

Article

Comparative Nutrient and Antioxidant Profile of High Lycopene Variety with *hp* Genes and Ordinary Variety of Tomato under Organic Conditions

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Abstract: Organic tomato cultivation is growing all over the world due to its healthy functional properties and environmental concerns. Recently, some new varieties with increased functional properties, particularly high lycopene, have been developed. However, few were assessed under organic farming systems. The objective of this study was to evaluate physico-chemical properties and the main bioactive compound contents, as well as lipophilic, hydrophilic and total radical scavenging activities of a recently developed high lycopene tomato variety homozygous for *hp-2^{ds}* genes (HLT-F71) grown under organic conditions for two non-consecutive years (2019 and 2021) compared to the ordinary control ‘Nemador’ variety. The lycopene, β -carotene, total phenol, flavonoid, vitamin C, radical scavenging activity and tocopherols were analyzed by using spectrophotometric and HPLC methods, respectively. The high lycopene content variety presented suitable marketable yield, average fruit weight, pH, titratable acidity, firmness and higher °Brix, pulp color than the control. Additionally, it has significantly higher lycopene, β -carotene, total phenol, flavonoid, vitamin C and particularly considerable α -tocopherol contents, as well as radical scavenging activity. The high lycopene tomato variety with *hp* genes proved to be an effective sustainable variety for enhancing tomato fruit yield and functional properties even under organic grown conditions.

Keywords: bioactive compounds; lycopene; organic farming; radical scavenging activity; tomato; yield



Citation: Romdhane, A.; Riahi, A.; Ujj, A.; Ramos-Diaz, F.; Marjanović, J.; Hdider, C. Comparative Nutrient and Antioxidant Profile of High Lycopene Variety with *hp* Genes and Ordinary Variety of Tomato under Organic Conditions. *Agronomy* **2023**, *13*, 649. <https://doi.org/10.3390/agronomy13030649>

Academic Editors: Dominika Średnicka-Tober, Renata Kazimierczak and Krystian Marszałek

Received: 17 January 2023

Revised: 20 February 2023

Accepted: 22 February 2023

Published: 23 February 2023



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1. Introduction

Tunisia is one of the main tomato (*Solanum lycopersicum* L.) producers in the world. Indeed, the tomato crop occupies a strategic place in Tunisia with a harvested area of 24,540 ha and a production of 1,416,000 tons [1]. Organic farming, among the sustainable solutions, occupies a very important place in Tunisia which is the result of policies supporting this sector and underlined in a national strategy. Organic crops intended for vegetables remain weak and fluctuating with 211 ha in 2019 far behind the olive with 251,569 ha and other fruit trees with 14,118 ha [2]. In Tunisia, the main organic cultivated species were peas, potatoes, garlic, parsley, tomatoes, artichoke, and pepper.

Tomatoes are popular, appreciated and widely consumed all over the world and particularly in Tunisia and there is an increasing demand for organic tomatoes. Consumers are now concerned with the quality of final food, as well as how it has been produced. In

fact, this demand is closely linked to increased awareness of the impact of foods on the well-being and health of people [3].

Tomatoes have several beneficial effects for health due to their richness in a multitude of antioxidant compounds and biofunctional properties [4–7]. However, most of the available data relating to the physicochemical and nutraceutical characteristics of tomato fruit are obtained in conventional cultivation mode, while those obtained in organic mode are still insufficient.

The choice of variety is an important criterion in the success of the organic tomato culture. Indeed, the genetic factors linked to the variety can influence several important parameters such as production potential, disease resistance and fruit quality [8,9]. Recently, there has been great interest in developing or enhancing, through conventional plant breeding or biotechnology, tomato varieties rich in several bioactive compounds such as lycopene, anthocyanin, β -carotene, zeaxanthin [10–13]. This has been performed more successfully for high lycopene variety with *hp* genes [14]. These were obtained by introgression of *high-pigment* (*hp*) and *old-gold* (*og*) mutations. *Hp* tomato mutants (*hp-1* and *hp-2*) determine an amplified phytochrome-mediated response to light causing higher levels of carotenoids, principally lycopene, and other antioxidant compounds [4]. However, little is known about the agronomic and quality performance of these promising new plant materials, particularly under organic farming. In fact, it would be interesting to combine the health beneficial effects of high bioactive compounds level varieties with those of organic cultivation. Then, assessing tomato cultivars rich in bioactive compounds suitable for organic conditions is necessary. Although there is great interest in these newly developed varieties, few studies [15–17] evaluated high lycopene tomato cultivars under organic farming systems and when it was the case several important quality attributes were not studied. In addition, the existence of important environmental effects and interactions suggests the need to assess the behavior of varieties under organic growing conditions specifically for each environment [17]. This current study aimed to evaluate agronomic, physico-chemical and phytonutrient properties of a recently developed high lycopene tomato variety with *hp* genes (HLT-F71) grown under organic conditions compared to the ordinary open pollinated tomato variety ‘Nemador’.

2. Materials and Methods

2.1. Growth Conditions

Open field tomato trials were conducted for two non-consecutive years in 2019 and 2021 seasons at the support research station certified organic farm in Manouba governorate, northern Tunisia (36°48'28" N 10°6'4" E) characterized by a classic Mediterranean climate. The trial in 2020 was canceled due to COVID-19. The soil was a clay loam in texture with 29% clay, 18% loam and 32% sand and was rich in total 30% and active 18% calcareous material, with electrical conductivity of 0.13 ms cm⁻¹. The soil was rich in total nitrogen (0.16%), phosphorus (45 mg kg⁻¹), potassium (869 mg kg⁻¹) and magnesium (728 mg kg⁻¹) but low in calcium (68 mg kg⁻¹) and organic matter 1.85% with suitable pH 7 for tomato. The means monthly air temperature (minimum/maximum), humidity and rainfall recorded for the same season during the growing of tomato for the years 2019 and 2021 are presented in Table 1. Two tomato varieties were used in this experiment. The promising field tomato high lycopene variety homozygous for *hp-2^{dg}* genes, named HLT-F71, selected by the National Agricultural Research Institute of Tunisia was characterized by a high content of lycopene and antioxidants. It has been selected from an advanced tomato breeding lines by conventional plant breeding methods mainly for the high-lycopene attribute which is generally due to the occurrence of light-responsive high-pigment (*hp*) mutations [18]. The open pollinated tomato variety ‘Nemador’, one of the most cultivated in Tunisia, was used as a control. The seeds are produced organically.

The experimental plot had artichoke as a previous crop in 2018 and was fallow in 2020 in previous rotations. The trial was carried out in an organic plot converted since 2004 and controlled by the ECOCERT certified organism. The organic growing techniques

used are in accordance with national organic requirements which are also in accordance with European standards. These techniques include organic fertilization at the rate of 20 tons per hectare of certified cattle manure from extensive farming [19]. Organic growing methods also comprised weed elimination by hand and plant pathogen treatments with some approved bio pesticides, employed when disease first indications were noticed, as indorsed by the Technical Center of Organic Agriculture, Chott Mariem and described by Riahi et al. [19]. Drip irrigation for 1 to 2.5 h with a flow rate of 4 L h⁻¹ at one to two day intermissions depending on potential evapotranspiration for research site climatic data and plant cultural coefficient was employed. The sowing was carried out on 15 March 2019 and 20 March 2021 in alveolar boxes. Plantlets were transplanted in an open field at the end of April in double rows with spacing within rows and between double rows of approximately 40 cm and 150 cm, respectively. The experimental design was a randomized complete block with four replications and a density of 3 plants m⁻². Each replication comprised 30 plants per variety.

Table 1. Climate data recorded for the same season during the growing of tomatoes for the years 2019 and 2021.

Period	Tmin (°C) ^a		Tmax (°C) ^b		Humidity (%)		Rainfall (mm)	
	2019	2021	2019	2021	2019	2021	2019	2021
April	14	14	20	21	79	78	45	46
May	17	19	23	25	70	65	51	41
June	24	25	31	31	62	63	26	15
July	27	28	34	34	60	52	04	01

Abbreviations: ^a minimum temperature; ^b maximum temperature.

2.2. Fruit Sampling

Tomato fruits were manually picked randomly from the central part of each plant at each twin rows on July 20 to 25 for both 2019 and 2021. A representative sample of fruits of about two kilograms of tomatoes belonging to the red stage for every cultivar and block was taken to the laboratory the same day. A portion of the samples were instantly used for certain analyses (°Brix, pH, titratable acidity and color) and the other portions were frozen at −20 °C and used within the limit of one week for the other analyzes of lycopene, β-carotene, total phenols, flavonoid, vitamins contents and antioxidant activity.

2.3. Agronomical and Physicochemical Determinations

Fruit weight was obtained to determine average fruit weight and extrapolated to calculate marketable yield per hectare. Soluble solid content was measured using a drop of filtered tomato juice and by the use of a digital refractometer (Atago PR-100, NSG Precision Cells, Inc., Farmingdale, NY, USA), taking into account the temperature and indicated in (°Brix). pH was determined using an electronic pH meter (WTW PH 539, Weilheim, Germany) and titratable acidity, expressed as percentage (%) of citric acid, was assessed after titration at pH 8.1 with sodium hydroxide (0.1 M). Firmness was assessed with an electronic penetrometer (Penefel, Penefel électronique, Setop-Giraud Technologies, Cavaillon, France) and expressed in kg×cm⁻². The tomato total (peel) and fresh cut (pulp) color of the fruits were determined in accordance with the CIELab system [Commission Internationale de l'Éclairage, L* (lightness), a* (redness) and b* (yellowness)] by means of a Minolta Chromameter (CR-400, Minolta corp.). Then, the ratio a*/b* was calculated.

2.4. Lycopene, β-Carotene, Total Phenols and Flavonoid Determinations

Lycopene and β-carotene were extracted and analyzed spectrophotometrically as previously and fully described by Fish et al. [20] and Lee [21], respectively. Results were expressed in milligram per kilogram of tomato fresh weight (mg × kg⁻¹, fw). Total phenols and flavonoid contents were extracted and analyzed spectrophotometrically, as previously

and fully described by Riahi and Hdider [22]. They were quantified in milligram gallic acid equivalent per kilogram of tomato fresh weight ($\text{mg GAE} \times \text{kg}^{-1}$, fw) and milligram of rutin equivalents per kilogram of fresh weight ($\text{mg RE} \times \text{kg}^{-1}$, fw), respectively.

2.5. Vitamins Determinations

Vitamin C content is the addition of both ascorbic acid (AsA) and dehydroascorbic acid contents. The latter were analyzed, as previously described in detail [23]. Briefly, 0.1 g duplicate of the homogenate tomato samples were extracted and detected spectrophotometrically at 525 nm in Cecil BioQuest CE 2501 (Cecil Instruments Ltd., Cambridge, UK). The standard curve used in the assay was from 0 to 700 mmol AsA. Vitamin C content was expressed in milligram per kilogram of fresh weight ($\text{mg} \times \text{kg}^{-1}$, fw).

Tocopherols were extracted on duplicate samples with n-hexane as earlier described by Daood et al. [24]. The determination of α -, β -, γ -tocopherol isomers content was carried out using a Hitachi Chromaster high-performance liquid chromatography with fluorescence detection (Hitachi Chromaster, Tokyo, Japan). The HPLC system consists of a model 5210 autosampler, a model 5110 gradient pump and a 5440 fluorescence detector. The separation was performed on Nucleosil 5 mm column (250×4.6 mm i.d.) with a mobile phase consisting of 99.5:0.5 n-hexane: ethanol. The excitation and emission wavelength were set at 295 nm and 320 nm, respectively, as reported in Duah et al. [25]. Peaks of tocopherols isomers α -, β - and γ - were obtained using external standards (Sigma-Aldrich, Budapest, Hungary) cochromatographed with the samples. The data obtained were expressed as a milligram of tocopherols per kilogram of fresh weight ($\text{mg} \times \text{kg}^{-1}$, fw).

2.6. Hydrophilic and Lipophilic Radical Scavenging Activity Determinations

The hydrophilic and lipophilic radical scavenging activities (HRSA and LRSA, respectively) were determined by the ABTS decoloration procedure described by Miller and Rice-Evans [26]. The extraction of the antioxidants from the hydrophilic and lipophilic fractions was performed on 0.3 g of the tomato sample (three independent replicates) using methanol (50%) acetone (50%), respectively, at 4 °C under constant shaking (300 rpm) during 12 h. A sample centrifugation was performed at $10,000 \times g$ during 7 min and the supernatant was recovered before its utilization for radical scavenging activity measurements. Absorbance was measured at 734 nm using a Cecil BioQuest CE 2501 spectrophotometer (Cecil Instruments Ltd., Cambridge, UK). Two different calibration curves were dressed using freshly prepared Trolox solutions for HRSA and LRSA determinations. A linear calibration curve was obtained with Trolox solution (0–16 μM Trolox) and results were recorded as μM of Trolox $\times 100 \text{ g}^{-1}$ of fresh weight (fw). Total radical scavenging activity is the sum of lipophilic and hydrophilic activities.

2.7. Statistical Analysis

Data were expressed as means $\pm$ standard error of four replicates. The differences in all agronomical and qualitative data were compared by analysis of variance (ANOVA) using the software package SAS Version 9.2 software (SAS Institute, Cary, NC, USA). The means were separated by a least significant difference (LSD) test at the 0.05 significance level.

3. Results

3.1. Climatic Data

Means monthly air temperature (minimum/maximum) and relative humidity during the two experimental years 2019 and 2021 were very close (Table 1). However, the total mean monthly precipitation recorded was higher in 2019 with 126 mm compared with 103 mm in 2021.

3.2. Agronomical and Physicochemical Properties

The high lycopene tomato variety with *hp* genes (HLT-F71) grown under the organic farming conditions of the study was characterized by dark foliage and dark green immature fruit compared to the ordinary tomato 'Nemador' variety. Results for the main agronomical and physicochemical properties of the varieties HLT-F71 and 'Nemador' grown under the organic farming conditions of study are shown in Table 2. Marketable yield, average fruit weight, soluble solids, pH, titratable acidity, firmness and color were not significantly influenced by the growing years ($p > 0.05$). The marketable yield of tomato fruit harvested in 2019 and 2021 ranged from $62.14 \text{ t} \times \text{ha}^{-1}$ in 'Nemador' to $69.11 \text{ t} \times \text{ha}^{-1}$ in HLT-F71. When averaged across year, the results showed no significant difference between tomato varieties in marketable yield ($p > 0.05$). Mean marketable yields recorded for the two varieties HLT-F71 and 'Nemador' were 66.52 and $63.68 \text{ t} \times \text{ha}^{-1}$, respectively. Average fruit weight ranged from 71.64 g in HLT-F71 to 76.61 g in 'Nemador' for the two growing seasons. There was no significant difference ($p > 0.05$) between the studied tomato varieties whatever the year and mean values were 72.63 g and 75.62 g for the high lycopene tomato variety and the ordinary tomato variety 'Nemador', respectively. Comparable fruit sizes were found in the different tomato varieties under study. Soluble solids of tomato fruit harvested in 2019 and 2021 varied from 5.03°Brix in 'Nemador' to 5.79°Brix in HLT-F71. Soluble solids varied significantly ($p < 0.05$) between the different tomato varieties under study whatever the year. Higher values were recorded in the variety HLT-F71 compared to the variety 'Nemador'. Mean soluble solids were 5.75 and 5.12°Brix in the high lycopene tomato variety and the ordinary tomato variety 'Nemador', respectively. pH, titratable acidity and firmness values showed no significant difference between tomato varieties whatever the year ($p > 0.05$). For color, when fruit peel readings were considered, a^* (redness), b^* (yellowness) and the ratio a^*/b^* were not significantly different between the two varieties whatever the year ($p > 0.05$). This is expected since it is difficult to differentiate between the fruit color of the two varieties by external visualization. When pulp readings were considered, the color readings a^* , b^* and the ratio a^*/b^* varied significantly in pulp-cut tomato fruits ($p < 0.05$). The mean index of redness a^* decreased from 35.80 in pulp HLT-F71 to 20.12 in pulp 'Nemador' fruits. Regarding the ratio a^*/b^* , pulp high-lycopene tomato berries had the highest values 1.23 compared to 0.91 for pulp 'Nemador'.

Table 2. Yield and physico-chemical properties of tomato varieties grown organically during the two growing seasons. Data are expressed as mean $\pm$ S.E. with four replicates.

Parameters	Varieties					
	HLT-F71			Nemador		
	2019	2021	Mean	2019	2021	Mean
Marketable yield ($\text{t} \times \text{ha}^{-1}$)	$69.11 \pm 4.8 \text{ a}$	$63.93 \pm 4.4 \text{ a}$	66.52 A	$65.22 \pm 4.6 \text{ a}$	$62.14 \pm 4.3 \text{ a}$	63.68 A
Average fruit weight (g)	$73.62 \pm 5.1 \text{ a}$	$71.64 \pm 4.9 \text{ a}$	72.63 A	$76.61 \pm 5.3 \text{ a}$	$74.63 \pm 5.2 \text{ a}$	75.62 A
Soluble solids ($^\circ\text{Brix}$)	$5.79 \pm 0.3 \text{ a}$	$5.71 \pm 0.3 \text{ a}$	5.75 A	$5.21 \pm 0.2 \text{ a}$	$5.03 \pm 0.2 \text{ a}$	5.12 B
pH	$4.45 \pm 0.2 \text{ a}$	$4.35 \pm 0.2 \text{ a}$	4.40 A	$4.38 \pm 0.2 \text{ a}$	$4.32 \pm 0.2 \text{ a}$	4.35 A
Titratable acidity (% citric acid)	$0.36 \pm 0.04 \text{ a}$	$0.34 \pm 0.03 \text{ a}$	0.35 A	$0.40 \pm 0.05 \text{ a}$	$0.38 \pm 0.04 \text{ a}$	0.39 A
Firmness ($\text{kg} \times \text{cm}^{-2}$)	$4.31 \pm 0.3 \text{ a}$	$4.13 \pm 0.2 \text{ a}$	4.22 A	$4.23 \pm 0.3 \text{ a}$	$4.01 \pm 0.2 \text{ a}$	4.12 A
Color						
$(a^*)_{\text{peel}}$	$27.48 \pm 1.9 \text{ a}$	$26.12 \pm 2.1 \text{ a}$	26.80 A	$24.83 \pm 1.5 \text{ a}$	$25.61 \pm 2.2 \text{ a}$	25.22 A
$(b^*)_{\text{peel}}$	$28.63 \pm 1.8 \text{ a}$	$27.79 \pm 2.2 \text{ a}$	28.21 A	$27.30 \pm 2.4 \text{ a}$	$27.42 \pm 2.5 \text{ a}$	27.36 A
$(a^*/b^*)_{\text{peel}}$	$0.96 \pm 0.08 \text{ a}$	$0.94 \pm 0.07 \text{ a}$	0.95 A	$0.91 \pm 0.06 \text{ a}$	$0.93 \pm 0.07 \text{ a}$	0.92 A
$(a^*)_{\text{pulp}}$	$36.49 \pm 2.5 \text{ a}$	$35.11 \pm 2.3 \text{ a}$	35.80 A	$20.21 \pm 1.4 \text{ a}$	$20.02 \pm 1.1 \text{ a}$	20.12 B
$(b^*)_{\text{pulp}}$	$29.79 \pm 2.1 \text{ a}$	$28.41 \pm 2.0 \text{ a}$	29.10 A	$22.45 \pm 1.6 \text{ a}$	$21.76 \pm 1.5 \text{ a}$	22.11 B
$(a^*/b^*)_{\text{pulp}}$	$1.22 \pm 0.08 \text{ a}$	$1.24 \pm 0.09 \text{ a}$	1.23 A	$0.90 \pm 0.05 \text{ a}$	$0.92 \pm 0.06 \text{ a}$	0.91 B

a Lower case letters indicate mean separation within row and variety; values with different letters were significantly different at $p < 0.05$ by LSD test. A,B Capital letters indicate mean separation among variety means within a row; values with different letters were significantly different at $p < 0.05$ by LSD test.

3.3. Fruit Nutrient Contents

Lycopene, β -carotene, total phenol, flavonoid, vitamin C and tocopherols contents of the high-lycopene tomato variety with *hp* genes (HLT-F71) and the ordinary 'Nemador' grown under the organic farming conditions of study are shown in Table 3. The fruit nutrient contents measured were not significantly influenced by the growing year ($p > 0.05$). The Lycopene content in tomato fruit harvested in 2019 and 2021 ranged from $72.2 \text{ mg kg}^{-1} \text{ fw}$ in 'Nemador' to $148.7 \text{ mg kg}^{-1} \text{ fw}$ in HLT-F71. Lycopene contents varied significantly among tomato varieties under study whatever the year ($p < 0.05$). Mean lycopene ranged from $74.5 \text{ mg kg}^{-1} \text{ fw}$ in 'Nemador' to $140.5 \text{ mg kg}^{-1} \text{ fw}$ in HLT-F71 variety. The Lycopene content of the high-lycopene tomato variety was 1.9 times that of the 'Nemador' variety. β -carotene content ranged from $4.2 \text{ mg kg}^{-1} \text{ fw}$ in 'Nemador' to $13.7 \text{ mg kg}^{-1} \text{ fw}$ in HLT-F71 for the two growing years. β -carotene contents varied significantly between the tomato varieties under study whatever the year ($p < 0.05$). Higher values were recorded in the variety HLT-F71 compared to the variety 'Nemador'. Mean values for β -carotene ranged from $4.5 \text{ mg kg}^{-1} \text{ fw}$ to $13.0 \text{ mg kg}^{-1} \text{ fw}$ in 'Nemador' and HLT-F71 variety, respectively. The β -carotene content of the high-lycopene tomato variety was 2.9 times that of the 'Nemador' variety.

Table 3. Lycopene, β -carotene, total phenols, flavonoid, vitamin C and tocopherol contents of tomato varieties grown organically during the two growing seasons. Data are expressed as mean $\pm$ S.E. with four replicates.

Properties	Varieties					
	HLT-F71			Nemador		
	2019	2021	Mean	2019	2021	Mean
Lycopene ($\text{mg} \times \text{kg}^{-1} \text{ fw}$)	$148.7 \pm 5.9 \text{ a}$	$132.2 \pm 5.2 \text{ a}$	140.5 A	$72.2 \pm 4.1 \text{ a}$	$76.8 \pm 4.4 \text{ a}$	74.5 B
β -carotene ($\text{mg} \times \text{kg}^{-1} \text{ fw}$)	$13.7 \pm 0.5 \text{ a}$	$12.2 \pm 0.4 \text{ a}$	13.0 A	$4.2 \pm 0.4 \text{ a}$	$4.7 \pm 0.5 \text{ a}$	4.5 B
Total phenols ($\text{mg GAE} \times \text{kg}^{-1} \text{ fw}$)	$265.6 \pm 10.6 \text{ a}$	$233.7 \pm 9.3 \text{ a}$	249.7 A	$138.9 \pm 8.1 \text{ a}$	$148.6 \pm 8.7 \text{ a}$	143.8 B
Flavonoids ($\text{mg RE} \times \text{kg}^{-1} \text{ fw}$)	$353.7 \pm 14.1 \text{ a}$	$311.3 \pm 12.4 \text{ a}$	332.5 A	$98.8 \pm 9.8 \text{ a}$	$107.3 \pm 10.6 \text{ a}$	103.1 B
Vitamin C ($\text{mg} \times \text{kg}^{-1} \text{ fw}$)	$218.1 \pm 8.7 \text{ a}$	$191.9 \pm 7.7 \text{ a}$	205.0 A	$150.1 \pm 9.1 \text{ a}$	$157.0 \pm 9.5 \text{ a}$	153.6 B
Vitamin E (α -tocopherol) ($\text{mg} \times \text{kg}^{-1} \text{ fw}$)	$27.7 \pm 0.8 \text{ a}$	$24.7 \pm 0.7 \text{ a}$	26.2 A	$17.0 \pm 0.5 \text{ a}$	$17.5 \pm 0.6 \text{ a}$	17.3 B

a Lower-case letters indicate mean separation within row and variety; values with different letters were significantly different at $p < 0.05$ by LSD test. A,B Capital letters indicate mean separation among variety means within a row; values with different letters were significantly different at $p < 0.05$ by LSD test.

Total phenols in tomato fruit harvested in 2019 and 2021 ranged from $138.9 \text{ mg GAE} \times \text{kg}^{-1} \text{ fw}$ in 'Nemador' to $265.6 \text{ mg GAE} \times \text{kg}^{-1} \text{ fw}$ in HLT-F71. Flavonoid content ranged from $98.8 \text{ mg RE} \times \text{kg}^{-1} \text{ fw}$ in 'Nemador' to $353.7 \text{ mg RE} \times \text{kg}^{-1} \text{ fw}$ in HLT-F71 for tomato berries harvested in 2019 and 2021, respectively. Total phenol and flavonoid contents varied significantly among tomato varieties under study whatever the year ($p < 0.05$). Mean total phenols ranged from $143.8 \text{ mg GAE} \times \text{kg}^{-1} \text{ fw}$ in 'Nemador' to $249.7 \text{ mg GAE} \times \text{kg}^{-1} \text{ fw}$ in HLT-F71 variety. Mean flavonoid content ranged from $103.1 \text{ mg RE} \times \text{kg}^{-1} \text{ fw}$ in 'Nemador' to $332.5 \text{ mg RE} \times \text{kg}^{-1} \text{ fw}$ in HLT-F71 variety. The total phenol and flavonoid contents of the high-lycopene tomato variety were 1.7 and 3.2 times that of the 'Nemador' variety, respectively.

Vitamin C in tomato berries harvested in 2019 and 2021 ranged from $150.1 \text{ mg} \times \text{kg}^{-1} \text{ fw}$ in 'Nemador' to $218.1 \text{ mg} \times \text{kg}^{-1} \text{ fw}$ in HLT-F71. Vitamin E content was quantified as α -tocopherol. For β -tocopherol and γ -tocopherol, contents were less than the limit of quantification in the samples and are not listed in the table. α -tocopherol in tomato berries harvested in 2019 and 2021 ranged from $17.0 \text{ mg} \times \text{kg}^{-1} \text{ fw}$ in 'Nemador' to $27.7 \text{ mg} \times \text{kg}^{-1} \text{ fw}$ in HLT-F71. Vitamin C and α -tocopherol contents recorded for the experiment were significantly different between tomato varieties whatever the year ($p < 0.05$). Mean vitamin C and tocopherol amounts ranged from 153.6 to $205.0 \text{ mg} \times \text{kg}^{-1} \text{ fw}$ and from 17.3 to $26.2 \text{ mg} \times \text{kg}^{-1} \text{ fw}$ in 'Nemador' and HLT-F71, respectively. The high lycopene tomato

variety showed significantly higher vitamin C and tocopherols contents than the ordinary tomato variety ‘Nemador’. Vitamin C and tocopherols contents of the high-lycopene tomato variety were 1.3 and 1.5 times that of the ‘Nemador’ variety, respectively.

3.4. Radical Scavenging Activity

Lipophilic, hydrophilic and total radical scavenging activity of the high-lycopene tomato variety HLT-F71 and the ordinary ‘Nemador’ are shown in Figure 1. LRSA, HRSA and total radical scavenging activity were not significantly influenced by the growing year ($p > 0.05$). LRSA ranged from $100.3 \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$ in ‘Nemador’ to $338.6 \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$ in HLT-F71 variety. HRSA ranged from $70.8 \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$ in ‘Nemador’ to $234.3 \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$ in HLT-F71 variety in 2019 and 2021. Total radical scavenging activity ranged from $171.1 \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$ in ‘Nemador’ to $572.9 \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$ in HLT-F71 variety in 2019 and 2021, respectively. LRSA, HRSA and total radical scavenging activity varied significantly between the tomato varieties under study whatever the year ($p < 0.05$). Mean LRSA ranged from $107.7 \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$ in ‘Nemador’ to $319.9 \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$ in HLT-F71 variety. Mean HRSA ranged from $74.8 \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$ in ‘Nemador’ to $219.4 \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$ in HLT-F71 variety. The LRSA and HRSA of the high-lycopene tomato variety were 3 and 2.9 times that of the ‘Nemador’ variety, respectively. Mean total radical scavenging activity was $182.5 \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$ in ‘Nemador’ and $547.3 \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$ in HLT-F71 variety. The total radical scavenging activity of the high-lycopene tomato variety was three times that of the ‘Nemador’ variety.

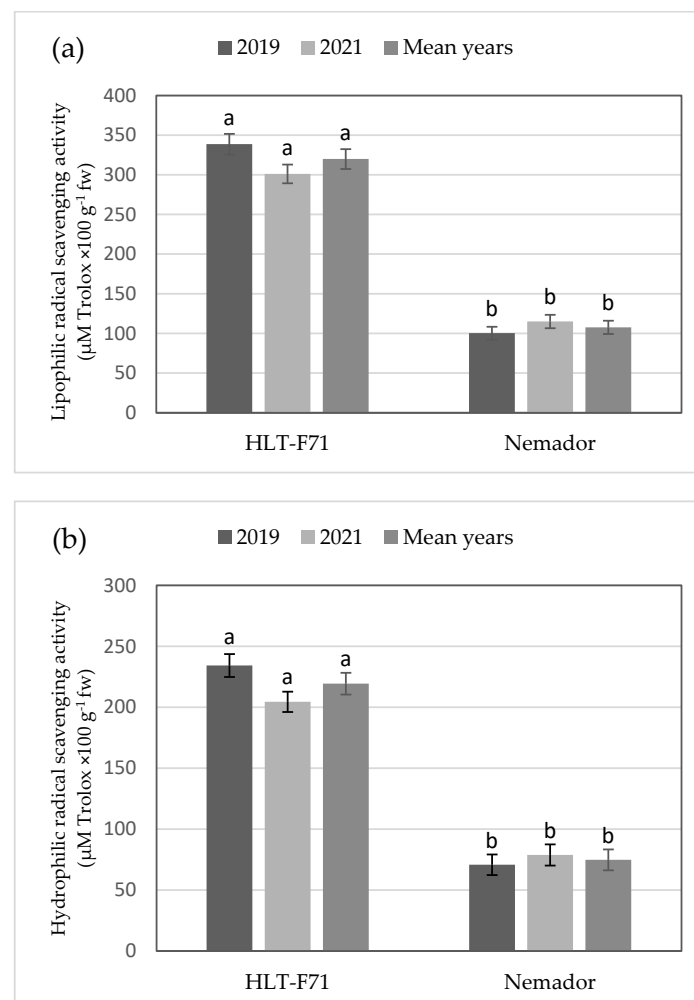


Figure 1. Cont.

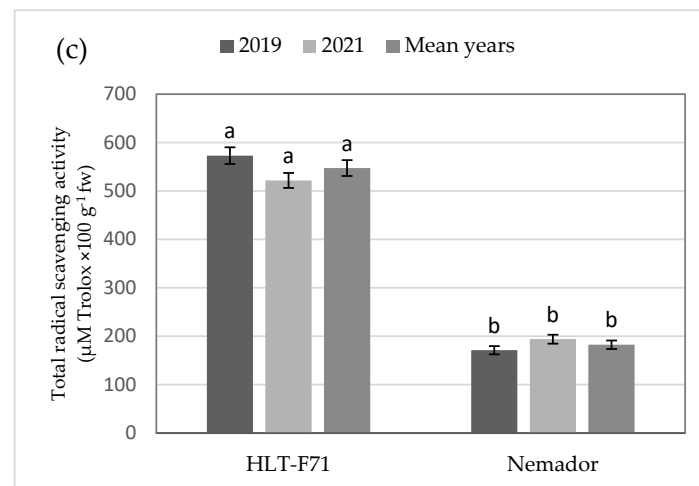


Figure 1. (a) Lipophilic, (b) hydrophilic and (c) total radical scavenging activity of the tomato varieties grown organically during the two growing seasons. Error bars represent $\pm$ S.E. ($n = 4$). ^{a,b} Within each year, means with different letters are significantly different at $p < 0.05$ by LSD test.

4. Discussion

The distinct varietal characteristics such as dark foliage and dark green immature fruit observed in high lycopene tomato variety (HLT-F71) grown under the organic farming conditions of study have also been reported previously in conventional farming practices [23]. Although yields recorded in the experimental trial are lower than those recorded among conventional elite farmers and which can exceed $100 \text{ t} \times \text{ha}^{-1}$, they are acceptable and above the average yield of $57.70 \text{ t} \times \text{ha}^{-1}$ in 2021 in Tunisia [1]. The yields were comparable to those of Giordano et al. [15] who reported that ‘BRS Tospodoro’, a high lycopene content tomato cultivar, has been evaluated under organic crop systems and attained a yield of $70.59 \text{ t} \times \text{ha}^{-1}$ significantly higher than the ordinary cultivar ‘Nemador’ with $51.63 \text{ t} \times \text{ha}^{-1}$. Furthermore, Lahoz et al. [16] found that the high lycopene organically grown cultivars ‘Kalvert’ and ‘ISI-24424’ obtained yields attaining 94.9 and $94.2 \text{ t} \times \text{ha}^{-1}$, respectively.

The results revealed that mean fruit fresh weight, soluble solids, pH, titratable acidity and firmness values were acceptable. Total soluble solids are among the most important quality parameters and represent an indicator of sweetness since tomatoes include glucose and fructose, as main components [27]. Our results are comparable to those obtained by Murariu et al. [28], who found values varied from 4.10 °Brix to 5.60 °Brix in tomatoes studied under organic field conditions. Nevertheless, De Sio et al. [29] obtain higher values arranging from 5.8 °Brix to 6.1 °Brix in three organic tomato hybrids cultivated in southern Italy. The results for pH content are comparable with those obtained by researchers from Romania, who found that pH values ranged from 4.32 to 4.53 in genotypes grown under organic field conditions [28]. The pH data obtained for organic pulp tomatoes are perhaps linked to a higher concentration of organic acids, especially malic and citric acids. These generally decrease during maturity, as they are the substrate for respiration [30]. Additionally, Sulieman et al. [31] related that the fresh tomato must maintain the pH level at four since it is favorable for storage and preservation. Regarding titratable acidity, the values found in the current study were slightly lower than those obtained for organic tomato grown in Spain by Ayuso-Yuste et al. [32] varying between 0.26 and 0.36% of citric acid in red ripening stage. This is probably due to varietal differences. The fruits of tomato varieties grown under organic conditions were firm and had quality characteristics which were comparable to those obtained in organic farming [19]. Tomato fruit firmness is a necessary trait of quality since it allows resistance to over-ripeness in the field, little juice loss and resistance to physical damage in transportation. Regarding color and although fruit peel readings were not significantly different between the two varieties, pulp high-

lycopene tomato HLT-F71 fruits obtained more intense red color than pulp of the ordinary 'Nemador' fruits. Similarly, Lahoz et al. [10] evaluated color of standard and high lycopene tomato cultivars under organic farming and found that the high lycopene cultivars 'Kalvert' and 'ISI-24424' obtained the more intense red color pulp with 2.43 and 2.31 Hunter a^*/b^* , respectively, compared to normal red color with 1.99 Hunter a^*/b^* . Additionally, a^*/b^* values obtained in tomato peel were in the range of those obtained by Ayuso-Yuste et al. [32] from 0.97 to 1.72.

The data also confirmed that genotype considerably affects lycopene and β -carotene contents in organic tomatoes. Lycopene values were higher than that obtained by Fracchiolla et al. [33]. Although the level of this major pigment was reduced under organic farming, HLT-F71 still accumulates a high content with respect to the commonly tomato varieties cultivated under organic farming conditions. Likewise, Lahoz et al. [16] found that the high lycopene organically grown cultivars 'ISI-24424' and 'Kalvert' obtained highest levels reaching 170.1 and 167.1 $\text{mg} \times \text{kg}^{-1}$ fw, respectively. Recently, similar high lycopene content was noticed in conventional farming with values attaining 348.8 $\mu\text{g} \times \text{g}^{-1}$ fw in the wild tomato followed by HLY18 and ISI12152 with 153.6 and 145.2 $\mu\text{g} \times \text{g}^{-1}$ fw, at the red ripe stage, respectively [10]. Similarly, high accumulation of carotenoids reaching 2.27 and 2.00 $\text{mg} \times \text{kg}^{-1}$ fw in the high lycopene cultivars 'Kalvert' and 'ISI-24424', respectively, were obtained under organic farming [16]. Lower β -carotene values with 1.42 $\text{mg} \times \text{kg}^{-1}$ fw [16] and with 0.6 $\mu\text{g} \times \text{g}^{-1}$ fw [33] were found in organically grown standard tomato cultivars.

For total phenol and flavonoid contents, the results agree with those found by Martí et al. [17] who reported that the use of high lycopene cultivars such as 'Kalvert' can offer increased levels of polyphenols, particularly rutin, chlorogenic acid, and naringenin. 'Nemador' values were in the range of previously reported data for total phenols and flavonoids for common field organically grown tomatoes with (0.154 to 0.162 $\text{mg GAE} \times \text{g}^{-1}$ fw) and (0.109 to 0.113 $\text{mg RE} \times \text{g}^{-1}$ fw), respectively [22]. However, total phenols values were lower than those reported by Rodríguez Ortiz et al. [34] between 21.5 and 21.6 $\text{mg GAE} \times 100 \text{ mg}^{-1}$ fw.

Tomato is one of the major sources of vitamin C in the Mediterranean diet. Vitamin C has been associated with various health-promoting effects in fruits plants including tomato [35–37]. These are the first data on vitamin C content in organic grown tomato in Tunisia. Similarly, Martí et al. [17] demonstrated that the use of high lycopene cultivars such as 'Kalvert' can offer increased levels of L ascorbic acid 135.02 $\text{mg} \times \text{kg}^{-1}$ fw which was lower than values obtained in this study.

Vitamins, particularly vitamin E, existing in tomato are naturally occurring micronutrients, necessary for regular metabolism of organisms. Vitamin E comprises eight diverse chemical arrangements with four tocopherols (α -, β -, γ - and δ), and four tocotrienols (α -, β -, γ - and δ). The α -tocopherol form is retained in human plasma and is used to precise dietary reference value for vitamin E. Vitamin E is an indispensable nutrient that cannot be provided by the human body and then needs to be furnished solely by food. The dietary reference values determined and recommended by The European Food Safety Authority for alpha-tocopherol were 13 mg/day and 11 mg/day for men and women, respectively. These standards are also endorsed for children over ten years old. Data related to vitamin E in organically grown tomatoes is very limited. To our knowledge, this is the first report on high lycopene tomatoes grown organically and the second report on vitamin E in organically grown tomato and provides evidence that tomato grown organically constitutes a significant source of vitamin E dependent on the variety. Fracchiolla et al. [33] evaluated the use of some cover crops on yield and several quality attributes on tomatoes grown organically in Puglia and found close values varied from 14.3 to 26.8 $\mu\text{g} \times \text{g}^{-1}$ fw for α -tocopherol. The amounts of α -tocopherol measured in the tomato grown organically under analysis are in the range or even superior to the values reported for conventionally ordinarily and high pigment tomatoes. In fact, values were close to those recently found in conventional tomato, ranging between 10.23–31.69 $\text{mg} \times \text{kg}^{-1}$ fw for α -tocopherol in 'Rio

Grande' variety [38]. Additionally, Lenucci et al. [39] determined a very high content of α -tocopherol in conventionally grown high pigment tomato hybrids ($22 \text{ mg} \times \text{kg}^{-1}$ of fw in HLY 13; $16 \text{ mg} \times \text{kg}^{-1}$ of fw in HLY 18).

Generally, when grown organically, tomatoes are reported to have high levels of total phenolics, flavonoid, vitamin C and vitamin E [33,40]. This is due to stress conditions leading to oxidative stresses in organic farming system [41,42]. In addition, having a higher content of α -tocopherol, the high lycopene tomato variety can probably better tolerate stress under organic farming conditions since increased α -tocopherol levels are known as an indicator of stress tolerance [43].

The TEAC method has been and remains one of the most recommended methods for the determination of radical scavenging activity of fruit and vegetables since it measures with high consistency and repeatability both hydrophilic and lipophilic radical scavenging activity [38]. The data obtained in this study were close to that previously reported for LRSA, HRSA and total radical scavenging activities in organically grown tomato with 123.8 , 81.5 and $205.3 \text{ } \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$, respectively [22]. The results were slightly lower than the antioxidant capacity values, between 273 and $296 \text{ } \mu\text{M Trolox} \times 100 \text{ g}^{-1} \text{ fw}$, determined by the ABTS method by Rodríguez-Ortiz et al. [34]. Higher antioxidant activities were probably due to the presence of the *hp* mutations increasing both hydrophilic (polyphenols and vitamins) antioxidants and lipophilic (lycopene and total carotenoids) antioxidants.

Although the rainfall in 2019 was higher, the year to year of climatic data was minor and with no or little effect on the agronomical, physicochemical and nutrient properties as well as radical scavenging activity of the studied varieties. This demonstrates minimal interaction between the new high lycopene tomato fruit variety (HLT-F71) and environmental conditions and, therefore, stable agronomic and quality properties.

5. Conclusions

The present study confirmed that the high lycopene content variety (HLT-F71) constitutes a reservoir of diverse antioxidant molecules, even when cultivated under organic growing conditions. Indeed, the lycopene, β -carotene, total phenols, flavonoids, vitamin C, α -tocopherol and total radical scavenging activity of the high-lycopene tomato variety were 1.9, 2.9, 1.7, 3.2, 1.3, 1.5 and 3 times that of the control 'Nemador' variety, respectively. This variety can therefore be recommended to combine the beneficial health effects of the variety rich in bioactive compounds with those of organic farming. In the future, further data over a longer period of time are needed. In addition, uses of this variety in future breeding programs or the study of the effect of cultivation techniques on its performance could be considered.

Author Contributions: Conceptualization, C.H.; methodology, A.R. (Amani Romdhane); software, C.H. and A.R. (Anissa Riahi); validation, C.H., A.U. and A.R. (Anissa Riahi); formal analysis, A.R. (Amani Romdhane) and A.R. (Anissa Riahi); investigation, C.H.; resources, C.H., A.U. and A.R. (Anissa Riahi); data curation, A.R. (Amani Romdhane); writing—original draft preparation, A.R. (Amani Romdhane); writing—review and editing, C.H., A.R. (Amani Romdhane), A.R. (Anissa Riahi), J.M., F.R.-D. and A.U.; visualization, C.H.; supervision, C.H. and A.R. (Anissa Riahi); project administration, C.H.; funding acquisition, C.H. F.R.-D. and A.U. All authors have read and agreed to the published version of the manuscript.

Funding: This work was supported by the Ministry of Higher Education and Scientific Research of Tunisia through the funding allocated to the research Laboratory of Horticulture (LR16INRAT03), National Agricultural Research Institute of Tunisia, Tunisia and the Doctoral School of Economics and Regional Sciences, Hungarian University of Agriculture and Life Sciences.

Data Availability Statement: The data presented in this study are available on request from the authors.

Acknowledgments: The authors thank Kamel Arfaoui and Gaith Bouzayen from the Vegetables Interprofessional Grouping GIL Manouba Tunisia, for the administrative and technical support.

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

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