Deficit Irrigation as a Suitable Strategy to Enhance the Nutritional Composition of HydroSOS Almonds

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Received: 6 October 2020; Accepted: 23 November 2020; Published: 27 November 2020

Abstract: The Mediterranean region is one of the most water-scarce areas worldwide and is considered a climate-change hotspot. To assure the viability and competitiveness of irrigated agriculture, it is vital to implement strategies that can maximize water saving without compromising yield. Deficit irrigation (DI) for cultivating drought-tolerant species such as almond (Prunus dulcis (Mill.) D.A. Webb) can help in achieving this goal, while at the same time improving fruit chemical composition. This work evaluated the effect of DI techniques and cultivars on the chemical composition of almonds (cvs. Marta, Guara, and Lauranne) in order to elucidate the most suitable irrigation dose under water-scarcity scenarios. Three irrigation regimes were imposed: a control treatment (FI), which was fully irrigated, receiving 100% of the irrigation requirement (IR), and two sustained-deficit irrigation (SDI) strategies that received 75% (SDI 75) and 65% (SDI 65) of IR. Significant differences among cultivars and irrigation treatments were observed for antioxidant activity and organic acid, sugar, and fatty acid content, which were increased by the SDI strategies. In addition, highly significant correlations were found between leaf-water potential and components such as fumaric acid, sugars, and fatty acids. In terms of the cultivars, cv. Marta showed the highest antioxidant activity, cv. Guara was the richest in organic acids, and cv. Lauranne had the highest fatty acid content. Consequently, SDI strategies improved almond quality parameters related to their nutritional and sensory composition, with significant water savings (reductions of 25–35%) and without important yield loss.

Keywords: water stress; bioactive compounds; HydroSOStainable products; fatty acids; Prunus dulcis

1. Introduction

In the last 10 years, due to climate change, the planet has warmed at a global average of around 1.41 °C [1]. In addition, together with the increased temperature, irregular rainfall and extreme events are the main effects caused by climate change, as described by the European Environment...
Agency [2]. These phenomena not only affect the amount of water available for agriculture but will also cause changes in the growing cycles of plants, affecting the final production. For example, for each degree of increase in global temperature, a 4–6% decrease in crop yields is expected [3]. Thus, considering water-scarcity and climate-change scenarios, the introduction of sustainable irrigation strategies to boost the proper water management for irrigated crops is crucial [4].

In this sense, many Mediterranean fruit crops are well adapted to drought, able to respond positively when water withholding is applied during different phenological stages, such as almond [5], olive [6], and citrus [7], among others. More concretely, almond (Prunus Dulcis (Mill.) D.A. Webb) is considered as a drought-tolerant species [8], with high positive responses under deficit-irrigation (DI) strategies. Many authors have described the almond response to DI, defining its behavior in terms of final yield [9,10], physiology [11], or nut quality [12].

Moreover, almond has high added value because of its nutritional and functional properties, with a composition of macro- and micronutrients that are beneficial to human health [13], with higher amounts of vegetable protein and fat-soluble bioactives. They are also dense in a variety of other nutrients and provide dietary fiber, vitamins, minerals, and many other phytochemicals such as phenolic acids, flavonoids, lignin, hydrolysable tannins, carotenoids, alkaloids, and phytates, among others [14]. In this line, many diseases such as type 2 diabetes, high blood pressure, and neurodegenerative and cardiovascular diseases can be prevented with a healthy diet and one serving of nuts per day [15].

In the last four years, the hydroSOStainable concept has gained great importance in agriculture. This concept arises from society’s dedication to the consumption of products that are sustainable for the environment and also have health benefits [16]. The definition of hydroSOStainable products as fruits and vegetables cultivated under regulated-deficit irrigation (RDI) strategies was first made by Noguera-Artiaga et al. [17]. HydroSOS products are those obtained from plants subjected to DI strategies and are characterized by high amounts of bioactive and functional compounds, among other properties. Taking this concept into consideration, the hydroSOStainable index was created to help farmers develop sustainable practices, ensuring recognition for their products [18]. Since then, hydroSOStainable strategies have been used in many crops, such as olives [19], pistachios [20], and almonds [21]. Although DI strategies slightly affect the final yield (compared with fully irrigated conditions), they improve the quality of the final product, with greater consumer acceptance, and produce environmentally friendly products. According to the scientific literature, previous studies on the effect of DI on almond quality were only focused on cv. Vairo. However, it is well known that each almond cultivar behaves differently in water-deficit conditions. In this line, Lipan et al. [22] evaluated the effect of RDI and overirrigation doses on the nut quality of three almond cultivars, Guara, Marta, and Lauranne. Later, Garcia-Tejero et al. [23] studied the effect of sustained-deficit irrigation (SDI) on the same cultivars, focusing their attention on quality parameters related to the almonds’ marketability and how this type of deficit irrigation strategy gave added value to the final product.

Considering these aspects, the aim of this work was to evaluate the irrigation and cultivar effects on the main chemical components of almonds (antioxidant activity, sugars, organic acids, and fatty acids), defining the most suitable irrigation dose that would ensure improvement of the almond quality in relation to its functional composition.

2. Materials and Methods

2.1. Experimental Design and Irrigation Treatments

The trial was carried out during 2019 in a commercial orchard of almonds (Prunus Dulcis (Mill.) D.A. Webb, cvs. Guara, Marta, and Lauranne) grafted onto GN15 (Garnem®) rootstock, located in the Guadalquivir river basin (SW Spain, 37°30′27.4″ N, 5°55′48.7″ W). The trees, which were 7 years old, were spaced 8 × 6 m² and drip-irrigated using two pipelines with emitters of 2.3 L·h⁻¹. More details about the experimental site can be found in Gutiérrez-Gordillo et al. [10].

Three irrigation treatments were designed: (1) a control treatment that was fully irrigated (FI) and received 100% of irrigation requirement (IR), (2) a sustained-deficit irrigation treatment that
received 75% IR (SDI\textsubscript{75}), and (3) a sustained-deficit irrigation treatment that received 65% IR (SDI\textsubscript{65}). The experimental design was of randomized blocks, with four replications per treatment and cultivar. Each replication consisted of 12 trees (three rows, and four trees per row), and the measurements were taken in the two central trees of each replication (eight trees per cultivar and irrigation treatment).

Irrigation was applied from the middle of March to the end of October, with IR estimated according to the methodology proposed by Allen et al. [24] and reference evapotranspiration (ET\textsubscript{0}) values obtained by using a weather station installed in the same experimental orchard (Davis Advance Pro2, Davis Instruments, Valencia, Spain). The local crop coefficient used ranged from 0.4 to 1.2 according to García-Tejero et al. [25]. Additionally, the IR was reduced for SDI\textsubscript{75} and SDI\textsubscript{65} by multiplying it by 0.75 and 0.65, respectively.

2.2. Plant Measurements

To assess the crops’ physiological response, crop water monitoring was done by means of leaf water potential (\(\Psi_{\text{leaf}}\)) in shaded leaves. These readings were taken between 12:00 and 13:30 p.m., with a 7–15-day frequency during the whole kernel-filling period (from day of year (DOY) 160 to 219). Measurements of \(\Psi_{\text{leaf}}\) were done using a pressure chamber (Soil Moisture Equipment Corp., Santa Barbara, CA, USA), monitoring eight trees per irrigation treatment and cultivar and two leaves on the north side of each tree that were totally mature, fresh, and shaded at approximately 1.5 m height and NW exposed.

At the end of the season, the almond yield was monitored, with harvesting done at 219, 221, and 235 DOY for cvs. Guara, Marta, and Lauranne, respectively. This process was carried out using a mechanical vibrator to throw the almonds to the ground. Collected almonds were processed with mechanical peeling to remove the hulls. Finally, cleaned almonds were left to air-dry and weighed once a humidity content of about 5–6% was reached. About 3 kg of in-shell almonds (per cultivar and irrigation treatment) were sent to Miguel Hernandez University (Orihuela, Alicante, Spain) for quality analysis. More details about harvesting can be found in [10].

2.3. Antioxidant Activity and Total Phenolic Content (TPC)

Extraction was done in 0.5 g of sample sonicated with 10 mL of extractant for 15 min and stored for 24 h at 4\degree C. Then the mixture was sonicated again under the same conditions and subsequently centrifuged at 10,000 rpm for 10 min. Two methods, DPPH\textsuperscript{*} (2,2-diphenyl-1-picrylhydrazyl) and ABTS\textsuperscript{+} (2,2-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)), were used to measure the antioxidant activity of the obtained extracts. More details about the methodology can be found in Lipan et al. [22].

2.4. Organic Acids and Sugars

Organic acids and sugars were determined by high-performance liquid chromatography (HPLC). The extraction consisted of 1 g of ground almond in a Moulinex grinder (AR110830) for 10 s, homogenized with 5 mL of phosphate buffer with a homogenizer (Ultra Turrax T18 Basic) at 11,300 rpm for 2 min, and the tube was maintained in an ice bath after it was centrifuged for 20 min at 15,000 rpm and 4\degree C (total of 3–18 K almond with 5 mL of phosphate buffer), followed by filtration and injection into the HPLC device. Sugar content was determined by using a Supelcogel TM C-610H column (30 cm \(\times\) 7.8 mm) with a precolumn (Supelguard 5 cm \(\times\) 4.6 mm; Supelco, Bellefonte, PA, USA) and detected by a refractive index detector. Organic acid absorbance was measured at 210 nm in the same HPLC condition using a diode-array detector. The analysis was done in triplicate and results were expressed as g kg\(^{-1}\) dry weight.

2.5. Fatty Acid Analysis

Fatty acid methyl esters (FAMEs) were measured using in situ methylation, with some modification [26], and analyzed according to Tuberoso et al. [27]. Ground almond (40 mg) was saponified with 100 \(\mu\)L of dichloromethane (Cl\(_2\)CH\(_2\)), 1 mL of sodium methoxide solution was added,
and it was refluxed for 10 min at 90 °C. Afterwards, 1 mL of BF3 methanolic was added and it was left to rest 30 min for reaction in the dark. Lastly, the FAMEs were extracted from the mixture by using 1.5 mL of hexane. Then, they were separated in a Shimadzu GC17 gas chromatograph coupled with a flame ionization detector and a CP-Sil 88 capillary column. The carrier gas used was He (at a constant pressure of 316 KPa, initial flow of 1.1 mL·min⁻¹), detector gases were H₂ (30 mL·min⁻¹) and air (350 mL·min⁻¹), and makeup gas was He (35 mL·min⁻¹). The injector temperature was 250 °C and the detector was at 260 °C. The preliminary temperature was 175 °C for 10 min, followed by a temperature gradient of 3 °C·min⁻¹ until 215 °C, then maintained at 215 °C for 15 min. The injection volume was 0.6 µL and the split ratio was 1:34. Methylated fatty acid peaks were identified by comparing the retention times of the standards (FAME Supelco MIX-37). Results were expressed quantitatively as g·kg⁻¹, using methyl nonadecanoate as internal standard.

2.6. Statistical Analysis

The Ψ_leaf readings were analyzed by SPSS statistical software (15.0 statistical package; SPSS, Chicago, IL, USA). An exploratory descriptive analysis of the dataset for each treatment and cultivar was done by applying Levene’s test to check the variance homogeneity of the variables. Additionally, with the aim of analyzing the whole dataset, a three-way analysis of variance (ANOVA) for repeated measures was also carried out (two between-subject factors, irrigation treatment and cultivar, and a within-subject factor, timing evolution), followed by Tukey’s multiple range test to compare the differences between treatments, cultivars, and their interactions, considering the variance homogeneity previously analyzed by Levene’s test.

Additionally, two-way ANOVA for Ψ_leaf values was applied to analyze the differences between irrigation treatments, cultivars, and their interactions for each measuring day, applying Tukey’s multiple range test for means separation with a significance level of p < 0.05.

For quality parameters, XLSTAT Premium 2016 (Addinsoft, New York, NY, USA) was used to detect statistically significant differences, with a significance level of p < 0.05. Eight replications per irrigation treatment and cultivar were considered. Two-way ANOVA was done, applying Tukey’s multiple range test for means comparison, taking into consideration the variance homogeneity previously analyzed by Levene’s test.

Yield data were subjected to one-way variance analysis (ANOVA; SPSS 15.0 statistical package, SPSS, Chicago, IL, USA), using Tukey’s test for mean separation (p < 0.05) to compare the differences between irrigation treatments within each cultivar.

Finally, taking into consideration the Ψ_leaf values and quality parameters, principal component analysis (PCA) was conducted with the aim of identifying those variables or underlying factors that would better explain the correlation or covariance matrix of several variables. Linear correlation analysis was done, calculating the Pearson’s correlation coefficient between Ψ_leaf and all quality parameters studied. For this purpose, and with the aim of minimizing the “cultivar effect”, the datasets for each parameter were homogenized according to the methodology proposed by Sterck and Stein [28], focusing attention on improvement or worsening due to water stress in the nutritional parameters of almonds.

3. Results and Discussion

3.1. Climatic Conditions, Physiological Response, and Final Yield

The climatic conditions during the irrigation period were characterized by mild average temperatures (19.8–26.9 °C) and low rainfall. The cumulative ET₀ and rainfall were 840 and 85 mm, respectively. Maximum evapotranspiration rates occurred in June (203 mm), July (239 mm), and August (170 mm), whereas rainfall was concentrated during April (71.2 mm) and October (10 mm). Thus, according to the climatic conditions, the total amount of irrigation received for FI, SDI75, and SDI65 at the end of the season was 7700, 5744, and 5159 m³·ha⁻¹, respectively.
In relation to the crop water status, significant differences in \( \Psi_{\text{leaf}} \) were found among cultivars and irrigation treatments (Table 1), taking into consideration not only all the data but also the daily measurements. In this regard, according to three-way ANOVA for repeated measures, significant differences \((p < 0.01)\) were found for timing evolution, cultivars, and irrigation treatments. Focusing on the cultivars, cv. Marta registered the highest average \( \Psi_{\text{leaf}} \) value \((-1.55 \text{ MPa})\), which was significantly different from cv. Guara \((-1.80 \text{ MPa})\) and cv. Lauranne \((-1.71 \text{ MPa})\), without differences between them. Regarding the irrigation dose, FI registered an average value of \(-1.55 \text{ MPa} \), which was significantly higher than those of SDI\(_{75}\) \((-1.76 \text{ MPa})\) and SDI\(_{65}\) \((-1.71 \text{ MPa})\), and the SDIs were similar. The interactions between irrigation and cultivars showed the highest \( \Psi_{\text{leaf}} \) value for cv. Marta under FI \((-1.45 \text{ MPa})\) and the lowest for cv. Guara under SDI\(_{75}\) \((-1.91 \text{ MPa})\) and SDI\(_{65}\) \((-1.90 \text{ MPa})\). A very similar trend was observed for each measuring day, which reflects that cv. Marta had a different water status than the other cultivars, whereas SDI treatments were similar.

### Table 1. Evolution of crop water status in terms of leaf water potential (\( \Psi_{\text{leaf}} \)).

| DOY  | 162–217 | 162 | 175 | 183 | 189 | 196 | 203 | 210 | 217 |
|------|---------|-----|-----|-----|-----|-----|-----|-----|-----|
| Timing |       |     |     |     |     |     |     |     |     |
| **   |     |     |     |     |     |     |     |     |     |
| Timing × Cultivar |     |     |     |     |     |     |     |     |     |
| **   |     |     |     |     |     |     |     |     |     |
| Cultivar |     |     |     |     |     |     |     |     |     |
| **   | 1.71a | 1.71b | 1.73b | 1.71ab | 1.69b | 1.48b | 1.88a | 1.45b | 1.82ab |
| **   | 1.75a | 1.42a | 1.49a | 1.58a | 1.46a | 1.24a | 1.76a | 1.32a | 1.65a |
| **   | 1.76b | 1.60ab | 1.81b | 1.85b | 1.74b | 1.54b | 1.85a | 1.47b | 1.90b |
| **   | 1.71b | 1.72b | 1.73b | 1.71ab | 1.69b | 1.48b | 1.88a | 1.45b | 1.82ab |
| **   | 1.53a | 1.52a | 1.49a | 1.47a | 1.47a | 1.21a | 1.72a | 1.26a | 1.67a |
| **   | 1.80b | 1.69a | 1.85b | 1.80b | 1.77b | 1.59b | 1.93b | 1.47b | 1.80b |
| **   | 1.71ab | 1.53a | 1.71b | 1.89b | 1.66b | 1.48b | 1.85b | 1.51b | 1.89b |

Note: ns, not significant at \( p < 0.05 \); *, ** significant at \( p < 0.05 \) and 0.01, respectively. † Values followed by the same letter within the same column and factor are not significantly different \( p < 0.05 \) according to Tukey’s multiple range test. DOY, day of year; FI, fully irrigated control treatment; SDI\(_{75}\), sustained-deficit irrigation at 75% of irrigation requirement (IR); SDI\(_{65}\), sustained-deficit irrigation at 65% of IR.

| Irrigation | (MPa) |      |      |      |      |      |      |      |      |
|------------|-------|------|------|------|------|------|------|------|------|
| FI         | -1.55a | -1.42a | -1.49a | -1.58a | -1.46a | -1.24a | -1.76a | -1.32a | -1.65a |
| SDI\(_{75}\) | -1.76b | -1.60ab | -1.81b | -1.85b | -1.74b | -1.54b | -1.85a | -1.47b | -1.90b |
| SDI\(_{65}\) | -1.71b | -1.72b | -1.73b | -1.71ab | -1.69b | -1.48b | -1.88a | -1.45b | -1.82ab |

| Cultivar |     |     |     |     |     |     |     |     |     |
|----------|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Marta    |     |     |     |     |     |     |     |     |     |
| Guara    |     |     |     |     |     |     |     |     |     |
| Lauranne |     |     |     |     |     |     |     |     |     |

| Irrigation × Cultivar |     |     |     |     |     |     |     |     |     |
|-----------------------|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Marta                 |     |     |     |     |     |     |     |     |     |
| Guara                 |     |     |     |     |     |     |     |     |     |
| Lauranne              |     |     |     |     |     |     |     |     |     |

| Note: ns, not significant at \( p < 0.05 \); *, ** significant at \( p < 0.05 \) and 0.01, respectively. † Values followed by the same letter within the same column and factor are not significantly different \( p < 0.05 \) according to Tukey’s multiple range test. DOY, day of year; FI, fully irrigated control treatment; SDI\(_{75}\), sustained-deficit irrigation at 75% of irrigation requirement (IR); SDI\(_{65}\), sustained-deficit irrigation at 65% of IR.
While the almond quality parameters are determined by water stress imposed during the kernel-filling period, final yield responded to the level of water stress produced over the entire period of crop development (including the first stages of vegetative and fruit growth, or even the postharvest conditions from the previous season) [29]. In our case, SDIs were applied, hence, water stress was imposed during the whole season (even during the postharvest period of the previous year, 2018). In response to this strategy, cv. Guara registered the highest yield reduction in SDI$_{65}$ (17% lower than FI), while the yield in SDI$_{75}$ was significantly similar to the yield in FI (Table 2). The yield reductions obtained with cv. Guara were in agreement with the $\Psi_{\text{leaf}}$ values previously registered by this cultivar (Table 1). In contrast, the yield of cvs. Marta and Lauranne was not significantly affected by SDI treatments (Table 2); these cultivars showed the best results in terms of $\Psi_{\text{leaf}}$ (Table 1), as has been previously discussed.

This differential response between cultivars is not new, as it has been proved by other authors. Gomes-Laranjo et al. [11] reported different physiological responses among cultivars when they were subjected to different DI strategies. The authors concluded that cv. Lauranne was less sensitive to water restrictions than Ferragnès. More recently, authors such as Miarnau et al. [30] highlighted that under the SDI strategy, with a total amount of water around 200 mm, the nut yield for cvs. Guara and Marta amounted to 1200 and 1900 kg·ha$^{-1}$, whereas under FI conditions (750 mm) the values were 2800 and 3600 kg·ha$^{-1}$, respectively. All these results suggest that cv. Marta may mitigate the water stress by means of an internal mechanism, yielding more than cv. Guara, as was observed in the present work.

### Table 2. Kernel yield for different cultivars and irrigation treatments.

| Cultivars | FI   | SDI$_{75}$ | SDI$_{65}$ |
|-----------|------|------------|------------|
| Marta     | 2218a| 2209a      | 2243a      |
| Guara     | 2254a| 2081ab     | 1871b      |
| Lauranne  | 2326a| 2105a      | 2196a      |

Note: FI, fully irrigated control treatment; SDI$_{75}$, sustained-deficit irrigation at 75% of irrigation requirement (IR); SDI$_{65}$, sustained-deficit irrigation at 65% of IR. Values followed by the same letter within the same row and factor are not significantly different ($p < 0.05$) by Tukey’s test.

### 3.2. Antioxidant Activity and Total Phenolic Content

The results of antioxidant activity (AA) and total phenolic content (TPC) are shown in Table 3. Highly significant effects ($p < 0.001$) were found in response to the applied irrigation treatments and cultivars.

AA was studied through two methodologies: ABTS$^{•+}$ and DPPH$^{•}$. The ABTS$^{•+}$ results showed significant differences among cultivars but not irrigation treatments, with cv. Marta showing the highest values. Moreover, it was notable that the highest values of ABTS$^{•+}$ were for cv. Guara under SDI$_{65}$, followed by cv. Marta under FI and SDI$_{65}$ strategies. These results evidence the importance of the cultivar in the antioxidant activity of almonds, which can be increased by using DI strategies, as has been observed for cv. Guara under SDI$_{65}$. The DPPH$^{•}$ assay corroborated the results obtained using ABTS$^{•+}$, showing that cv. Marta recorded the highest AA. In terms of irrigation strategies, it was observed that AA increased with the highest level of stress (SDI$_{65}$). Regarding the interaction irrigation × cultivar for DPPH$^{•}$, cv. Marta under SDI$_{65}$ registered the highest values, followed by cv. Marta SDI$_{75}$ and FI. Overall, the obtained results highlight how cultivar and irrigation can positively affect almond’s antioxidant activity.

For TPC, the highest values were also reached with SDI treatments and, in terms of cultivar, Lauranne had the highest value. Regarding the interaction irrigation × cultivar, the highest values were shown for cv. Guara SDI$_{65}$, followed by cvs. Lauranne SDI$_{75}$ and Marta SDI$_{65}$; this trend was consistent with the water-stress values found in this experiment. These findings agreed with Lipan et al. [16], who found a positive correlation between TPC and imposed water stress.
Antioxidant activity and TPC are parameters of great importance for health properties. Ros et al. [31] and Lopez-Uriarte et al. [32] reviewed a total of 21 clinical studies that evaluated almond’s antioxidant activity, reporting numerous cardiovascular health benefits such as decreased blood pressure and visceral adiposity. Polyphenols contribute to almond color and astringency and increase its shelf life [33]. The highest polyphenol concentration is found in almond skin, and many times this is eliminated through the operations of processing [34]. Moreover, almond skin also has antimicrobial properties, which are promoted by synergistic interactions between phenolic acids and flavonoids and involved in fighting against diseases caused by *Salmonella enterica*, *Listeria monocytogenes*, and *Escherichia coli* [35]. The main contributions of almond polyphenols to health are in reducing inflammation and type 2 diabetes [36]; they also have antiproliferative and antitumoral effects, including proanthocyanins, the major polyphenol found in almonds.

**Table 3.** Antioxidant activity (ABTS**••** and DPPH• indices) and total phenolic content (TPC) for different cultivars and irrigation treatments.

|                     | ABTS•• | DPPH• | TPC               |
|---------------------|--------|-------|------------------|
|                     | (mmol Trolox·kg⁻¹) | (g GAE·kg⁻¹) |       |
| **ANOVA †**         |        |       | ***              |
| **Irrigation**      | ns     | ***   | ***              |
| **Cultivar**        | ***    | ***   | ***              |
| **Irrigation × Cultivar** | ***    | ***   | ***              |
| **Tukey Multiple Range Test ‡** |        |       |                  |
| **Irrigation**      |        |       |                  |
| FI                  | 10.8   | 40.2b | 2.97b            |
| SDI75               | 10.0   | 39.8b | 3.81a            |
| SDI65               | 10.9   | 42.0a | 3.80a            |
| **Cultivar**        |        |       |                  |
| Marta               | 11.2a  | 45.0a | 3.40b            |
| Guara               | 10.5ab | 38.8b | 3.50b            |
| Lauranne            | 9.99b  | 38.2b | 3.68a            |
| **Irrigation × Cultivar** |        |       |                  |
| Marta               |        |       |                  |
| FI                  | 12.8ab | 41.7bc| 2.79de           |
| SDI75               | 9.77d  | 45.3ab| 3.44cd           |
| SDI65               | 11.1abc| 48.0a | 3.98bc           |
| Guara               |        |       |                  |
| FI                  | 8.85cd | 37.3ef| 2.29e            |
| SDI75               | 9.53cd | 39.2cde| 3.14cde         |
| SDI65               | 13.1a  | 40.0cde| 5.06a           |
| Lauranne            |        |       |                  |
| FI                  | 10.8bc | 41.6bcd| 3.82c           |
| SDI75               | 10.8bc | 35.0f | 4.86ab           |
| SDI65               | 8.40d  | 37.9def| 2.37e           |

Note: † ns, not significant at p < 0.05; *** significant at p < 0.001. ‡ Values (mean of eight replications) followed by the same letter within the same column and factor are not significantly different (p < 0.05) according to Tukey’s multiple range test. FI, fully irrigated control treatment; SDI75, sustained-deficit irrigation at 75% of irrigation requirement (IR); SDI65, sustained-deficit irrigation at 65% of IR.

The relationship between cultivar and antioxidant activity in almond has been investigated [33], and it was concluded that each cultivar has its own characteristic antioxidants. Considering our results, cv. Marta and Guara had greater amounts of antioxidants than cv. Lauranne, as shown by ABTS••• and DPPH•. The values obtained by ABTS••• and DPPH• may be a result of the activity of tocopherols rather than polyphenols, since cv. Lauranne had the highest TPC index. It is well known that food
antioxidant activity can be given by polyphenols but also by other compounds such as tocopherols, which can increase under water-stress conditions, as previously reported by Zhu et al. [37] and Lipan et al. [34], who demonstrated the importance of DI strategies to increase the antioxidant activity and TPC in raw almonds.

3.3. Organic Acid and Sugar Content

In relation to organic acid results, strongly significant effects were observed between irrigation treatments and cultivars. That is, cv. Guara showed the highest amounts of all studied organic acids, followed by cvs. Lauranne and Marta (Table 4). Thus, as described for antioxidant activity and TPC, the cultivar factor was of great importance. In relation to the imposed irrigation strategy, it is noticeable that SDI$_{75}$ treatment obtained the highest values in all organic acids and, consequently, in total organic acid amount.

Table 4. Organic acids in raw almonds as affected by irrigation dose and cultivar.

| Organic Acids | Oxalic | Citric | Tartaric | Malic | Fumaric | Total |
|---------------|--------|--------|----------|-------|---------|-------|
| Irrigation ANOVA Test† | ns | *** | *** | *** | *** | *** |
| Cultivar | ** | *** | *** | *** | *** | *** |
| Irrigation × Cultivar | *** | *** | *** | *** | *** | *** |

Tukey’s Multiple Range Test‡

| Irrigation (g·kg$^{-1}$) | FI | SDI$_{75}$ | SDI$_{65}$ |
|--------------------------|----|------------|------------|
| Marta | 2.04a | 2.98b | 2.05b |
| Guara | 2.14b | 3.14a | 2.20a |
| Lauranne | 1.88b | 3.05b | 2.08b |

| Cultivar | FI | SDI$_{75}$ | SDI$_{65}$ |
|----------|----|------------|------------|
| Marta | 2.00abc | 2.43c | 1.23c |
| Guara | 2.13abc | 3.63a | 2.98a |
| Lauranne | 1.88b | 3.05b | 1.87b |

| Irrigation × Cultivar | Marta | Guara | Lauranne |
|----------------------|-------|-------|----------|
| FI | 2.04abc | 2.36e | 2.53de |
| SDI$_{75}$ | 1.99abc | 2.00a | 2.37a |
| SDI$_{65}$ | 2.13abc | 2.61b | 1.89b |

Note: † ns, not significant at $p < 0.05$; **, *** significant at $p < 0.01$ and 0.001, respectively. ‡ Values (mean of eight replications) followed by the same letter within the same column and factor are not significantly different ($p < 0.05$) according to Tukey’s multiple range test. FI, fully irrigated control treatment; SDI$_{75}$, sustained-deficit irrigation at 75% of irrigation requirement (IR); SDI$_{65}$, sustained-deficit irrigation at 65% of IR.

Regarding the interaction irrigation × cultivar, it can be highlighted that all cultivars were positively influenced by SDI$_{75}$. The interaction of cv. Guara and SDI$_{75}$ treatment registered the highest amount of organic acids except fumaric acid; cv. Lauranne × SDI$_{75}$ had the highest value of fumaric acid. Similar results were reported by Lipan et al. [16] for cv. Vairo, which showed a clear relationship...
between water stress and total organic acid content, although other authors concluded that water stress did not have such effects on these compounds [12,38]. Comparing these results with those obtained for $\Psi_{\text{leaf}}$, it is noticeable that the best results in relation to organic acid content were found in the cultivar that registered the highest water-stress values, which is in line with the results of Lipan et al. [22], who concluded there was a direct relationship between imposed water stress and organic acid content for these cultivars (Guara, Marta, and Lauranne).

Sugar content was also highly influenced by the cultivar and the irrigation strategy (Table 5). All studied sugars showed a clear response to the irrigation treatments and cultivars, except for maltotriose. In terms of irrigation treatment, $\text{SDI}_{75}$ and $\text{SDI}_{65}$ showed the highest total sugar content of 62.9 and 62.2 g·kg$^{-1}$, respectively. In terms of cultivar, Guara (65.5 g·kg$^{-1}$) and Lauranne (63.7 g·kg$^{-1}$) had the highest amount of sugars. Total sugars, and specifically sucrose, tend to be higher as water stress increases, which is in line with other studies [39] that observed that total sugars and sucrose increased with water stress. Relating to the interaction of irrigation $\times$ cultivar, it can be highlighted that all cultivars were positively influenced by DI, especially by $\text{SDI}_{75}$, as occurs with organic acids. For maltotriose and sucrose, the best combination was observed in cv. Guara under $\text{SDI}_{75}$, for glucose, the best combination irrigation $\times$ cultivar was in cv. Marta $\text{SDI}_{65}$, and for fructose, cv. Lauranne under the $\text{SDI}_{65}$ strategy reached the highest value. According to Sánchez-Bel et al. [38], sucrose is the principal sugar in almond cultivars, due to its preferential production and accumulation in the almond during ripening, and probably its synthesis and accumulation would be influenced by water stress. In this line, there is a strong effect between water stress and the sugar composition of nuts. During the water-stress period, almond leaves begin to lose turgor due to dehydration and, to avoid this, the tree closes the stoma by decreasing the transpiration process. Therefore, to restore the osmotic balance, the tree concentrates sugars to recover the turgor that was lost by dehydration. This is in line with Prigomet et al. [40], who found in a two-year experiment that non-irrigated trees accumulated more leaf water soluble sugars than control trees to maintain cell turgor, similar to what occurred in almonds, as previously discussed [16,21,22].

3.4. Fatty Acids

Almond’s fatty acid profile has a broad spectrum, which makes this product a food that is a good energy source and does not increase cholesterol levels [41]. A total of 25 fatty acids were identified in this experimental work, classified as saturated fatty acids (SFA) (Table 6) and unsaturated fatty acids (Table 7), which are subdivided into monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA). The almond lipid fraction identified was mainly composed of oleic acid (C18:1n9), linoleic acid (C18:2n6), palmitic acid (C16:0), and stearic acid (C18:0). These were significantly affected by water stress, which increased their concentration in both SDI treatments compared to the FI treatment. In particular, oleic and linoleic acids increased 6.0% and 10%, respectively, in SDI treatments.

Regarding the cultivar, Guara had the highest amount of oleic acid, while cv. Lauranne had the highest amount of linoleic acid (6.2% and 3.8% more than Marta and Guara, respectively).

The interaction irrigation $\times$ cultivar showed that cv. Guara in $\text{SDI}_{75}$ and cv. Lauranne in $\text{SDI}_{65}$ led to the highest content of palmitic acid; whereas cv. Marta in the three treatments and cv. Guara in $\text{SDI}_{65}$ and cv. Lauranne in both SDI treatments showed the highest amounts of oleic acid.

The best results in terms of the oleic/linoleic (O:L) ratio were found for $\text{SDI}_{75}$ (this index improves as its value decreases), while in terms of the cultivar, the lowest values were obtained for cvs. Guara and Lauranne. Linoleic acid is related to the stability of the oil. Thus, the lower the amount, the greater the stability [42]. This fatty acid is essential for humans and cannot be synthesized by itself. Thus, although stability is reduced, the increase in linoleic acid by deficit-irrigation treatment gives the almond added value since this acid plays a fundamental role in the death of cardiac cells [39]. Additionally, as water stress increases, total phenolic content and antioxidant activity increase, which could help in protecting against lipid oxidation.
Regarding the total monounsaturated fatty acid (MUFA) and polyunsaturated fatty acid (PUFA) content (Table 8), these values were higher in both SDI treatments; cv. Marta had a higher amount of MUFA and cv. Lauranne of PUFA. These results agree with those in the study of Lipan et al. [22], which found the same relation of these cultivars under different irrigation strategies. In contrast, in other cultivars, such as cv. Vairo, decreased MUFAs and increased PUFAs with increasing water stress after three years of experimentation was highlighted by Lipan et al. [39]. In other nuts, such as pistachio, these compounds were not affected by water stress [43]. However, significant differences were found with values higher than 50% of MUFA and 30% of PUFA depending upon the pistachio cultivar used. In olive cv. Arbequina, Garcia et al. [44] reported an increase in the MUFA/PUFA ratio in the RDI treatment compared to the control treatment due to the desaturation of oleic acid with high stress and consequent linoleic formation. Bitok and Sabaté [45] reported that products rich in MUFA and PUFA could contribute to the prevention of coronary heart and cardiovascular diseases as well as diabetes and obesity. To prevent these diseases, the US Food and Drug Administration recommends 42.5 g of almond [46]. This provides greater functionality for almonds cultivated under water-stress conditions. The PUFA:SFA ratio provides information about whether a diet is atherogenic or could promote coronary heart disease. In this case, this ratio had a high relation with irrigation, cultivar, and irrigation × cultivar, and was higher in SDI75 with cv. Lauranne.

Table 5. Sugar content in raw almonds as affected by irrigation dose and cultivar.

| Sugars       | Maltoheptaose | Maltotriose | Sucrose | Glucose | Fructose | Total |
|--------------|---------------|-------------|---------|---------|----------|-------|
| **ANOVA Test‡**          |               |         |         |         |          |       |
| Irrigation     | ns            | ***      | ***     | ***     | ***      | ***   |
| Cultivar       | ns            | ***      | ***     | ***     | ***      | ***   |
| Irrigation × Cultivar | ns           | ***      | ***     | ***     | ***      | ***   |

| **Tukey’s Multiple Range Test †**          |               |         |         |         |          |       |
| Irrigation (g·kg⁻¹)                        |               |         |         |         |          |       |
| FI            | 3.34          | 3.20ab   | 33.5b   | 9.78b   | 2.80b    | 52.6b |
| SDI75         | 3.45          | 3.56a    | 41.2a   | 10.9a   | 3.81a    | 62.9a |
| SDI65         | 3.35          | 3.08b    | 40.5a   | 11.2a   | 4.06a    | 62.2a |

| Cultivar      |               |         |         |         |          |       |
| Marta         | 3.38ab        | 3.39b    | 26.5b   | 12.5a   | 2.85c    | 48.5b |
| Guara         | 3.56a         | 4.45a    | 44.3a   | 9.66b   | 3.55b    | 65.5a |
| Lauranne      | 3.20b         | 2.00c    | 44.4a   | 9.79b   | 4.27a    | 63.7a |

| Irrigation × Cultivar |               |         |         |         |          |       |
| Marta            |               |         |         |         |          |       |
| FI                | 3.40          | 3.27c    | 25.2e   | 10.0bcde| 2.52c    | 44.4f |
| SDI75            | 3.50          | 3.38bc   | 26.6e   | 11.4b   | 2.78c    | 47.6ef|
| SDI65            | 3.24          | 3.51bc   | 27.6e   | 15.9a   | 3.24bc   | 53.6d |
| Guara            |               |         |         |         |          |       |
| FI                | 3.31          | 3.57bc   | 33.4d   | 9.46cde | 2.35c    | 52.1de|
| SDI75            | 3.76          | 5.60a    | 52.0a   | 10.5bcd | 4.21ab   | 76.1a |
| SDI65            | 3.60          | 4.19b    | 47.5b   | 9.05de  | 4.09ab   | 68.4b |
| Lauranne         |               |         |         |         |          |       |
| FI                | 3.30          | 2.74c    | 41.8c   | 9.87bcd | 3.52bc   | 61.3c |
| SDI75            | 3.10          | 1.69d    | 44.9b   | 10.9bc  | 4.44ab   | 65.1bc|
| SDI65            | 3.22          | 1.56d    | 46.4b   | 8.62e   | 4.86a    | 64.7bc|

Note: † ns, not significant at p < 0.05; *** significant at p < 0.001. ‡ Values (mean of 8 replications) followed by the same letter within the same column and factor are not significantly different (p < 0.05) according to Tukey’s multiple range test. FI, fully irrigated control treatment; SDI75, sustained-deficit irrigation at 75% of irrigation requirement (IR); SDI65, sustained-deficit irrigation at 65% of IR.
Table 6. Saturated fatty acid (SFA) profile in raw almonds affected by cultivar and irrigation dose.

|                | C14:0 (Myrist) | C15:0 (Pentadecylic) | IsoC16:0 | C16:0 (Palmitic) | C17:0 (Margaric) | C18:0 (Stearic) | C20:0 (Arachidic) | C21:0 (Heneicosylic) | C22:0 (Behenic) | C23:0 (Tricosylic) |
|----------------|----------------|----------------------|----------|-----------------|-----------------|----------------|------------------|---------------------|------------------|-------------------|
| **ANOVA Test†**|                |                      |          |                 |                 |                |                  |                     |                  |                   |
| Irrigation     | ***            | *                    | *        | ***             | ***             | ***            | *                | ***                 | ***              | ns                |
| Cultivar       | ***            | *                    | ns       | ***             | ***             | ***            | *                | ***                 | ***              | ***               |
| Irrigation × Cultivar | ***            | *                    | *        | ***             | ***             | ***            | *                | ***                 | ***              | ***               |
| **Tukey Multiple Range Test‡** |                |                      |          |                 |                 |                |                  |                     |                  |                   |
| **Irrigation** |                |                      |          |                 |                 |                |                  |                     |                  |                   |
| FI             | 0.11b          | 0.05b                | 0.04b    | 21.3b           | 0.30b           | 9.92b          | 0.56ab           | 0.09a               | 0.14a            | 0.19              |
| SDI75          | 0.13a          | 0.06a                | 0.05a    | 23.4a           | 0.36a           | 11.0a          | 0.63a            | 0.09a               | 0.14a            | 0.19              |
| SDI65          | 0.11b          | 0.05b                | 0.04b    | 23.0a           | 0.28b           | 10.7ab         | 0.53b            | 0.08b               | 0.13b            | 0.19              |
| **Cultivars**  |                |                      |          |                 |                 |                |                  |                     |                  |                   |
| Guara          | 0.12a          | 0.05b                | 0.04     | 21.8b           | 0.37a           | 9.92b          | 0.57b            | 0.09a               | 0.13b            | 0.25a             |
| Marta          | 0.11b          | 0.06a                | 0.04     | 22.5ab          | 0.25b           | 12.9a          | 0.67a            | 0.08b               | 0.16a            | 0.18b             |
| Lauranne       | 0.12a          | 0.06a                | 0.04     | 23.6a           | 0.33a           | 8.70c          | 0.47c            | 0.09a               | 0.12b            | 0.15c             |
| **Irrigation × Cultivar** |            |                      |          |                 |                 |                |                  |                     |                  |                   |
| FI             |                |                      |          |                 |                 |                |                  |                     |                  |                   |
| Marta          | 0.12ab         | 0.05ab               | 0.04b    | 21.6bc          | 0.34c           | 9.30de         | 0.59b            | 0.09ab              | 0.12bc           | 0.25a             |
| Guara          | 0.10c          | 0.05b                | 0.04b    | 20.8c           | 0.25b           | 11.8bc         | 0.63ab           | 0.08ab              | 0.17a            | 0.17b             |
| Lauranne       | 0.11bc         | 0.06ab               | 0.04b    | 24.1ab          | 0.26b           | 13.2ab         | 0.57b            | 0.08ab              | 0.14abc          | 0.18b             |
| **SDI75**      |                |                      |          |                 |                 |                |                  |                     |                  |                   |
| FI             | 0.12ab         | 0.05ab               | 0.04b    | 21.6bc          | 0.34c           | 9.30de         | 0.59b            | 0.09ab              | 0.12bc           | 0.25a             |
| Marta          | 0.12ab         | 0.05ab               | 0.04b    | 21.8bc          | 0.35a           | 9.90de         | 0.59b            | 0.09ab              | 0.12bc           | 0.25a             |
| Guara          | 0.12ab         | 0.05ab               | 0.04b    | 21.9bc          | 0.25b           | 10.6cd         | 0.59b            | 0.10ab              | 0.14abc          | 0.26a             |
| Lauranne       | 0.11bc         | 0.06ab               | 0.04b    | 24.1ab          | 0.26b           | 13.2ab         | 0.57b            | 0.08ab              | 0.14abc          | 0.18b             |
| **SDI65**      |                |                      |          |                 |                 |                |                  |                     |                  |                   |
| FI             | 0.12ab         | 0.05ab               | 0.04b    | 21.6bc          | 0.34c           | 9.30de         | 0.59b            | 0.09ab              | 0.12bc           | 0.25a             |
| Marta          | 0.12ab         | 0.05ab               | 0.04b    | 21.8bc          | 0.35a           | 9.90de         | 0.59b            | 0.09ab              | 0.12bc           | 0.25a             |
| Guara          | 0.12ab         | 0.05ab               | 0.04b    | 21.9bc          | 0.25b           | 10.6cd         | 0.59b            | 0.10ab              | 0.14abc          | 0.26a             |
| Lauranne       | 0.11bc         | 0.06ab               | 0.04b    | 24.1ab          | 0.26b           | 13.2ab         | 0.57b            | 0.08ab              | 0.14abc          | 0.18b             |

Note: † ns, not significant at $p < 0.05$; * and *** significant at $p < 0.05$ and 0.001, respectively. ‡ Values (mean of three replications) followed by the same letter within the same column and factor are not significantly different ($p < 0.05$), according to Tukey’s least significant difference test. FI, fully irrigated control treatment; SDI75, sustained-deficit irrigation at 75% of irrigation requirement (IR); SDI65, sustained-deficit irrigation at 65% of IR.
### Table 7. Monounsaturated and polyunsaturated fatty acid (MUFA and PUFA) profile in raw almonds affected by cultivar and irrigation dose.

| MUFA | C15:1 (Pentadecenoic) | C16:1c7 (Palmitoleic) | C16:1c10 (Eicosenoic) | C17:1 (cis-Heptadecenoic) | C18:1t9 (Elaidic) | C18:1c9 (Palmitoleic) | C18:1c11 (Eicosenoic) | C20:1c9 (cis-Vaccenic) | C20:1c11 (Eicosenoic) | C20:1c15 (Nervonic) | C24:1 (Linoleadic) | C18:2t8c13 (Linoleaidic) | C18:2n6c9,12 (Linoleic) | C18:3n3 (α-Linolenic) |
|------|----------------------|----------------------|----------------------|-------------------------|------------------|----------------------|----------------------|----------------------|----------------------|----------------------|---------------------|----------------------|----------------------|---------------------|
| ANOVA Test † | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** |
| Tukey Multiple Range Test ‡ | | | | | | | | | | | | | | | |
| Irrigation | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** |
| Cultivar | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** |
| I × C | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** | ns *** *** *** |

**Note:** †, ns, not significant at \( p < 0.05; *, **, ***, and **** significant at \( p < 0.05, 0.01, \) and 0.001, respectively. ‡ Values (mean of three replication) followed by the same letter, within the same column and factor, were not significantly different \( p < 0.05 \), according to Tukey's least significant difference test. I, irrigation; C, cultivar; FI, fully irrigated control treatment; SDI75, sustained-deficit irrigation at 75% of irrigation requirement (IR); SDI65, sustained-deficit irrigation at 65% of IR.
Table 8. Types of fatty acids, ratios, and health indices in different almond cultivars under sustained deficit irrigation.

|                        | O:L | SFA | MUFA | PUFA | PUFA:SFA | PUFA:MUFA | (MUFA + PUFA)/SFA | Total fatty acids |
|------------------------|-----|-----|------|------|----------|------------|-------------------|------------------|
| **ANOVA Test †**       |     |     |      |      |          |            |                   |                  |
| Irrigation             | *** | *** | **   | *    | ***      | ***        | ***               | *                |
| Cultivar               | *** | *** | **   | *    | ***      | ***        | ***               | *                |
| Irrigation × Cultivar  | *** | *** | **   | *    | ***      | ***        | ***               | *                |

**Tukey Multiple Range Test ‡**

|                      | Irrigation (g·kg⁻¹ dry weight) |                      |                      |                      |                      |                      |                  |
|----------------------|--------------------------------|----------------------|----------------------|----------------------|----------------------|----------------------|------------------|
| FI                   | 2.73a 32.9b 158b 51.2b 1.56a 0.32b 6.38a 242b | SDI75 2.51b 36.0a 160ab 56.8a 1.57a 0.35a 6.05b 253a | SDI65 2.79a 35.0a 166a 53.2ab 1.52b 0.32b 6.30a 254a |                      |                      |                      |                  |

|                      | Cultivar |                      |                      |                      |                      |                      |                  |
|----------------------|----------|----------------------|----------------------|----------------------|----------------------|----------------------|------------------|
| Marta                | 2.88a 33.3b 169a 52.1b 1.56b 0.31b 6.63a 254a |                      |                      |                      |                      |                      |                  |
| Guara                | 2.59b 36.9a 154b 53.5b 1.45c 0.35a 5.64b 245b |                      |                      |                      |                      |                      |                  |
| Lauranne             | 2.57b 33.7b 162ab 55.6a 1.65a 0.34a 6.47a 251a |                      |                      |                      |                      |                      |                  |

|                      | Irrigation × Cultivar |                      |                      |                      |                      |                      |                  |
|----------------------|----------------------|----------------------|----------------------|----------------------|----------------------|----------------------|------------------|
| Marta                |                      |                      |                      |                      |                      |                      |                  |
| FI                   | 2.82ab 32.5c 164ab 51.9ab 1.59ab 0.31cd 6.65ab 249b | SDI75 2.80ab 33.4bc 165ab 52.5ab 1.57abc 0.32d 6.51ab 251ab | SDI65 3.02a 34.0bc 177a 51.9ab 1.53abc 0.29d 6.72a 262a |                      |                      |                      |                  |
| Guara                |                      |                      |                      |                      |                      |                      |                  |
| FI                   | 2.81ab 34.0bc 154b 48.8b 1.44bc 0.32cd 5.97d 237c | SDI75 2.32d 38.1a 150b 57.6ab 1.51abc 0.38a 5.44e 245b | SDI65 2.63bc 38.7a 159ab 54.2ab 1.40c 0.34bc 5.50de 251ab |                      |                      |                      |                  |
| Lauranne             |                      |                      |                      |                      |                      |                      |                  |
| FI                   | 2.57bcd 32.0c 156b 53.0ab 1.65a 0.34bc 6.51ab 241b | SDI75 2.41cd 36.5ab 166ab 60.3a 1.65a 0.36ab 6.21bc 263a | SDI65 2.71b 32.4c 163ab 53.6ab 1.65a 0.3c 6.68ab 249b |                      |                      |                      |                  |

Note: † *, **, and *** significant at p < 0.05, 0.01, and 0.001, respectively. ‡ Values (mean of three replications) followed by the same letter within the same column and factor were not significantly different (p > 0.05), according to Tukey’s least significant difference test. FI, fully irrigated control treatment; SDI75, sustained-deficit irrigation at 75% of irrigation requirement (IR); SDI65, sustained-deficit irrigation at 65% of IR.
3.5. Relationship between Crop Water Status at Different Stages and Nutritional Composition of Almond

With the aim of finding the potential relationships between $\Psi_{\text{leaf}}$ and quality parameters and estimate which of them were more affected by imposed water stress, PCA was done (Figure 1).

![Figure 1](image.png)

**Figure 1.** Principal component analysis (PCA) score biplot showing relationships among leaf water potential (LWP) and quality parameters. Black dots indicate samples. O/L, oleic/linoleic ratio; MUFA, monounsaturated fatty acid; PUFA, polyunsaturated fatty acid; SFA, saturated fatty acid; Total_FA, total fatty acids; TPC, total phenolic content.

According to the obtained results, the two main components (F1 and F2) explained 41.9% and 21.4% of total variance. As observed, samples of each irrigation treatment were grouped mainly separately, except for SDI$_{75}$ for cvs. Lauranne and Marta grouped together in FI treatments, and totally opposite to SDI$_{65}$ treatments. In this regard, SDI$_{65}$ samples were mainly surrounded by sucrose, glucose, fructose, SFA, and total fatty acids. In addition, cv. Guara under SDI$_{75}$ was characterized by total organic acids (especially malic, citric, and tartaric), maltoheptaose, maltotriose, and the PUFA/MUFA ratio. Finally, FI samples were characterized by $\Psi_{\text{leaf}}$ and the (MUFA + PUFA/SFA) ratio.

Significant relationships were observed between $\Psi_{\text{leaf}}$ values and the quality parameters that had previously shown significant differences between irrigation treatments. No significant correlations were obtained between $\Psi_{\text{leaf}}$ and antioxidant activity (in terms of ABTS$^{\bullet+}$ and DPPH$^{\bullet-}$ index with $r = -0.10$ and 0.08, respectively). However, a significant correlation was observed for TPC ($r = -0.59$). On the contrary, Lipan et al. [39] found a significant relationship between accumulated water stress and the ABTS$^{\bullet+}$ index ($r = 0.79$), which was not detected in the present study, probably because this experiment was developed with different cultivars.

More interesting were the Pearson’s coefficients among $\Psi_{\text{leaf}}$, organic acids, and sugar content. Within the organic acids, fumaric showed the best correlations ($r = -0.96^{**}$), whereas for the sugar profile, the best relationships were found for sucrose ($r = -0.85^{**}$), fructose ($r = -0.92^{**}$), and total sugars ($r = -0.86^{**}$). Similar results were reported by Lipan et al. [39], who noted improvements in terms of sucrose ($r = -0.42^{**}$), fructose ($r = -0.36^{*}$), and total sugar content ($r = -0.39^{**}$). Thus, according to these findings, high water-stress levels (lower values of $\Psi_{\text{leaf}}$) would be accompanied by higher sugar and fumaric acid content.

Finally, the fatty acid content increased directly with the imposed water stress, with SFA ($r = -0.88^{**}$), PUFA ($r = -0.80^{*}$), and total fatty acid content ($r = -0.84^{**}$) showing the highest
correlation with $\Psi_{\text{leaf}}$, whereas for MUFA these relationships were not significant ($r = -0.51$). In line with these results, the most important variations in the fatty acid composition was observed for the oleic/linoleic ($r = 0.46$) and (MUFA+PUFA)/SFA ($r = 0.71$ *) ratios. These results are in agreement with those reported by Lipan et al. [39], who found strong relationships between $\Psi_{\text{leaf}}$ values and the oleic/linoleic ratio ($r = -0.82 **$), SFA ($r = 0.81 ***$), MUFA ($r = -0.84 ***$), PUFA ($r = 0.83 ***$), and PUFA/MUFA ($r = 0.85 ***$).

Even though the present work corresponds to a single year, the relationships found between the studied quality parameters and imposed water stress are in accordance with similar works developed over a single year or for long-term experiments. As already mentioned, authors such as Egea et al. [12], in a two-year experiment with cv. Marta, concluded that DI strategies did not affect the final content of sugars and organic acids. Moreover, Lipan et al. [39], in a long-term experiment on cv. Vairo, found significant relationships between quality parameters and imposed water stress. Those authors reported significant increases in fructose, sucrose, and total sugars when water stress was imposed, and similarly for antioxidant activity, SFA, and PUFA and decreased O/L rate, all in accordance with the results obtained in the present work. In addition, Lipan et al. [22] reported significant improvements in organic acid and sugar content in the same cultivars considered in this work but subjected to RDI strategies, along with reductions in the oleic/linoleic ratio and increases in the PUFA, PUFA/SFA, and PUFA/MUFA ratios. Thus, despite presenting results for a single year, there is a clear pattern between the obtained results and other recently published results, reinforcing the hypothesis that DI strategies improve some of the most relevant quality parameters related to the nutritional composition of raw almonds.

4. Conclusions

In view of the obtained results, almond cultivars that have been subjected to SDI strategies have improved fruit quality parameters, so these can be suitable strategies to enhance the nutritional composition of almonds. Specifically, SDI50 treatment improved the content of fatty acids and TPC, while SDI75 improved sugars and organic acids. These findings also demonstrate that these parameters were strongly affected by the almond cv. In addition, organic acids, sugars, and fatty acids were the most affected parameters, showing strong correlations with imposed water stress. These results open a new line of research in which it may be possible to evaluate how the quality parameters evolve during the kernel-filling period, even to define different threshold values of $\Psi_{\text{leaf}}$ to increase specific components, and with different objectives depending on the cultivar factor. Furthermore, SDI strategies not only would improve the almond quality, with concurrent reductions in water consumption, but also give added value to the final product, within the concept of hydroSOS products.

Encouragingly, there are advances in almond production and water productivity through a combination of SDI techniques, as shown by the findings described in this study, making it possible to implement deficit irrigation practices as a tool to achieve environmental, health, and economic benefits.

**Author Contributions:** Writing: S.G.-G. and I.F.G.-T.; Conception or design: S.G.-G., I.F.G.-T., and L.L. Acquisition, analysis, and interpretation of data: S.G.-G., I.F.G.-T., and L.L.; Critical revision of the manuscript for important intellectual content: A.A.C.-B., V.H.D.Z., E.S., F.H., and M.S.H.-Z.; Statistical analysis: S.G.-G. and L.L.; Reagents/materials/analysis tools’ contribution: S.G.-G., L.L., F.H., E.S., and A.A.C.-B. All authors have read and agreed to the published version of the manuscript.

**Funding:** S.G.-G. has a contract co-financed by the National Institute of Agrarian and Food Research and Technology (FPI-INIA 2016) and European Social Fund (ESF) “The European Social Fund invests in your future”. This work has been partially sponsored by the research project “Impact of climate change and adaptation measures (INNOVA-Climate)” (AVA.AVA2019.051), both co-financed by the European Regional Development Fund (ERDF) within the Operational Program Andalusia 2014–2020 “Andalucia is moving with Europe” and the Spanish Ministry of Economy, Industry and Competitiveness, through the research project (hydroSOS mark) including the Universidad Miguel Hernández de Elche (AGL2016-75794-C4-1-R, hydroSOS foods) and the Universidad de Sevilla (AGL2016-75794-C4-4-485 R).

**Conflicts of Interest:** The authors declare no conflict of interest.
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