Changes in Ripening-Related Quality Traits of Long Shelf Life Tomatoes as Influenced by Water Deficit and Short-Term Postharvest Storage

Joan Casals 1,2,*, Aurora Rull 1,2 and Jordi Giné-Bordonaba 3

Abstract: The diversity preserved within the European long shelf life tomato landraces (LSL) is a unique source to design high quality tomato products better adapted to changing environmental conditions and, thereby, to reduce food losses. The adaptation of LSL to water deficit (WD) management practices and their postharvest keeping ability can be used as tools to concomitantly enhance fruit quality and sustainable production. In this study, we investigated the effect of WD conditions and the plant growing environment (open field vs. tunnel) on quality traits of two genotypes of the Penjar LSL variety (modern hybrid (MV) and landrace (LR)). Changes in ripening-related quality traits (fruit ethylene production, respiration rate, firmness, color, soluble solids content, titratable acidity and the content of antioxidants, as well as specific sugars and acids) in response to the different preharvest factors were evaluated at the time of harvest and after a short period of storage (30 days), following actual commercial practices. Significant differences among genotypes were encountered for most quality traits at the time of harvest and higher intra- and inter-environment heterogeneity was observed in the LR than in the MV genotype. In general, Penjar tomatoes exhibit a low physiological activity (ethylene production, 0.56–1.33 μL kg⁻¹ h⁻¹, respiration rate: 0.015–0.026 mg CO₂ kg⁻¹ h⁻¹) at harvest. In both genotypes, WD increased to a different extent the fruit external color (redness, lightness) as well as the sensory (SSC) and nutritional (antioxidant capacity) fruit profiles. By contrast, the growing environment had little impact on most fruit quality traits. Postharvest storage only led to a slight reduction in the fruit respiration and ethylene production, lower sugars and acids content, enhanced color and no firmness changes. Overall, the results from this study demonstrate that selecting the appropriate genotypes is the most important step towards the design of high-quality LSL tomatoes, while WD and short-term storage can be used by farmers as a strategy to differentiate the product quality in specific market niches.

Keywords: Penjar tomato; landrace; ripening mutant; postharvest; ethylene production; respiration rate

1. Introduction

Tomato (Solanum lycopersicum L.) fruit quality, both sensory and nutritional, is becoming the first appeal for consumers, who complain about the deterioration of flavor in modern commercial cultivars [1] produced under high-input cropping systems [2]. Fruit quality traits, which are the result of the interaction between the chemical composition of the fruit and human sense-receptors, are regulated by complex interactions between genetic, environmental, agronomical and postharvest factors [3,4]. Achieving high yields and quality under the highest efficiency cropping systems is, to date, the main challenge for tomato growers.
In the past years, and boosted in part by consumers demanding flavorful fruit, tomato landraces have gained interest among growers, given their singular quality traits, combining in some cases good taste with high nutritional properties, but also their better adaptation to low-input agriculture [5–8]. Among them, Mediterranean long shelf life (LSL) landraces are highly appreciated in Italy and Spain [9], and exhibit a rich sensory and nutritional profile when they are grown under traditional cropping systems (i.e., low-input or water deficit conditions) [9–12]. Several LSL landraces are described in the literature, among which the Balearic “Ramellet” [13], the Catalan “Penjar” [14], and the Italian “Da Serbo” [15] are the most popular.

LSL landraces display specific adaptations to drought, as described in Balearic [16] and Italian [17] landraces. These varieties were historically grown in low-input, non-irrigated and non-trellised cropping systems. Farmers describe that under low-input cropping systems shelf life is improved [18], although this effect seems genotype-dependent. Water deficit (WD) induces in tomato, and multiple other fruits, a “concentration” effect leading to enhanced levels of specific antioxidants (i.e., vitamin C) but also of flavor-related compounds such as soluble sugars [3,4,19,20]. In the specific case of LSL landraces, several metabolites have been described as being positively affected by water shortage, such as soluble solids content (SSC) [21], carotenoids, polyphenols or L-ascorbic acid [22,23]. Nevertheless, much less is known regarding the effect of WD on physiological processes such as respiration and ethylene production in LSL [9], as well as the differences between traditional and modern LSL varieties. Moreover, under low watering regimes the water-use efficiency (WUE) of the tomato crop is optimized, although WD produces a negative effect on yield [24]. Thus, the production of LSL landraces under low-input farming practices allows to concomitantly maximize fruit quality and crop efficiency [25].

The ability to store fruits for several months is among the key traits distinguishing LSL landraces from other tomato cultivars. During storage the chemical composition of the tomato fruit evolves [26,27], triggering significant changes in the organoleptic and nutritional profile. These changes are more pronounced in the LSL tomatoes because of their longer shelf life than normal ripening varieties, and the common practice to consume the LSL fruits after a postharvest period [28–32]. For instance, the rearrangement of the fruit volatilome [33] during this postharvest phase enables the appearance of new aromas and tastes, and the fruit becomes soft, which generally impacts positively on consumer acceptance [29].

Recent success of LSL varieties in the market has triggered the transfer to modern and high-input cropping systems, in some cases diluting their singular quality traits. In the case of the Penjar landrace, current commercial fruits belong to modern and high yielding hybrids produced under high-input and protected cultivation [25,34]. Fruits are consumed not only after prolonged storage but also early after harvest or after a short storage period (30 days). Under this new commercial production scheme, both WD and postharvest storage can be used as strategies to increase tomato fruit quality while targeting specific consumers’ demands. The effects of the growing environment, water irrigation and fertilization on the agronomic performance of Penjar tomatoes has been recently studied, showing that low-input practices optimize crop efficiency and increase shelf life, boosting the profitability of farmers interested in selling “aged” tomatoes [25]. In this study we present the effects that WD, growing environment (tunnel vs. open-air) and short-term postharvest storage (30 days) have on ripening-related quality traits of Penjar tomatoes.

2. Materials and Methods

2.1. Plant Materials and Growing Conditions

A modern hybrid (545, Semillas Fitó, Barcelona, Spain) and a landrace (LC649, collected in Valencia, Spain) belonging to the Penjar variety were selected for this study. Plants were grown in 2018 during the summer season (April–September) in contiguous plots with the following treatments: (i) open field/tunnel (open-sided unheated tunnels, 3.4 m tall, 7 m wide, covered with a 200 µm thick polyethylene film with >91% global and <45%...
diffuse light transmission capacities, and thermicity factor >82%); (ii) irrigation dose, with a well-watered (WW) and a water deficit (WD) treatment. Previous to the opening of the first flower, irrigation was adjusted daily to the requirements of the crop calculated following the crop evapotranspiration (ETc) method based on soil–water balance [35]; subsequently, we followed two different irrigation strategies that farmers normally use in the area, which set the irrigation volume to water for 1 h (WD) or 4 h (WW) per week, resulting in irrigation amounts of 4795 m$^3$ ha$^{-1}$ (WW, 70% ET$_c$) and 2334 m$^3$ ha$^{-1}$ (WD, 35% ET$_c$). In order to simulate farmers’ practices, fertilization was coupled with the amount of irrigation, adjusting the electrical conductivity to 2 dS m$^{-1}$. The fertilizer used was potassium nitrate (13.8-0-45.1 NPK). Total amount of fertilizer applied was 152 kg ha$^{-1}$ N-NO$_3$− and 289 kg ha$^{-1}$ K$_2$O in the WD treatment, and 339 kg ha$^{-1}$ N-NO$_3$− and 580 kg ha$^{-1}$ K$_2$O in the WW treatment. Fields were managed using conventional farmer practices of the area, following integrated pest management (IPM) guidelines and conducting the plants to one stem by pruning lateral stems each 2 weeks. Plant density was 1.4 plants/m$^2$. More details on the growing and climatic conditions can be found in Casals et al. [25].

2.2. Fruit Sampling and Storage Conditions

For each combination genotype*treatment, 120 fruits from the 2nd to 4th trusses were harvested at the red-ripe stage, and separated in 3 biological replicates (40 fruits). Fruits were immediately placed in a dark room at 16 °C and 85–90% relative humidity to conduct the postharvest experiment. All measurements were performed in each replicate at harvest (0 days (d), 20 fruits) and after a short storage period (30d, 20 fruits). The 20 fruits per replicate used to perform measurements at 30d were individually weighted at the beginning of the experiment and after 30 days of storage, in order to calculate water loss (%). Prior to any quality evaluation, 10 fruit per replicate were used for physiological measurements (ethylene production and respiration rate). Physical traits were studied individually in 20 fruits per replicate by means of non-destructive measurements (external fruit color (fec), index of absorbance difference (IAD), acoustic firmness (AF)) at each time. Subsequently, these 20 fruits per replicate were used to destructively assess fruit firmness. From the same fruit used for destructive firmness evaluations, 4 pools per biological replicate of 5 fruit each were cut in half and used to further evaluate SSC and TA. The other halves from each pool were quickly snap-frozen in liquid nitrogen, grounded to a fine powder and kept at −80 °C until further biochemical analysis.

2.3. Physical Measurements

The index of absorbance difference (IAD = $A_{670} - A_{720}$), as an indicator of the fruit maturity, was measured with a DA-Meter (TR Turoni, Forli, Italy) on opposite sides of the equatorial parts of the fruit. Objective color values (L*, a*, b*, Chroma and Hue coordinates from the CIELAB color space) were measured using a portable spectrophotometer CM-2600d (Konica Minolta Sensing, Tokyo, Japan). All color measurements were taken on two equatorial sides of the fruit. Different tests aiming to determine the textural properties of the fruit were conducted: acoustic firmness (AF) using an AWETA AFS (AWETA International Ltd., Nootdorp, The Netherlands), obtaining both F0 (resonant frequency; Hz) and acoustic firmness values (AF; 10$^8$ Hz$^2$ g$^{2/3}$), and uniaxial compression test (peak positive force; PPF (Kg)) followed by a puncture test (firmness; Kg) performed using an Aname TA-XT2i texture analyser (Stable Micro Systems Ltd., Surrey, UK) as described in Camps and Gili [36].

2.4. Sugar and Organic Acid Content

Bulks of 5 fruits (ca. 350 g) per biological replicate were rinsed, cut and then blended to obtain a juice. Soluble solids content (SSC) was determined from the juice with a PAL-1 Pocket refractometer (ATAGO, Tokyo, Japan). Titratable Acidity (TA) was measured by diluting 5 mL of juice with 10 mL of deionized water and titrating with NaOH 0.1 N until pH 8.2.
Malic acid and sugars (sucrose, glucose and fructose) were extracted from frozen flesh tissue as described by Giné-Bordonaba et al. [37]. For sugars’ determination, 2 g of frozen flesh tissue were diluted in 5 mL of 62.5% (v/v) aqueous methanol solvent and placed in a thermostatic bath at 55 °C for 15 min, mixing the solution with a vortex every 5 min to prevent layering. Then, the samples were centrifuged at 24,000× g for 15 min at 20 °C. The supernatants of each sample were recovered and used for enzyme-coupled spectrophotometric determination of glucose and fructose (hexokinase/phosphoglucose isomerase) and sucrose (β-fructosidase) using commercial kits (BioSystems S.A., Barcelona, Spain) and following the manufacturer instructions. Malic acid was extracted dissolving 2 g of frozen tissue in 5 mL of distillate water. The samples were slightly shaken for 10 min at room temperature and then centrifuged at 24,000× g for 7 min at 20 °C. The resulting supernatant was recovered and used for enzyme-coupled spectrophotometric determination (L-malate dehydrogenase) of malic acid using commercial kits (BioSystems S.A., Barcelona, Spain) and following the manufacturer instructions.

2.5. Total Phenolic Content and Antioxidant Activity

Total phenolic compounds (TPC) and antioxidant capacity (FRAP) were determined using frozen tissue by mixing 3 g of tomato snap-frozen powder with 10 mL of 79.5% (v/v) methanol and 0.5% (v/v) HCl in Milli-Q water. Sample extraction was held at 20 °C with constant shaking for 2 h and mixing the samples every 30 min. The extract was centrifuged at 24,000× g for 30 min at 20 °C. From the same extract, TPC (mg gallic acid equivalents (GAE) g⁻¹ fresh weight (fw)) were measured by means of the Folin–Ciocalteu method and FRAP (mg Fe²⁺ g⁻¹ fw) measured by the Ferric Reducing Antioxidant Power assay as described elsewhere [20].

2.6. Ethylene Production Capacity and Respiration Rate

Ethylene production (µL kg⁻¹ h⁻¹) was measured as described by Giné-Bordonaba et al. [37] with some modifications. Samples of 10 fruit (700 g) per biological replicate were placed immediately after harvest in 2 L flasks sealed with a silicon septum for sampling the gas of the headspace after 2 h incubation in an acclimatized chamber at 20 °C. For the analysis of ethylene production, gas samples (1 mL) were taken using a syringe and injected into a gas chromatograph (GC; Agilent Technologies 6890, Wilmington, Germany) fitted with an FID detector and an alumina column F1 80/100 (2 m × 1/8 × 2.1, Tecknokroma, Barcelona, Spain), as previously described [38]. Fruit respiration (mg CO₂ kg⁻¹ h⁻¹) was determined from the same flasks used for ethylene measurements. After 2 h incubation at 20 °C, the headspace gas composition was quantified using a handheld gas analyser (CheckPoint O₂/CO₂, PBI Dansensor, Ringsted, Denmark).

2.7. Statistical Analyses

Previous to statistical analyses, data were curated and assumptions of homogeneity of variance (Levene’s test) and normal distribution (plot of residuals) of the data were confirmed. At each postharvest time (0d, 30d) a 3-way analysis of variance (ANOVA) was performed considering the fixed factors: genotype (G), environment (E) and irrigation (I). For significant factors (p < 0.05), mean separation (Student–Newman–Keuls, p < 0.05) was used to analyze differences between groups (G*E*I). Two principal component analyses (PCA) were built by using phenotypical values (3 replicates per G*E*I) at both 0d and 30d, with unit variance normalization. Finally, to identify traits affected by storage, a 4-way ANOVA was performed considering G, E, I and storage time (S) as fixed factors. Traits that were significantly affected by storage time or its interactions were further studied by a box plot and ANOVA, comparing the evolution of replicates and treatments between 0 and 30 days. Pearson correlation coefficients between variables at each period (0d, 30d) were calculated by using means per G*E*I. Statistical analyses were performed by using R software [39] using “ggplot2”, “factoextra”, “corrplot” and “ggfortify” packages.
3. Results and Discussion

3.1. Effect of Water Deficit and Growing Environment on Fruit Quality and Physiological Traits

At harvest, the main factors affecting fruit quality traits were first the genotype followed by water regime (Figure 1, Tables 1 and 2), while the growing environment (tunnel/open field) did not significantly alter the quality profile or the biochemical/physiological traits. The results of the ANOVA show that the genotype was significant for 12 out of 18 measured traits, the irrigation for 7 traits, and the environment for 4 traits (mainly fruit color coordinates (fec.L, fec.B, and fec.Hue) and FRAP) (Table S1). These results are in agreement with previous studies describing the growing environment’s significant impact on the agronomic behavior of five Penjar genotypes, although it did not significantly affect fruit quality traits [25].

![Figure 1](image-url)

**Figure 1.** Dissimilarities based on fruit’s physical and chemical traits recorded at harvest showing the effects of water regime (water deficit, WD; well-watered, WW) and growing environment (open field, OF; tunnel, T) on two Penjar genotypes (landrace, LR; modern cultivar, MV). Scores are represented on the two first principal components (Dim1–Dim2), accounting for 52.8% of the total variation. (a) Phenotypic values grouped by treatment (the center of the ellipse is signaled); (b) correlation loadings of the variables with the principal components. Abbreviations: (a) the abbreviations indicate the genotype (MV/LR), the water regime (WD/WW), and the environment (OF/T); for instance, MV_WD_T indicates: modern variety, water deficit, tunnel; (b) FW, fruit weight; AF, acoustic firmness; PPF, peak positive force; Firm, firmness; fec., fruit external color CIELAB coordinates (L, a, b, Chroma, Hue); FRAP, antioxidant capacity; TPC, total phenolic content; SSC, soluble solids content; TA, titratable acidity; IAD, Index of absorbance difference; Glu + fru, glucose + fructose content.
Table 1. Effect of water regime (water deficit, WD; well-watered, WW) and growing environment (open field, OF; tunnel, T) on fruit’s physical characteristics of two Penjar genotypes (landrace, LR; modern cultivar, MV) at harvest. Within columns, data indicated with different letters are significantly different ($p < 0.05$). Significance of the ANOVA for each variable is shown on the bottom line (significance levels: * $<0.05$, ** $<0.01$, *** $<0.001$, ns = not significant).

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Irrigation</th>
<th>Environment</th>
<th>Diameter (mm)</th>
<th>FW (g)</th>
<th>Firmness (kg)</th>
<th>L</th>
<th>a</th>
<th>b</th>
<th>Chroma</th>
<th>Hue</th>
</tr>
</thead>
<tbody>
<tr>
<td>MV</td>
<td>WD</td>
<td>OF</td>
<td>49.3 ab</td>
<td>64.1</td>
<td>ns</td>
<td>3.8 ab</td>
<td>44.3 bc</td>
<td>23.4 a</td>
<td>19.6 c</td>
<td>30.6 b</td>
</tr>
<tr>
<td>MV</td>
<td>WW</td>
<td>OF</td>
<td>49.0 ab</td>
<td>65.0 ns</td>
<td>3.7 ab</td>
<td>46.4 a</td>
<td>20.2 b</td>
<td>22.9 b</td>
<td>31.1 b</td>
<td>47.8 ab</td>
</tr>
<tr>
<td>MV</td>
<td>WD</td>
<td>T</td>
<td>47.0 b</td>
<td>60.3 ns</td>
<td>3.4 bc</td>
<td>44.1 bc</td>
<td>23.4 a</td>
<td>19.0 c</td>
<td>30.2 b</td>
<td>38.9 d</td>
</tr>
<tr>
<td>MV</td>
<td>WW</td>
<td>T</td>
<td>52.7 a</td>
<td>74.2 ns</td>
<td>4.1 a</td>
<td>46.0 ab</td>
<td>21.9 ab</td>
<td>20.2 c</td>
<td>30.0 b</td>
<td>42.4 cd</td>
</tr>
<tr>
<td>LR</td>
<td>WD</td>
<td>OF</td>
<td>51.4 a</td>
<td>70.3 ns</td>
<td>2.9 c</td>
<td>43.7 c</td>
<td>24.0 a</td>
<td>25.3 a</td>
<td>35.5 a</td>
<td>46.4 abc</td>
</tr>
<tr>
<td>LR</td>
<td>WW</td>
<td>OF</td>
<td>52.2 a</td>
<td>75.6 ns</td>
<td>3.3 bc</td>
<td>44.3 bc</td>
<td>21.8 ab</td>
<td>26.1 a</td>
<td>34.3 a</td>
<td>49.8 a</td>
</tr>
<tr>
<td>LR</td>
<td>WD</td>
<td>T</td>
<td>49.7 ab</td>
<td>65.9 ns</td>
<td>2.8 c</td>
<td>42.6 c</td>
<td>24.2 a</td>
<td>24.1 ab</td>
<td>34.4 a</td>
<td>44.6 bc</td>
</tr>
<tr>
<td>LR</td>
<td>WW</td>
<td>T</td>
<td>52.3 a</td>
<td>74.4 ns</td>
<td>3.3 bc</td>
<td>43.6 c</td>
<td>22.7 a</td>
<td>26.0 a</td>
<td>34.8 a</td>
<td>48.7 ab</td>
</tr>
</tbody>
</table>

Abbreviations: FW, fruit weight; fec., fruit external color CIELAB coordinates (L, a, b, Chroma, Hue).

Table 2. Effect of water regime (water deficit, WD; well-watered, WW) and growing environment (open field, OF; tunnel, T) on fruit’s chemical characteristics of two Penjar genotypes (landrace, LR; modern cultivar, MV) at harvest. Within columns, data indicated with different letters are significantly different ($p < 0.05$). Significance of the ANOVA for each variable is shown on the bottom line (significance levels: * $<0.05$, ** $<0.01$, *** $<0.001$, ns = not significant).

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Irrigation</th>
<th>Environment</th>
<th>Malic Acid (mg g$^{-1}$ fw)</th>
<th>Glucose + Fructose (mg g$^{-1}$ fw)</th>
<th>Citric Acid (mg g$^{-1}$ fw)</th>
<th>FRAP (mg Fe$^{2+}$ g$^{-1}$ fw)</th>
<th>TPC (mg GAE g$^{-1}$ fw)</th>
<th>Ethylene (µL kg$^{-1}$ h$^{-1}$)</th>
<th>Respiration (mg CO$_2$ kg$^{-1}$ h$^{-1}$)</th>
<th>SSC (°Brix)</th>
<th>TA (g Malic Acid L$^{-1}$)</th>
<th>Chlorophyll (IAD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MV</td>
<td>WD</td>
<td>OF</td>
<td>0.28 b</td>
<td>27.2 ab</td>
<td>3.03 ab</td>
<td>3.59 a</td>
<td>0.52 ns</td>
<td>1.18 b</td>
<td>0.026 a</td>
<td>6.9 a</td>
<td>3.2 a</td>
<td>0.05 ns</td>
</tr>
<tr>
<td>MV</td>
<td>WW</td>
<td>OF</td>
<td>0.22 b</td>
<td>25.2 abc</td>
<td>4.24 abc</td>
<td>2.77 bc</td>
<td>0.47 ns</td>
<td>1.00 b</td>
<td>0.021 ab</td>
<td>6.8 a</td>
<td>3.3 a</td>
<td>0.02 ns</td>
</tr>
<tr>
<td>MV</td>
<td>WD</td>
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<td>28.8 a</td>
<td>3.03 ab</td>
<td>3.31 abc</td>
<td>0.41 ns</td>
<td>0.63 b</td>
<td>0.021 ab</td>
<td>6.8 a</td>
<td>3.0 ab</td>
<td>0.00 ns</td>
</tr>
<tr>
<td>MV</td>
<td>WW</td>
<td>T</td>
<td>0.25 b</td>
<td>25.9 abc</td>
<td>4.23 abc</td>
<td>2.57 c</td>
<td>0.43 ns</td>
<td>0.56 b</td>
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<tr>
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<td>WD</td>
<td>OF</td>
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<td>1.96 ab</td>
<td>3.55 a</td>
<td>0.39 ns</td>
<td>1.06 b</td>
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<td>2.8 ab</td>
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<td>WW</td>
<td>OF</td>
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<td>3.02 abc</td>
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<td>0.88 b</td>
<td>0.017 b</td>
<td>5.1 c</td>
<td>2.4 b</td>
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<td>1.68 b</td>
<td>3.48 ab</td>
<td>0.39 ns</td>
<td>1.33 b</td>
<td>0.021 ab</td>
<td>6.8 a</td>
<td>3.2 ab</td>
<td>0.04 ns</td>
</tr>
<tr>
<td>LR</td>
<td>WW</td>
<td>T</td>
<td>0.49 a</td>
<td>16.0 d</td>
<td>1.43 b</td>
<td>2.61 c</td>
<td>0.35 ns</td>
<td>2.48 a</td>
<td>0.026 a</td>
<td>5.0 c</td>
<td>2.7 ab</td>
<td>0.07 ns</td>
</tr>
</tbody>
</table>

Abbreviations: FRAP, antioxidant capacity; TPC, total phenolic content; SSC, soluble solids content; TA, titratable acidity; IAD, index of absorbance difference.
Indeed, differences between tomato genotypes regarding firmness and the content of specific sugars and organic acids are well documented [4]. The average monosaccharide content reported herein (26.8 and 20.5 mg g\(^{-1}\) fw for MV and LR, respectively) was in line with that described previously for the Penjar variety [32,34] and standard fresh market tomatoes [40]. The content of citrate (average 2.7 mg g\(^{-1}\) fw), the main organic acid found in tomato fruit, was, on average, two-fold higher in the modern hybrid (MV) than in the landrace (LR) genotype, while the opposite trend was observed for malate (Table 2). The LR genotype was also characterized by enhanced color coordinates b* and Chroma, together with lower firmness than the MV (Table 1). Several correlations were identified between traits both at harvest and after the short storage period. FW, b*, Chroma and Hue coordinates were negatively correlated with sugars and fruit firmness (Figure S1). Texture-related traits (AF, Firmness, PPF) were highly correlated between them. By employing a principal component analysis (PCA), the first dimension of the PCA plot (capturing 34.4% of the total variance) clearly differentiated the two genotypes investigated herein, whereas clustering of the samples along the second dimension of the PC2 (18.2% of the total variance) was mainly related to differences in water regimes (Figure 1).

Within each genotype, WD enhanced the fruit color (redness (fec.a)), antioxidant capacity (FRAP) and SSC (Figure 1, Tables 1 and 2). Considering both growing environments, the mean increase under WD conditions was 9.8% for fec.a (MV: increase of 11.3% between WD and WW treatments; LR: 8.3%), 27.3% for FRAP (MV: 29.2%; LR: 25.4%) and 15.7% for SSC (MV: 5.6%; LR: 25.8%) (Tables 1 and 2). The effect of WD on fruit external color was more pronounced on the modern cultivar, showing enhanced redness (higher a*, lower Hue) and lower lightness (L*) in fruits from WD plants (Table 1). These results agree with previous findings of the positive effect of WD on fruit quality traits of LSL landraces [6,10,21,25] and general reports in tomato suggesting that sugars, acids and antioxidants are the main flavor and nutritional components affected by WD [4,41]. In contrast to the fruit antioxidant capacity measured by the FRAP assay, the total phenolic content (TPC) was not affected by the environmental/irrigation conditions tested herein. It is likely that specific phenolic compounds, but not the overall TPC content, was affected by WD conditions, as previously reported in other fruits [20].

In tomato, the majority of quality traits reported above are known to be ethylene dependent [42]. Ethylene production at harvest ranged from 0.56 to 1.33 µL kg\(^{-1}\) h\(^{-1}\), and was significantly higher (\(p < 0.001\)) in the LR (mean: 1.44 µL kg\(^{-1}\) h\(^{-1}\)) in comparison with the MV (mean: 0.84 µL kg\(^{-1}\) h\(^{-1}\)) (Table S1). Compared to the mean of the genotype, a two-fold higher value was recorded in the LR cultivated under WW and tunnel conditions (2.48 µL kg\(^{-1}\) h\(^{-1}\)). Likewise, the respiration rate ranged from 0.015 to 0.026 mg CO\(_2\) kg\(^{-1}\) h\(^{-1}\) with no significant differences between genotypes or environmental/irrigation conditions (Table S1). In general, both respiration rates and ethylene production have been poorly studied in LSL landraces [9], yet our results confirm the expected lower values in the Penjar tomato [28] than in commercial standard cultivars [43–45]. Although the ripening mutation responsible for the LSL phenotype was not confirmed in the genotypes evaluated in this study, most of the Penjar tomatoes bear the alc mutation affecting the NOR gene belonging to the NAC transcription factor family [14,28] and resulting in lower ethylene production rates accompanying extended fruit shelf life. The lower firmness observed in LR fruit was not related to differences in ethylene production and was rather likely associated to differences in cuticle composition, which are widely known among Penjar genotypes [28,46]. Reduced ethylene production in Penjar genotypes is also generally associated with an inhibition in the accumulation of carotenoids [28], the compounds responsible for the red color of tomato fruit. That said, in our study, differences in objective color (i.e., Hue angle) among genotypes were not associated to differences in the fruit ethylene production capacity (Figure S1).

Despite the general conception that a key trait distinguishing landraces is their phenotypic stability across environments [47,48], Figure 1 shows that the intra- and inter-environment heterogeneity for fruit quality traits was much higher in the LR than in the
MV genotype. It is noteworthy that the heterogeneity in the fruits' chemical profile was double in the LR (e.g., coefficient of variation (CV) across treatments of 36% for malic acid, 40% for citric acid or 32% for TA)) than in the MV (18% 26% and 21%, respectively), signaling the high heterogeneity of the LR regarding fruit quality traits, both within and across the growing environments.

Overall, the results from our study confirm that the genotype is the major source of variability for specific fruit quality traits, thereby, they are in agreement with previous studies reporting the high intra-varietal diversity affecting not only quality traits but also agronomic performance in Penjar tomatoes [11,14,32,34]. Based on this and previous studies, it is therefore evident that the selection of appropriate genotypes is the most important step for farmers aiming to obtain fruit of optimum organoleptic quality. Moreover, the high phenotypic heterogeneity of the LR in comparison with the MV, especially in response to WD treatment, points to the difficulty of standardizing the fruit quality attributes of this traditional genotype, limiting to some extent its marketability if grown under diverse environmental conditions (i.e., water regimes). By contrast, the MV variety shows a much more stable phenotype within each growing environment, facilitating the claim for specific fruit quality traits.

3.2. Effect of Short-Term Storage on Fruit Quality and Physiological Traits

After harvesting, LSL fruits are traditionally stored in dark rooms and consumed after an aging period that can vary from 1 to 6 months. This said, given their increased acceptability among consumers [9], current commercial practices have introduced the strategy to consume Penjar tomatoes already at harvest (fresh) or after a short storage period (few weeks). During storage, fruit metabolism continues leading to substantial changes in the fruit’s chemical profile [27], generally accompanied by a dramatic fruit weight loss and high incidence of postharvest rots. In our study, fruit lost 4.2–7.6% of their initial weight during the first 30 days of storage, hence, in accordance with previous results for the Penjar variety [14,32] (Table S2).

Multiple quality or ripening traits (SSC, TA, color (Chroma, L*), malic acid, sugars (glucose + fructose), ethylene production and respiration rate) significantly changed during storage at 16 °C (Figure 2). In detail, most of them tended to decline as the fruit ripened during storage while only certain color attributes (Chroma) increased during storage (i.e., fruit color saturation tended to diminish). The decline in sugars and acids during storage is not unusual and can be explained by their consumption as respiratory substrates [49,50] yet such a decline may be underestimated due to the concentration effect resulting from the observed fruit weight loss [31,32].

Ethylene production and respiration rate were reduced during storage but solely in the MV variety, showing a reduction of 44% in ethylene production and of 28% in the respiration rate. By contrast, in the LR we did not observe this reduction, and the postharvest pattern seemed to be strongly influenced by the higher heterogeneity at harvest. The reduction in the physiological activity of tomato fruit during postharvest storage has been described in other commercial tomato cultivars [45] and in some Penjar accessions [28] where the highest ethylene production rate is normally observed close to the time of harvest. The non-homogenous response of the LR to the postharvest suggests that other factors not analyzed in this study may govern the postharvest activity of the fruit, and possibly its shelf life potential. Elucidating the factors underlying this specific and diverse postharvest behavior can be an important step towards a better understanding of the regulation of the LSL phenotype.
Figure 2. Box plots (25th–75th percentile; center line, median) showing the evolution of physical, chemical and physiological traits after 30 days of storage. Lines connect the values of replicates. Within each genotype (LR: landrace; MV: modern cultivar) ***, **, *, and ns indicates significant at <0.001, <0.01, <0.05 levels, and non-significant differences between 0 and 30 days, respectively.

In order to evaluate if the fingerprint that provokes preharvest factors on fruit profile was maintained during storage, the PCA analysis was repeated with the results after 30 days of storage (Figure 3). The Dim1-Dim2 scatterplot explained 57.8% of the total variance, and depicted a similar pattern of dissimilarity between genotypes and treatments to that observed at harvest (Figure 1). After 30 days of storage, the overall differences between genotypes and treatments were maintained, although smoothed (less significant differences between treatments and genotypes were found (Tables S1–S3)). Variables contributing to the distinction between genotypes (mainly Dim1) and watering regimes (mainly Dim2) were similar to the time of harvest, with a major contribution of ethylene production and respiration rate on Dim2.

In general, the compounds that clearly impact the fruit sensory profile decreased during storage, making the fruit less sweet and sour (Figure 2). Nevertheless, fruits of both genotypes still maintained high levels of sugars and acids after 30 days, as well as high values of antioxidant compounds. In combination with the rearrangement of the fruit volatilome, enabling the appearance of new aromas [29], the results from this study confirm that after one month of postharvest storage, the sensory profile of the Penjar tomato evolves towards a differentiated product. Reports on the evolution of the fruit’s chemical profile during longer storage periods (>6 months) in LSL landraces [29–32] seem to indicate that such high quality is maintained till 3 months after harvesting. One-month-aged tomatoes can have a high potential for farmers aiming to differentiate local productions by offering a singular sensory profile to consumers while paying a lower cost in terms of fruit weigh loss (average 4–7%) than in the case of more aged fruits (average fruit weight loss at 4 months = 12–18% [32]).
Figure 3. Dissimilarities based on fruit’s physical and chemical traits recorded after 30 days of storage showing the effects of water regime (water deficit, WD; well-watered, WW) and growing environment (open field, OF; tunnel, T) on two Penjar genotypes (landrace, LR; modern cultivar, MV). Scores are represented on the two first principal components (Dim1-Dim2), accounting for 52.8% of the total variation. (a) Phenotypic values grouped by treatment (the center of the ellipse is signaled); (b) correlation loadings of the variables with the principal components. Abbreviations: (a) the abbreviations indicate the genotype (MV/LR), the water regime (WD/WW), and the environment (OF/T); for instance, MV_WD_T indicates: modern variety, water deficit, tunnel; (b) FW, fruit weight; AF, acoustic firmness; PPF, peak positive force; Firm, firmness; fec., fruit external color CIELAB coordinates (L, a, b, Chroma, Hue); FRAP, antioxidant capacity; TPC, total phenolic content; SSC, soluble solids content; TA, titratable acidity; IAD, Index of absorbance difference; Glu + fru, glucose + fructose content.

4. Conclusions

Long shelf life landraces cultivated under low-input cropping systems possess unique nutritional and organoleptic traits that make them good candidates for recovering sensory and nutritional quality in tomato. Considering the enormous variability for fruit quality traits that can be found within these landraces, farmers from the Mediterranean area have a good opportunity to build new niche markets based on singularity and tradition. Water deficit can be used as a strategy to increase crop efficiency and fruit quality, enhancing some sensory (SSC) and nutritional (antioxidant capacity) attributes of LSL tomatoes. The positive effect of these preharvest factors is maintained during the short-term storage of the fruits (30 days), which boosts the profitability of these tomato varieties as they can be sold in the market for a longer period.

Supplementary Materials: The following are available online at https://www.mdpi.com/article/10.3390/agronomy1112304/s1, Figure S1: Correlation matrix for fruit quality traits at harvest (0d) and after the short-term storage period (30d). Table S1: The p-values for the 3-way ANOVA for quality parameters analyzed at harvest and after 30 days of storage. Table S2: Effect of water regime (water deficit, WD; well-watered, WW) and growing environment (open field, OF; tunnel, T) on fruit physical characteristics of two Penjar genotypes (landrace, LR; modern cultivar, MV) after 30 days.
of storage. Table S3: Effect of water regime (water deficit, WD; well-watered, WW) and growing environment (open field, OF; tunnel, T) on fruit chemical characteristics of two Penjar genotypes (landrace, LR; modern cultivar, MV) after 30 days of storage.

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