43±0.052 ^{cf} | 11±0.13 ^h | 16.9±0.044 ^c | 30837±448 ^{bc} | 31497.3±538.7 ^h | 2006.8±43.2 ^{fg} |
| SSP | 3.86±0.060 ^{gh} | 8.4±0.01 ^j | 18.7±0.057 ^{bc} | 12831.4±252.9 ^f | 29714±546 ^h | 5670.6±47.6 ^a |
| SS | 5.35±0.11 ^{dg} | 9.45±0.10 ⁱ | 19.2±0.050 ^b | 18028.6±286.9 ^d | 82638±692.8 ^c | 1214.4±46.3 ^h |
| Z | 7.96±0.060 ^{dg} | 14.25±0.13 ^f | 10.2±0.057 ^{gh} | 8390.1±115 ^g | 78896.6±404.6 ^c | 1032.6±45.7 ^h |
| DST | 9.6±0.086 ^{cf} | 15.05±0.17 ^{ef} | 12.9±0.050 ^f | 12164±201.2 ^f | 40743.3±587.7 ^f | 1593.06±56.6 ^g |
| GT | 6.99±0.045 ^{eh} | 12.4±0.10 ^g | 18.3±0.065 ^{bc} | 19302.6±196.3 ^d | 69425.3±531.4 ^c | 539.8±32.8 ^j |
| MAT | 13.55±0.043 ^{bd} | 11.5±0.16 ^h | 10.4±0.052 ^{gh} | 20568.4±329.2 ^c | 50219.3±480.7 ^e | 1193.8±61.5 ^h |
| AT | 13.33±0.070 ^{bd} | 14.2±0.07 ^f | 16.9±0.050 ^c | 18313±286.3 ^d | 27611.3±392.9 ^h | 1494.2±54.7 ^h |
| ZT | 8.64±0.069 ^{dg} | 16.2±0.13 ^e | 6.7±0.050 ^k | 7703.7±185.8 ^g | 16315.3±136.1 ^j | 2801.2±60.5 ^d |
| ST | 3.86±0.043 ^{gh} | 15.65±0.10 ^{ef} | 20.8±0.072 ^b | 6356.6±130.4 ^{gh} | 24221.3±462.6 ⁱ | 3197.8±62.8 ^{de} |
| HSY | 5.09±0.026 ^{eh} | 21.2±0.11 ^c | 21.4±0.044 ^b | 13564.6±278.2 ^f | 128162±496.2 ^a | 3261.5±53.8 ^{de} |
| SSSY | 5.44±0.072 ^{eh} | 17.6±0.10 ^d | 11.85±0.080 ^g | 9554.1±272.2 ^g | 8136±340.4 ^l | 1427.1±47.2 ^g |
| PSSY | 12.56±0.052 ^{bd} | 21.3±0.13 ^c | 22.1±0.072 ^a | 7549.8±211.6 ^g | 35440±448.7 ^g | 2075.2±72.5 ^{fg} |
| SSY | 14.27±0.052 ^{bc} | 22.4±0.12 ^c | 14.3±0.072 ^e | 22707.3±277.3 ^c | 41550.6±222.6 ^f | 2044±28.5 ^{fg} |
| MMSY | 4.19±0.043 ^{gh} | 20.5±0.14 ^{cd} | 9.3±0.072 ^h | 8789.6±212.4 ^g | 49646.1±786.9 ^e | 1743.3±53.8 ^g |
| SN | 14.24±0.060 ^{bc} | 17.3±0.20 ^d | 21.6±0.065 ^b | 2565.6±96.3 ^l | 31228±478.4 ^h | 3240±54.9 ^{de} |
| RN | 19.51±0.051 ^a | 16.3±0.14 ^e | 11.3±0.033 ^g | 14225.3±246.8 ^f | 58611.3±503.2 ^d | 2865.4±79.3 ^e |
| AN | 14.13±0.079 ^{bc} | 17.9±0.08 ^d | 17.3±0.058 ^c | 19374±191.2 ^d | 54220.6±453.3 ^d | 1032.5±47.5 ^h |
| KSN | 11.02±0.052 ^{b-c} | 25.03±0.10 ^b | 11.01±0.072 ^{gh} | 36068.3±306.6 ^b | 55663.3±390.7 ^d | 2331.5±34.1 ^f |
| NS | 3.94±0.036 ^{gh} | 29.7±0.12 ^a | 19.9±0.10 ^b | 4543.9±196.4 ⁱ | 44310.5±472.6 ^e | 1979.2±58.9 ^{fg} |
| DS | 3.11±0.060 ^{gh} | 26.9±0.11 ^b | 15.3±0.050 ^d | 16569.3±227 ^e | 55936±493.9 ^d | 5232.8±83.7 ^b |
| MS | 4.7±0.85 ^{fh} | 22.3±0.07 ^c | 14.5±0.050 ^e | 2013.3±132.2 ^l | 48202±401.4 ^e | 712.3±30.6 ⁱ |
| DMS | 4.65±0.051 ^{fh} | 18.8±0.14 ^d | 13.3±0.043 ^{ef} | 639.9±33.8 ^m | 34908.8±613.2 ^g | 3613.2±60 ^d |

Values are means ± SE of three replicates. Different letters indicated significant (P<0.05) differences

2019). The antioxidant ability of pomegranate depends on growing area, cultivar, fruit ripening, and agricultural causes (Cam *et al.*, 2009). Our results showed similarity with Li *et al.* (2006), who proved that pomegranate peel extract had noticeably higher antioxidant ability. Orak *et*

al. (2012) showed that the DPPH scavenging activity of peel ranged from 77.02 to 86.36%, while our results were higher than the values reported. This range was found to be between 12.07 and 12.65% in seed, so our results were lower. The difference in comparison with

Table 4. DPPH radical scavenging ability (%) in different parts of pomegranate cultivars.

| Cultivars | Leaf | Peel | Seed |
|-----------|--------------------------|---------------------------|-------------------------|
| TMT | 857.16±0.43 ^d | 947.3±0.53 ^{bc} | 58.51±0.28 ^e |
| SST | 877.7±2.9 ^c | 937.02±1.07 ^{cd} | 13.99±0.23 ^j |
| SDT | 923.9±1 ^a | 923.4±0.64 ^f | 51.1±0.76 ^{ef} |
| MT | 901.2±1.4 ^{bc} | 945.88±1.8 ^{bc} | 71.5±0.15 ^c |
| ST | 852.8±1.2 ^d | 938.99±0.49 ^{cd} | 89.9±0.21 ^a |
| PSK | 861.1±0.94 ^{cd} | 944.65±0.64 ^{bc} | 64.5±0.19 ^d |
| MDSK | 766.4±1.04 ^f | 913.6±0.85 ^g | 61.5±0.60 ^{de} |
| MDSK | 847.6±1.1 ^d | 928.4±0.85 ^{fd} | 25.4±0.67 ^h |
| GBK | 404.6±5.05 ^k | 933.58±0.85 ^d | 66.2±0.50 ^d |
| GA | 877.38±2.3 ^c | 942.8±0.42 ^{cd} | 83.07±0.46 ^b |
| PNA | 903.25±1.5 ^{bc} | 903.8±0.96 ^h | 71.82±0.29 ^c |
| SDA | 903.8±1.06 ^{bc} | 934.07±1.4 ^d | 74.8±0.26 ^c |
| SA | 877.6±.75 ^c | 927.4±0.61 ^{fd} | 28.68±0.43 ^h |
| SB | 849.7±1.5 ^d | 940.1±1.3 ^c | 44.3±0.20 ^f |
| GB | 915.52±1.9 ^b | 925.83±0.77 ^f | 66.01±0.71 ^d |
| ZB | 915.4±0.52 ^b | 957.9±1.1 ^a | 54.2±0.47 ^e |
| PNS | 768.59±1.9 ^f | 931.86±0.88 ^{cd} | 51.2±0.51 ^{ef} |
| AS | 777.74±1.6 ^f | 902.71±0.86 ^h | 77.5±0.24 ^c |
| PKS | 807.1±2.2 ^e | 952.6±0.74 ^b | 54.1±0.63 ^e |
| SS | 865.7±2.3 ^{cd} | 903.4±1.3 ^h | 34.1±0.42 ^g |
| AG | 864.38±1.8 ^{cd} | 933.4±0.86 ^d | 57.9±0.65 ^e |
| SDG | 477.62±2.1 ^d | 923.4±0.53 ^f | 13.8±0.83 ^j |
| SK | 550.54±2.2 ⁱ | 930.5±1.3 ^{cd} | 49.6±0.76 ^{ef} |
| S | 333.09±4.4 ^l | 946±0.74 ^c | 29.6±0.79 ^h |
| MS | 815.8±1.4 ^e | 928.1±0.53 ^{fd} | 49.8±0.50 ^{ef} |
| SSP | 547.05±1.9 ⁱ | 911.4±1.06 ^g | 66.1±0.28 ^d |
| SS | 462.9±2.9 ^j | 950.06±0.53 ^{bc} | 40.4±0.60 ^f |
| Z | 652.2±2.6 ^h | 934.32±0.42 ^d | 60.8±0.53 ^{de} |
| DST | 736.34±2.08 ^g | 932.35±0.53 ^d | 49.4±0.91 ^{ef} |
| GT | 907.7±1.2 ^{bc} | 950.8±1.1 ^{bc} | 86.9±0.46 ^b |
| MAT | 908.5±1.4 ^{bc} | 951.29±0.21 ^b | 69.6±0.66 ^d |
| AT | 898.3±1.8 ^c | 939.3±0.80 ^{cd} | 51.2±1.05 ^{ef} |
| ZT | 909.5±0.84 ^b | 956.08±0.97 ^a | 43.7±0.84 ^f |
| ST | 800.9±3.5 ^e | 923.7±0.61 ^f | 55.8±0.61 ^e |
| HSY | 857.16±1.8 ^d | 952.3±1.06 ^b | 35.4±0.19 ^g |
| SSSY | 910.7±1.3 ^b | 917.34±0.76 ^g | 27.3±0.83 ^h |
| PSSY | 909.8±3.2 ^b | 952.3±1.18 ^b | 67.4±0.26 ^d |
| SSY | 893.6±1.4 ^c | 924.96±1.62 ^f | 22±0.19 ^h |
| MMSY | 812.03±2 ^e | 943.05±1.09 ^{cd} | 5.5±1.7 ^k |
| SN | 920.9±1.5 ^a | 940.7±1.07 ^{cd} | 16.4±1.2 ⁱ |
| RN | 762.8±1.5 ^f | 923.7±2.2 ^f | 55.7±1.2 ^e |
| AN | 787±0.83 ^f | 916.36±0.74 ^g | 35.7±0.89 ^g |
| KSN | 894.95±2.1 ^c | 911.31±0.88 ^g | 61.1±0.85 ^{de} |
| NS | 905.1±2.2 ^{bc} | 909.9±0.85 ^g | 87.08±0.28 ^b |
| DS | 289.65±3.3 ^m | 938.99±0.96 ^{cd} | 46.8±0.56 ^f |
| MS | 767.3±0.86 ^f | 923.74±1.2 ^f | 79.2±0.26 ^c |
| DMS | 732.9±1.3 ^g | 947.8±0.49 ^{bc} | 39.88±0.65 ^f |

Values are means ± SE of three replicates. Different letters indicated significant (P<0.05) differences

the results of the present study may be the result of other reasons, such as the various pomegranate cultivars and extraction manners used.

Correlation among the studied parameters: The potential correlations among the examined variables in different sections of pomegranate were analyzed based on HCA (Figure 1). This analysis organized the studied

variables into the two main groups, which were then divided into multiple minor groups. In peel, antioxidant activity exhibited positive correlation with total phenol, anthocyanin, and ascorbate, but showed negative correlation with soluble sugar, flavonol, and flavonoid (Figure 1a). These correlations showed that these compounds contributed to the antioxidant activity of

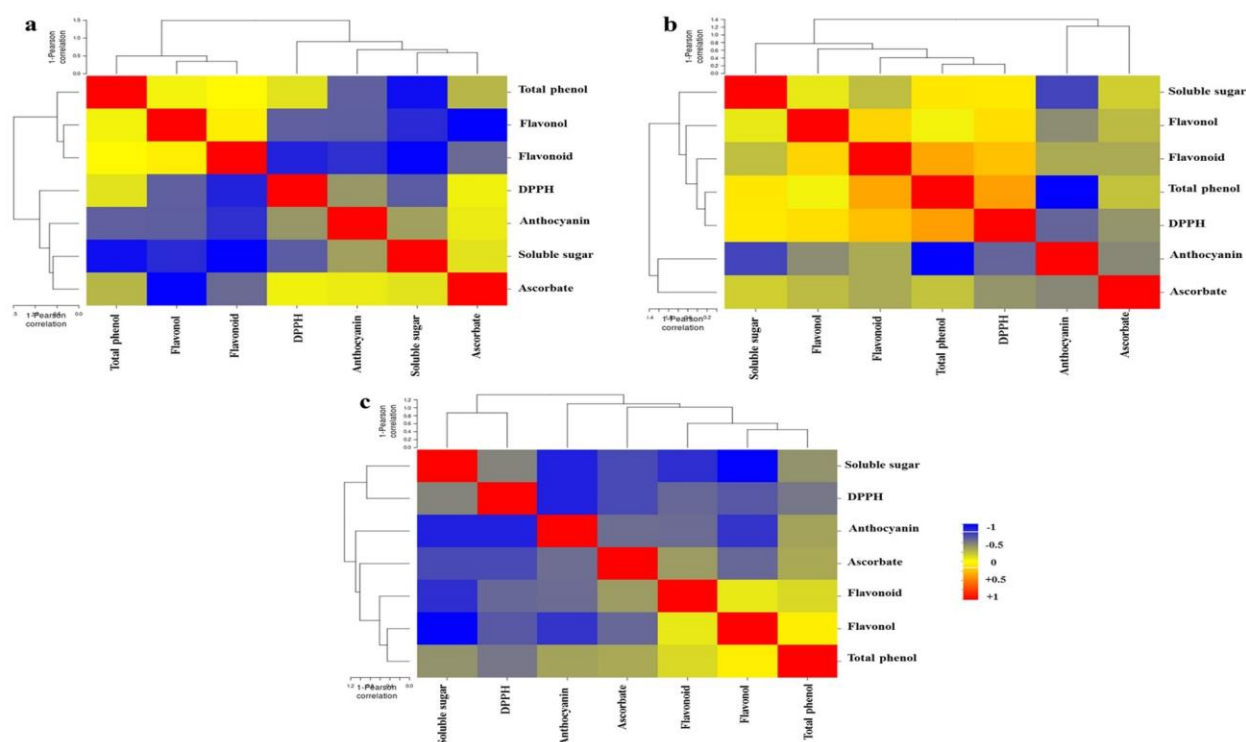


Figure 1. Hierarchical cluster analysis of the examined parameters of peel (a), leaf (b), and seed (c) in different cultivars. Colors show the strength of correlation: Intense red and blue represent strong positive and negative correlations, respectively.

pomegranate peel. The maximum correlation value was observed between antioxidant activity and ascorbate in peel. Total phenol, flavonol, and flavonoid in peel showed positive correlations to each other (Figure 1a). Antioxidant activity in leaves positively correlated with total phenol, flavonoid, soluble sugar, and flavonol, but it revealed a negative correlation with ascorbate and anthocyanin. Also, antioxidant activity with total phenol had the strongest correlation; it can be concluded that antioxidant activity in leaves is much more related to total phenol. Total phenol, flavonol, and flavonoid in leaf had positive correlations to each other, but these showed a negative correlation with anthocyanin (Figure 1b). A positive correlation was observed between antioxidant activity and total phenol, flavonol, flavonoid, soluble sugar, and ascorbate in seed. The highest correlation value was obtained between antioxidant activity and soluble sugar in seed. Anthocyanin in seed had a negative correlation with antioxidant activity (Figure 1c). Same as leaf, anthocyanin was not found as such a significant contributor to antioxidant ability in seed. These findings may be due to the scarcity of anthocyanin in leaf and seed. Also, it is possible that anthocyanins do not play a major function in the antioxidant ability of pomegranate, which would be in agreement with the results by Gil *et al.* (2000). Similarly, Tzulker *et al.* (2007) reported that the DPPH of pomegranate significantly correlated with the polyphenol content, but it did not correlate with anthocyanin content. In other work, total phenol content was not highly correlated

with anthocyanin content, while DPPH was correlated with total phenol content but not with anthocyanin content (Radunic *et al.*, 2015).

Conclusion

In current work, statistically significant alterations were observed in cultivars and, especially, different parts of pomegranate in parameters examined. This indicated that cultivar is the key factor determining the antioxidant properties in pomegranates. Total phenol, flavonol, flavonoid, soluble sugar, ascorbate, and antioxidant ability in peels were higher than in leaves and seeds. So, this investigation supported that pomegranate extracts have powerful antioxidant ability, and the peels are more effective than leaf and seed extracts, such as a good source of natural antioxidants. Our results exhibited that the involvement level of bioactive compounds in seed, leaf, and peel parts of pomegranate differently affected the degree of antioxidant ability. Results of this work can recommend some specific cultivars of pomegranate with upper levels of antioxidant activity and the above-mentioned compounds that are suitable for fresh consumption and health benefits. Therefore, depending on the need, appropriate pomegranate cultivars can be selected for bringing to commercial cultivation.

Compliance with ethical standards conflict of interest

All the authors declare that there is no conflict of interest.

Author contributions

AM has contributed to the major bench experiments.
VN and MHR designed the experiments and supervised

the entire work. MR drafted the manuscript and critically revised the final version.

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