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Effects of Agricultural Practices on Color, Carotenoids Composition, and Minerals Contents of Sweet Peppers, cv. Almuden

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Consumers demand organic products because they believe they are more flavorful and respectful to the environment and human health. The effects of conventional, integrated, and organic farming, grown in a controlled greenhouse, on color, minerals, and carotenoids of sweet pepper fruits (*Capsicum annuum*), cv. Almuden, were studied. Experimental results proved that organic farming provided peppers with the highest (a) intensities of red and yellow colors, (b) contents of minerals, and (c) total carotenoids. Integrated fruits presented intermediate values of the quality parameters under study, and conventional fruits were those with the lowest values of minerals, carotenoids, and color intensity. As an example, the concentrations of total carotenoids were 3231, 2493, and 1829 mg kg^{-1} for organic, integrated, and conventional sweet peppers, respectively. Finally, organic red peppers could be considered as those having the highest antioxidant activity of all studied peppers (agricultural farming and development stage).

KEYWORDS: Capsanthin; *Capsicum annuum*; CIELab coordinates; nutrients; organic farming

INTRODUCTION

Organic markets are thriving, as a growing number of consumers worldwide prefer organic products. Growth in the industry is fuelled in part by consumers' attitudes toward food production systems and product quality. With respect to product quality, surveys indicate that consumers consider organic foods to be more positive for human health and the environment and more flavorful than their conventionally grown counterparts (*1*).

The total organic area in the European Union (EU-15), fully converted and under conversion, increased from 2 300 000 ha in 1998 to 4 900 000 ha in 2002; the organic farming area reached 3.8% of the total utilized agricultural area of the EU-15 in 2002, up from 1.8% in 1998. The number of registered organic producers increased from 100 280 in 1998 to 135 191 in 2003, which represents a 6% average annual increase over the 1998–2003 period (*2*).

In Spain, 5841 ha of sweet peppers were cultivated under greenhouse conditions and 1352 ha in Murcia (*3*). In Murcia, about 70 ha of sweet peppers are under organic agriculture, and yields range between 6 and 10 kg m^{-2}, often very close to those under conventional agriculture as reported by del Amor (*4*). In this way, higher nutrient availability in the conventional treatment could contribute more to the vegetative components (leaves and stems) of the plant than the generative components (fruits).

Conventional, organic, and sustainable agriculture are the primary cultural practices used in the production of foods in Europe and the United States of America. The goal of each of these practices differs greatly with respect to crop yield, land and pesticide use, and environmental impact. Conventional agricultural practices utilize high-yield crop cultivars, chemical fertilizers and pesticides, irrigation, and mechanization. Although conventional practices result in reliable high-yield crops, there is concern regarding the negative biological and environmental consequences and long-term sustainability associated with these practices (*5*).

Some studies have been performed to evaluate the impact of cultural practices (organic versus conventional production) in

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Table 1. Biological Control during the Crop Cycle in Conventional, Integrated, and Organic Sweet Pepper Farming

days after transplanting	agricultural practice		
	conventional	integrated	organic
63		*Aphidius colemani* *Ambliseius cucumeris*	*Aphidius colemani* *Ambliseius cucumeris*
69		*Aphidius colemani* *Orius laevigatus*	*Aphidius colemani* *Orius laevigatus*
100		*Aphidius colemani* *Phytoseilus persimilis*	*Aphidius colemani* *Phytoseilus persimilis*
118		*Aphidius colemani* *Eretmocerus mundos*	*Aphidius colemani* *Eretmocerus mundos*
128		*Aphidius colemani* *Ambliseius californicus* *Orius laevigatus*	*Aphidius colemani* *Ambliseius californicus* *Orius laevigatus*
149		*Phytoseilus persimilis*	*Phytoseilus persimilis*
154		*Aphidius colemani* *Aphidoletes aphidimyza* *Phytoseilus persimilis*	*Aphidius colemani* *Aphidoletes aphidimyza* *Phytoseilus persimilis*
163		*Phytoseilus persimilis* *Ambliseius californicus* *Orius laevigatus*	*Phytoseilus persimilis* *Ambliseius californicus* *Orius laevigatus*
189	*Bacillus thuringensis*	*Bacillus thuringensis*	*Bacillus thuringensis*
210			*Bacillus thuringensis*
226		*Phytoseilus persimilis*	

different fruit species on nutrients and antioxidant activity content (*6–8*).

Peppers, like other fruits, vary in chemical composition, even within the same variety, depending on maturity, the location of production, and agricultural practices as well as on numerous environmental factors (*9*, *10*).

Peppers are popular vegetables because of the combination of color, taste, and nutritional value. Fresh peppers are one of the vegetables with a high content of vitamin C and are a good source of pro-vitamin A carotenoids (*11–13*). The importance of carotenoids in the diet has been recognized, as vitamin A precursors and antioxidants in cell protection, in the prevention of degenerative diseases, and for human epithelial cell differentiation (*14*). Moreover, carotenoid pigments are responsible for the red color of the peppers.

As previously mentioned, many people believe that organic foods are healthier than conventionally produced foods and that they are produced in a more environmentally compatible manner (*7*). However, this consumers' expectation must be properly demonstrated by scientific studies. The main problem in a comparative study of organically and conventionally produced foods is the selection of the experimental fields in such a manner that they truly represent the cultivation forms which must be compared. The culture system in a controlled greenhouse is ideal to compare the effect of culture types on different quality parameters because it removes environmental factors such as climate and soil conditions.

Our objectives were to determine the effect of maturation and the type of agricultural practices (organic, integrated, and conventional) on the CIEL*a*b* color parameters, mineral content, total carotenoid content, and carotenoid composition of sweet peppers grown in a controlled greenhouse.

MATERIALS AND METHODS

Plant Material, Growth Conditions, and Experiment Design. Sweet pepper plants (*Capsicum annuum* L.), cv. Almuden, were transplanted from a commercial nursery on December 14th, 2005. Plants were grown in a plastic greenhouse, which was divided into eight independent lysimeters. Each lysimeter was 7.6-m-long and 6.5-m-wide, with seven lines of drip irrigation and its own fertilizer and water control unit. Each lysimeter contained a total of 126 plants with a 4 L h^{-1} dripper per plant. The irrigation schedule was applied according to the U.S. Weather Bureau Class A evaporation plan, which was placed inside the greenhouse. Each lysimeter received, before transplanting, 4 kg m^{-2} of bovine manure. The greenhouse had automated control of the relative humidity by a fog system.

The conventional treatment consisted of the application of the local farmers' fertilizer (chemicals) dosage and pesticides (**Tables 1** and **2**) as soon as infestation was detected; the integrated treatment received half the fertilizer dosage applied in the conventional treatment, and the organic treatment only received water without fertilizer after transplanting, and natural products and predators were used to control infestations and pathogens (**Tables 1** and **2**). Organic and integrated cultivations followed all rules established by the Board of Organic Agriculture of the Murcia region (*15*).

The experiment was designed in three blocks according to the culture type: organic, integrated, and conventional. The fruit were harvested at different times throughout ripening, thus providing three samples per block in terms of their maturity stage: immature green, green, and red. So, the total of samples was nine (3 agricultural practices × 3 maturity stages). Each sample consisted of 20 fruits and was analyzed in triplicate.

Color Coordinates. Color determinations were made, at 25 ± 1 °C, using a Hunterlab Colorflex spectrophotometer (Hunterlab, Reston, VA). This spectrophotometer uses an illuminant D_{65} and a 10° observer as references.

Peppers were dried until a constant weight was reached at 65 °C, and 0.5 g of dried peppers were diluted to 100 mL with ultrapure water, and finally a homogenized aliquot was used for color measurements. A sample cup for reflectance measurements was used (5.9 cm internal diameter × 3.8 cm height) with a path length of light of 10 mm. Blank measurements were made with the cup filled with distilled water against a reference white background.

Color data are provided as CIEL*a*b* coordinates, which define the color in a three-dimensional space: L^* indicates lightness, and a^* and b^* are the chromaticity coordinates, green–red and blue–yellow coordinates, respectively. L^* is an approximate measurement of luminosity, which is the property according to which each color can be considered as equivalent to a member of the gray scale, between black and white, taking values within the range 0–100; a^* takes positives values for reddish colors and negative values for the greenish ones, whereas b^* takes positive values for yellowish colors and negative values for the bluish ones (*16*, *17*).

C^* is chroma $[C^* = \sqrt{(a*^2)+(b*^2)}]$, 0 being at the center of a color sphere and increasing according to the distance from the center.

Table 2. Chemical Control during the Crop Cycle in the Conventional, Integrated, and Organic Sweet Pepper Farming

	agricultural practice		
days after transplanting	conventional	integrated	organic
97	pymetrozine	pymetrozine	potash soap
	potash soap	potash soap	
104	sulfur	sulfur	sulfur
132	avermectin		
147	avermectin	pyriproxyfen	sulfur
		sulfur	
154	cyproconazole	cyproconazole	sulfur
160		avermectin	
	pyriproxyfen	pyriproxyfen	
170	kresoxim-methyl	kresoxim-methyl	sulfur
185			neem oil
	cyproconazole		sulfur
189	sulfur	sulfur	
197			neem oil
202		neem oil	neem oil
		kresoxim-methyl	sulfur
210		sulfur	
202		pymetrozine	
		sulfur	
229		pymetrozine	sulfur

Finally, h_{ab} is the hue angle [$h_{ab} = \text{arc}\, tg\,(b^*/a^*)$], which is defined as starting at the $+a^*$ axis and is expressed in degrees; 0° would be $+a^*$ (red), 90° would be $+b^*$ (yellow), 180° would be $-a^*$ (green), and 270° would be $-b^*$ (blue) (*17*).

The color analyses were run for eight replicates.

Carotenoids Profile. The analysis of the carotenoid composition requires an extraction with organic solvent followed by de-esterification of the carotenoid fraction. Dry pepper samples were extracted with 50 mL of acetone, by using a Kika Labortechnik, T25 basic homogenizer (Jankel & Kunkel Gmbh & Co., Staufen, Germany), until the complete exhaustion of color occurred (extraction was repeated until filtrates were colorless). All extracts were pooled in a separator and shaken with diethyl ether. A sufficient quantity of 10% NaCl was added at the end to aid in the separation of the phases. Subsequently, the organic phase was dried over anhydrous Na_2SO_4. This phase, containing the pigments, in various stages of esterification with fatty acids, was saponified with 100 mL of 20% KOH–methanol for 1 h at room temperature. The pigments were subsequently extracted with diethyl ether, evaporated in a rotary evaporator, and taken up in a maximum of 10 mL of acetone. Aliquots (1 mL) of this were centrifuged at 12 000 rpm and stored at –30 °C until analyzed.

The carotenoid profile of the extract was quantified using β-apo-8′-carotenal (Sigma Chemical Co.; St. Louis, MO) as an internal standard, added at the beginning of the extraction process, 1 mL of 100 μg mL^{-1}. This chemical was used as an internal standard because it is absent in peppers and, under the proposed conditions, it separates well from other carotenoids (*11*).

The high-performance liquid chromatography system consisted of an HP-1100 series unit with a photodiode array detector equipped with HP ChemStation software (Hewlett Packard, Palo Alto, CA). The column used was a 250 mm × 4.6 mm i.d., YMC C30, S-5 μm (YMC, Schermbeck, Germany). The mobile phase for this column was 81:15:4 methanol/methyl tertiary butyl ether (MTBE)/H_2O (solvent A) and 91:9 MTBE/methanol (solvent B). The gradient elution was 100% A to 50% A and 50% B in 45 min followed by 100% B in the next 10 min and 100% A in the next 5 min at a flow rate of 0.8 mL min^{-1} (*18, 19*). Carotenoids were monitored at 450 nm.

All analyses were carried out in triplicate.

Identification was based on the order of elution, retention time, and spectra of absorbance maxima of a particular peak. Standards of β-carotene and β-cryptoxanthin were obtained from Sigma and Extrasynthese (Genay, France), respectively. Finally, standards of capsanthin and capsorubin were isolated from a saponified extract of red pepper (*20*). The average fortification recoveries (for a 10 μg carotenoid spike on 10 mL of a 20 mg L^{-1} standard) through the complete method were 94, 89, 90, and 92 % for capsorubin, capsanthin, β-cryptoxanthin, and β-carotene, respectively.

Mineral Content. A multiplace digestion block, Selecta Block Digest 20 (Barcelona, Spain), was used for sample mineralization. A total of 1 g of dried pepper was treated with 5 mL of 65% (w/v) HNO_3 in Pyrex tubes, placed in the digestion block, and heated at 60 °C for 60 min and at 130 °C for 120 min (*7*). Solutions were left to cool to room temperature, transferred to a volumetric flask, and diluted to a final volume of 25 mL with ultrahigh-purity deionized water.

Determination of Ca, Mg, K, Cu, Fe, Mn, and Zn in previously mineralized samples was performed with a Unicam Solaar 969 atomic absorption spectrometer (Unicam Limited, Cambridge, U.K.).

Instruments were calibrated using certified standards. In each analytical batch, at least two reagent blanks, one international reference material (CRM) and one spike, were included to assess precision and accuracy for chemical analysis. The certified material selected for the current experiment was GBW07603 (bush, branches, and leaves); this material was provided by LGC Deselaers S.L. (Barcelona, Spain) and produced by the Institute of Geophysical and Geochemical Exploration of China (GBW07603). Analyses were run in five replicates.

Statistical Analysis. Physico-chemical analyses, instrumental color measurements, carotenoid profiles, and mineral contents were run, at least, in three replications. All data were subjected to an analysis of variance and the Tukey least significant difference multicomparison test to determine significant differences among agricultural practices and maturity stages. The statistical analyses were done using SPSS 12.0 (SPSS Science, Chicago, IL) and figures using Sigma Plot 8.0 (SPSS Science, Chicago, IL).

RESULTS AND DISCUSSION

Color Coordinates. The external appearance of fruits, particularly their color, is of prime importance when considering the different attributes which define quality. It is especially important in the case of fruits destined for fresh consumption (*21, 22*). The change in color of the pepper surface takes places as a result of chlorophyll degradation and a considerable increase in its carotenoid content, which is influenced by the temperature and illumination to which the fruit is exposed (*23*).

Table 3 summarizes color data for changes due to development stage (immature green, green, and red) and agricultural practice (conventional, integrated, and organic). The statistical study showed that significant differences were found for both of the factors under study, development stage and agricultural practice ($p \leq 0.001$).

Table 3. CIEL*a*b* Color Coordinates throughout Development and Maturation in Sweet Pepper (*Capsicum annuum*) cv. Almuden Growing under Organic, Integrated, and Conventional Systems[a]

ripening stage	agricultural practice	L^*	a^*	b^*	C^*	h_{ab}
immature green	conventional	67.34 ± 0.10 f	−2.09 ± 0.04 a	23.25 ± 0.03 a	23.34 ± 0.05 a	95.14 ± 0.09 f
	integrated	71.02 ± 0.05 g	−2.50 ± 0.08 b	24.73 ± 0.06 b	24.86 ± 0.05 b	95.77 ± 0.06 f
	organic	75.34 ± 0.03 h	−3.08 ± 0.02 c	25.04 ± 0.02 b	25.22 ± 0.03 b	97.02 ± 0.06 g
green	conventional	61.50 ± 0.04 d	6.23 ± 0.05 d	31.36 ± 0.05 c	31.97 ± 0.02 c	78.75 ± 0.05 e
	integrated	64.75 ± 0.06 e	7.90 ± 0.05 e	35.34 ± 0.09 d	36.21 ± 0.02 d	77.40 ± 0.04 d
	organic	67.59 ± 0.02 f	11.68 ± 0.02 f	46.78 ± 0.01 e	48.21 ± 0.01 e	75.98 ± 0.05 c
red	conventional	30.76 ± 0.03 a	31.81 ± 0.04 g	49.71 ± 0.07 f	59.01 ± 0.03 f	57.38 ± 0.03 a
	integrated	46.31 ± 0.04 b	34.51 ± 0.07 h	76.86 ± 0.06 g	84.25 ± 0.05 g	65.82 ± 0.08 b
	organic	54.32 ± 0.04 c	39.61 ± 0.05 i	89.59 ± 0.06 h	97.95 ± 0.02 h	66.14 ± 0.02 b

[a] Values in this table are the mean ± standard error of eight replicates. Data with the same letters were not significantly different at $p < 0.001$ for the attribute evaluated (Tukey multiple range test).

Chlorophylls are the most abundant pigments in immature green peppers, while carotenoids will predominate in red peppers (*23*). In this way, it can be expected that green coordinates take lower values while red coordinates increase their values.

As peppers mature, they become darker (L^* takes higher values), starting with a value of 71.23 (immature green) and changing to 64.61 (green) and 43.80 (red). In addition, organic peppers were always darker than the integrated and conventional ones.

The green–red coordinate, a^*, initially took negative values (−2.56) and increased significantly as peppers matured (8.60 and 35.31 for green and red peppers, respectively). Again, organic peppers were more reddish than the integrated and conventional ones. Similar trends were found for the blue–yellow coordinate, b^*, with the yellow component increasing with the maturity of the peppers and from conventional to integrated and from integrated to organic peppers.

Gómez-Ladrón de Guevara and Pardo-González (*23*) studied the changes in color of selected paprika peppers during their ripening. They studied 13 different cultivars and reported chroma values ranging from 15.00 up to 45.12. The values obtained in the present study ranged from 59.01 up to 97.95 for red peppers; these experimental values indicated that the cultivar Almuden provides red peppers with a more intense and vivid color than other cultivars.

Linear regressions among total carotenoid concentrations and the different CIELAB color coordinates, at the different maturation stages and for the different agricultural practices under study, showed that L^* (lightness) was the most suitable attribute ($R^2 = 0.961$) for distinguishing the different concentrations of total carotenoids. In this way, as the total carotenoid concentration increased, the value of L^* decreased, and the peppers became darker.

Summarizing this section, it can be concluded that organic farming will produce more attractive red peppers than other agricultural practices, giving an intense, vivid, and dark red–yellow color.

Total Carotenoid Content and Carotenoid Composition. The total carotenoid content of peppers, expressed as the sum of the carotenoid concentrations measured at 450 nm (**Figure 1**), was significantly ($p < 0.001$) influenced by both developmental stage and agricultural practice.

The synthesis of carotenoid pigments takes place specially during ripening of the pepper, giving the different varieties a final characteristic color. Levels of carotenoids may be affected by maturity, genotype, processing (*24*), and picking time (*25*).

The total carotenoid content of Almuden peppers increased along the development and ripening stages; the experimental values were 124 ± 13 mg kg^{-1} fresh weight (f.w.), 689 ± 173 mg kg^{-1} f.w., and 2468 ± 433 mg kg^{-1} f.w. (values are the means of the three agricultural practices under study) for the immature green, green, and red stages (**Figure 1**). A similar pattern was previously reported by Deli et al. (*26*) when studying the carotenoid composition in the fruits of *Capsicum annuum* cultivar Szentesi Kosszarvú during ripening. These authors reported total carotenoids concentrations of 115, 168, 448, 1327, 6107, and 9947 mg kg^{-1} f.w. for the green, pale yellow, yellow, orange, red, and deep red stages.

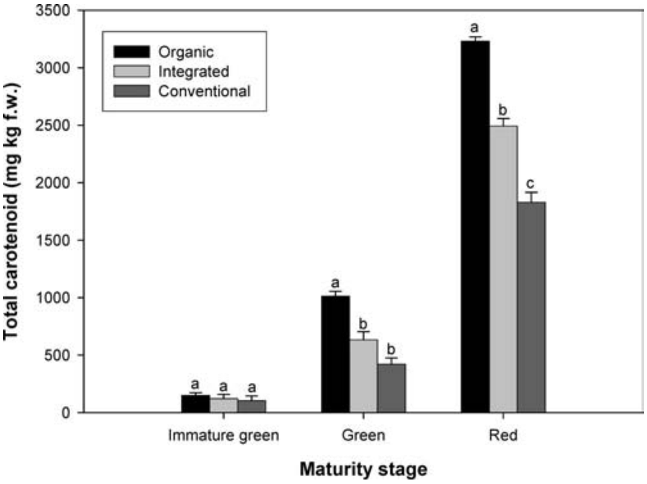

Figure 1. Effects of agricultural practices and ripening stage on total carotenoid contents (mg kg^{-1} fresh weight) of sweet peppers. Data with the same letter in the same maturity stage were not significantly different at $p < 0.001$ (Tukey multiple range test).

Abellán-Palazón et al. (*27*) studied the carotenoid content in two Spanish pepper cultivars (Albar and Negral) and four Hungarian cultivars (KKM-622, Mihályteleki, and Gorogled-6). These authors found mean carotenoid contents of 4470 and 3698 mg kg^{-1} f.w. in fresh pericarp of the Spanish and Hungarian cultivars, respectively. The carotenoid content of red Almuden peppers used in this study (2468 ± 433 mg kg^{-1} f.w.) was slightly lower than those described above because of the seeds, which are largely devoid of pigments (*28*). However, the carotenoid contents found in Almuden fruits were within or above the reported range of other studies; for instance, Guil-Guerrero et al. (*29*) studied the antioxidant activity of 10 Spanish pepper varieties and found total carotenoid contents ranging from 5 mg kg^{-1} f.w. in Green Pricking peppers up to 1031 mg kg^{-1} f.w. in Red Lamuyo peppers, with a mean value of 141 mg kg^{-1} f.w.

Regarding the effects of the agricultural practices under study on the total carotenoid content, no statistically significant differences were found for immature green peppers (124 ± 13 mg kg^{-1} f.w., mean of all treatments). On the other hand, in the last two stages studied, green and red, organic peppers

Table 4. Mean Carotenoid Composition of Red Sweet Peppers (*Capsicum annuum*) cv. Almuden Growing under Organic, Integrated, and Conventional Systems

	agriculture		
pigment	organic concentration (mg kg^{-1} f.w.)	integrated concentration (mg kg^{-1} f.w.)	conventional concentration (mg kg^{-1} f.w.)
capsorubin	255 ± 9[a]	190 ± 7	130 ± 2
violaxanthin	265 ± 11	224 ± 13	185 ± 4
capsanthin	1510 ± 29	1076 ± 32	750 ± 16
cis-capsanthin	353 ± 7	325 ± 9	303 ± 10
zeaxanthin	238 ± 14	220 ± 21	211 ± 12
β-cryptoxanthin	315 ± 23	238 ± 19	165 ± 8
β-carotene	295 ± 8	219 ± 2	85 ± 5
red fraction[b]	2038	1542	1088
yellow fraction	1193	902	639
total[c]	3231	2493	1829

[a] Mean of three replicates ± standard error. [b] Red fraction = capsorubin + capsanthin and isomers. Yellow fraction = violaxanthin + zeaxanthin + β-cryptoxanthin + β-carotene. [c] Total = red + yellow.

Table 5. Minerals Contents (in Fresh Weight) throughout Development and Maturation in Sweet Pepper (*Capsicum annuum*) cv. Almuden Growing under Organic, Integrated, and Conventional Systems

growing stage	agricultural practice	Fe (mg kg^{-1})	Cu (mg kg^{-1})	Mn (mg kg^{-1})	Zn (mg kg^{-1})	Ca (mg kg^{-1})	Mg (mg kg^{-1})	K (g kg^{-1})
immature green	conventional	3.86 ± 0.26 c[a]	0.40 ± 0.07 bc	0.39 ± 0.14 cd	1.96 ± 0.62 c	175 ± 7 d	133 ± 9 d	2.57 ± 0.04 d
	integrated	3.46 ± 0.20 c	0.45 ± 0.03 c	0.19 ± 0.07 a	2.77 ± 0.56 cd	156 ± 3 d	109 ± 8 c	2.62 ± 0.04 d
	organic	3.13 ± 0.84 c	1.17 ± 0.46 d	0.23 ± 0.07 a	5.07 ± 3.91 e	162 ± 10 d	149 ± 18 d	2.26 ± 0.20 d
green	conventional	2.33 ± 0.13 b	0.34 ± 0.02 ab	0.13 ± 0.07 a	0.93 ± 0.34 a	88.1 ± 1.7 b	74.4 ± 3.3 b	1.59 ± 0.02 c
	integrated	2.51 ± 0.14 b	0.38 ± 0.03 b	0.29 ± 0.01 b	1.64 ± 0.47 bc	93.4 ± 3.9 bc	62.8 ± 8.1 a	1.32 ± 0.04 ab
	organic	1.87 ± 0.07 a	0.17 ± 0.04 a	0.50 ± 0.01 d	0.62 ± 0.20 a	58.8 ± 1.5 a	56.2 ± 1.9 a	1.20 ± 004 a
red	conventional	1.70 ± 0.11 a	0.22 ± 0.07 a	0.37 ± 0.01 c	1.35 ± 0.21 b	81.8 ± 3.7 b	62.2 ± 4.4 a	1.45 ± 0.02 b
	integrated	2.16 ± 0.10 b	0.31 ± 0.03 ab	0.15 ± 0.04 a	1.08 ± 0.26 b	86.0 ± 3.6 b	65.1 ± 4.5 ab	1.47 ± 0.04 bc
	organic	2.39 ± 0.10 b	0.44 ± 0.10 c	0.17 ± 0.08 a	1.72 ± 0.87 bc	103 ± 7 c	55.3 ± 3.3 a	1.40 ± 0.01 b
range literature[b]	conventional	2.7–7.5	0.5–1.0	0.5–1.3	1.2–2.6	90–186	100–148	1.69–2.60

[a] Values in this table are the mean ± standard error of three replicates. Data with the same letters were not significantly different at $p < 0.001$ for the attribute evaluated (Tukey multiple range test). [b] Rubio et al. (*31*).

presented significantly ($p < 0.001$) higher contents of total carotenoids than integrated and conventional peppers; no differences were found among integrated and conventional peppers at the green stage. At the last stage, the total carotenoid content was 3231 ± 37 mg kg^{-1} for organic peppers and 2493 ± 65 mg kg^{-1} f.w. and 1829 ± 87 mg kg^{-1} f.w. for integrated and conventional peppers, respectively, implying that organic farming provides peppers with a higher antioxidant activity than other agricultural practices.

Lutein and neoxanthin, both characteristic chloroplast pigments, decreased in concentration with ripening and eventually disappeared (*12*), justifying the fact that these two carotenoids were not found in the present study. β-Carotene, antheraxanthin, and violaxanthin increased in concentration, and other pigments were biosynthesized *de novo*: zeaxanthin, β-cryptoxanthin, capsanthin, capsorubin, capsanthin-5,6-epoxide, and cucurbitaxanthin A (*12*). In the present study, only the carotenoid composition was studied at the red stage. No significant effect of agricultural practice was found (**Table 4**). As previously reported in different studies (*27, 28, 30*), capsanthin was the main carotenoid of red peppers, with capsorubin and *cis*-capsanthin being the other red carotenoids and having concentrations close to 200 mg kg^{-1} f.w., while all yellow carotenoids found, violaxanthin, zeaxanthin, β-cryptoxanthin, and β-carotene, were found at similar concentrations with values slightly over 200 mg kg^{-1} f.w. Capsanthin represented 43.6% of the total carotenoids found in red Almuden peppers. Capsanthin had similar percentages in other studies, for instance, Pérez-Gálvez and Mínguez-Mosquera (*30*) and Marín et al. (*11*) reported capsanthin being 37.8% and 43.6% of the total carotenoid concentration of red pepper fruits, respectively.

Hornero-Méndez et al. (*12*) studied the changes in the biosynthesis of individual carotenoid pigments during fruit ripening of five cultivars of red pepper: Mana, Numex, Belrubi, Delfin, and Negral. These authors concluded, in all the red varieties, that there was an inverse relationship between total carotenoid content and the red-to-yellow isochromic pigment fraction ratio (R/Y) and the capsanthin-to-zeaxanthin ratio (Caps/Zeax). The R/Y ratio increased during fruit ripening; for instance, for the Negral cultivar, the R/Y ratio value started at 0.67 when the green areas were more prevalent in the fruit than the red ones and went up to 1.80 for completely red fruits. The R/Y ratio values for the red peppers ranged from 1.25 (Mana cultivar) up to 1.80 (Negral cultivar). In addition, the cultivar with the highest Caps/Zeax ratio was Numex, with a value of 7.17, while the lowest value was for the cultivar Mana (3.38). The R/Y ratio in Almuden peppers was 1.7, independent of the agricultural practice, at the red stage. This experimental value showed that Almuden fruits had a high intensity of red color. However, the Caps/Zeax ratio was dependent on the agricultural practice, with values being 6.34, 4.88, and 3.55 for organic, integrated, and conventional fruits.

Mineral Content. Certified values for Ca (%), Mg (%), K (%), Cu (mg kg^{-1}), Fe (mg kg^{-1}), Mn (mg kg^{-1}), and Zn (mg kg^{-1}) were 1.81 ± 0.07, 0.65 ± 0.03, 1.38 ± 0.04, 274 ± 10, 9.3 ± 0.5, 45 ± 2, and 37 ± 1, respectively, while measured values for the same elements were 1.80 ± 0.05, 0.64 ± 0.02, 1.40 ± 0.02, 275 ± 11, 9.3 ± 0.3, 46 ± 3, and 37 ± 2, respectively. These results show the goodness of the digestion and spectroscopy analyses of minerals.

Rubio et al. (*31*) studied the mineral composition of red and green peppers from Tenerife Island (Canary Islands, Spain) and showed the influence of the ripening stage on the mineral content of this plant species. These authors concluded that red peppers present higher K, Mg, P, Fe, Cu, Zn, Mn, and B concentrations than green peppers. In the present study, however, red pepper cultivar Almuden presented significantly lower concentrations of Fe, Cu, Zn, K, Ca, and Mg than green peppers. In any case, what it was clear is that the mineral contents depend on the development or ripening stage. Concentrations of minerals in red peppers from the study of Rubio et al. (*31*) were included in **Table 5** to show the normal range of nutrients in this plant species (*Capsicum annuum*); data in this table could be transformed into dry weight considering that the moisture contents were 91.8%, 93.4%, and 93.5% for immature green, green, and red stages, respectively. All macronutrients (Ca, Mg, and K) were within the normal range found in the literature (*31*), even though some of the contents, especially those of red peppers, were close or below the lower limit of this normality range. For instance, the mean K concentration in Almuden peppers (mean value of all agricultural practices and development stages) was 1.38 g kg^{-1} f.w., while the reported normal range was 1.77–2.60 g kg^{-1} f.w. On the other hand, the experimentally found concentrations of some micronutrients (Fe, Cu, and Mn) were low compared with those from the literature (*31*). For instance, the mean Cu concentration in Almuden peppers was 0.28 mg kg^{-1} f.w., while the reported normal range was 0.5–1.0 mg kg^{-1} f.w. These low concentrations of Fe, Cu, and Mn could be due, at least in part, to the effects of the pepper cultivar. In this way, Guil-Guerrero et al. (*29*) studied the nutrient composition of 10 pepper varieties and found a wide range of variability in the nutrient content. For instance, these authors reported that Cu varied from 0.15 mg kg^{-1} f.w. in the Red Italian variety up to 3.03 mg kg^{-1} f.w. in the Green Italian variety. Finally, Zn contents in Almuden peppers were within the normal range reported in the literature (*31*).

The agricultural practice significantly ($p < 0.001$) influenced concentrations of minerals in peppers (**Table 5**). The concentrations of almost all nutrients analyzed in this study were higher in organic peppers compared to those in integrated and conventional products, with integrated products presenting intermediate values. This trend was observed in the first and third development stages (immature green and red) but was not followed in the green stage. For instance, the Fe and Ca contents, at the red stage, increased from 1.70, 2.16, and 2.39 mg Fe kg^{-1} and 81.8, 86.0, and 103 mg Ca kg^{-1}, for conventional, integrated, and organic farming, respectively. Worthington (*5*) reviewed the nutritional quality of organic versus conventional fruits, vegetables, and grains and concluded that there appears to be genuine differences in the nutrient content of organic and conventional crops. This author stated that organic crops contained significantly more vitamin C, Fe, Mg, and P and significantly less nitrates than conventional crops. In addition, Pérez-López et al. (*7*) studied the effect of organic farming on the mineral content of Clemenules mandarin juice and concluded that higher concentrations of all studied minerals (Fe, Cu, Mn, Zn, Ca, Mg, K, and Na) were found in the organic juice.

Conclusions. It can be concluded that the agricultural practice (conventional, integrated, and organic) had a significant effect on red and yellow colors, mineral content, and total carotenoid content in sweet peppers (*Capsicum annuum* cv. Almuden). Organic farming had a positive effect on all studied quality parameters of peppers, including intensities of red–yellow color, mineral content, and total carotenoid content, with the latter implying a higher antioxidant activity of organic vegetables. Integrated agriculture presented intermediate results between organic and conventional agricultures, but without any doubt, integrated peppers have better quality than conventional ones.

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