

Impact of Cultivation Conditions, Ethylene Treatment, and Postharvest Storage on Selected Quality and Bioactivity Parameters of Kiwifruit “Hayward” Evaluated by Analytical and Chemometric Methods

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Organic, semiorganic, and conventional “Hayward” kiwifruits, treated with ethylene for 24 h and stored during 10 days, were assessed by UV spectrometry, fluorometry, and chemometrical analysis for changes in selected characteristics of quality (firmness, dry matter and soluble solid contents, pH, and acidity) and bioactivity (concentration of polyphenols via Folin-Ciocalteu and *p*-hydroxybenzoic acid assays). All of the monitored qualitative parameters and characteristics related to bioactivity were affected either by cultivation practices or by ethylene treatment and storage. Results obtained, supported by statistical evaluation (Friedman two-way ANOVA) and chemometric analysis, clearly proved that the most significant impact on the majority of the evaluated parameters of quality and bioactivity of “Hayward” kiwifruit had the ethylene treatment followed by the cultivation practices and the postharvest storage. Total concentration of polyphenols expressed via *p*-hydroxybenzoic acid assay exhibited the most significant sensitivity to all three evaluated parameters, reaching a 16.5% increase for fresh organic compared to a conventional control sample. As a result of postharvest storage coupled with ethylene treatment, the difference increased to 26.3%. Three-dimensional fluorescence showed differences in the position of the main peaks and their fluorescence intensity for conventional, semiorganic, and organic kiwifruits in comparison with ethylene nontreated samples.

Kiwifruits are popular mostly because of their unique taste and aroma. In addition to other components, they contain several constituents that are responsible for high quality, which include polyphenols (1). To reach the ripening stage, kiwifruit can be treated with a number of substances. A common procedure is to use different concentrations of spermine and spermidine and then to store the kiwifruit in ambient conditions for 15 days (2). Additional experiments were performed with 1-methylcyclopropene (1-MCP). The effect of different concentrations of 1-MCP on the postharvest life and quality of *Actinidia deliciosa* “Allison” kiwifruit was studied (3).

In some reports, new kiwifruit cultivars were treated with 1-MCP at room temperature for 16 h, and then this effect was studied during the cold storage at 2°C and during shelf life at room temperature (4–6). Hardy kiwifruits (*Actinidia arguta*) were treated with 20 µL/L 1-MCP for 16 h at 10°C and were subsequently stored at 1 ± 0.5°C (7). The influence of controlled atmospheres on the postharvest quality and storage stability of “Hongyang” kiwifruit at 2°C was investigated (8). “Qinmei” kiwifruits were treated with 0.5 µL/L 1-MCP for 24 h during the period of precooling. After 50 days of cold storage at 0°C, fruits were returned to room temperature for 7 days. The fruits were then treated by ethylene, salicylic acid, and methyl jasmonate. Results showed that all these treatments promote normal ripening of kiwifruit treated by 1-MCP (9). Kiwifruits cold stored for 1 month were treated with 200 µL/L NO and were subsequently transferred to room temperature to monitor quality changes (10). Cold storage (1.5°C) for two or four months of fruit matched for firmness and soluble solids concentration. It resulted in a significant reduction in aroma-related esters such as methyl/ethyl propanoate, methyl/ethyl butanoate and methyl/ethyl hexanoate. Levels of these esters, however, were restored by ethylene treatment (100 ppm, 24 h) before ripening (11).

Interest in organically grown (OG) fruits is increasing nowadays in response to intensive agricultural practices and

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their effects on human health as well as on the environment (12). Many consumers prefer to buy organic foods because of their alleged greater nutritional benefits. Nevertheless, the current literature and the results of the published studies on this topic are rather ambiguous regarding the accuracy of this claim about nutritional value (13). Different types of fruits were compared from nutritional and antioxidant-component points of view (14), including organic and conventional oranges, limes, and apples. In general, small differences were observed in the chemical quality parameter, however, organic fruits showed higher antioxidant activity than conventional fruits, probably because of modulation of these parameters. The results suggest that other factors are more influential on nutrient content, particularly vitamin C levels, than whether fruits are OG or conventionally grown (CG). The authors claimed (15) that the organic system helped to increase the pH of acid soils and to improve the availability of nutrients for the plant. OG and CG kiwifruit, mangos, acerolas, persimmons, and strawberries were compared (16–18). There is a scarcity of reports about the ethylene treatment and storing effects on selected quality parameters of kiwifruit produced under different cultivation systems. Therefore, the aim of this research was to evaluate these effects-cultivation conditions, ethylene treatment, and postproduction storage for 10 days—on physicochemical properties related to quality and bioactivity of kiwifruit cultivar “Hayward” by new analytical methods and statistical processes.

Experimental

Preparation of Fruit Samples

Kiwifruit cultivar “Hayward” was grown under conventional, low-chemical, and organic conditions in the orchard Heanam County, Jeonnam Province, Korea. The harvest date was October 30, 2014, and October 18, 2015. The fruits were treated with ethylene and then ripened at 20°C in the growth chamber (Percival Scientific, Perry, IA) for 10 days. The samples were treated with liquid nitrogen to prevent oxidation of phenolic compounds and then were freeze-dried as previously described (17, 19, 20).

Reagents and Chemicals

Folin–Ciocalteu (FC) phenol reagent was purchased from Sigma Chemical Co. (St Louis, MO). Sodium nitrite, acetic acid, and hydrochloric acid were purchased from Samchun Chemical Co. (Korea). The latter chemicals were of analytical grade purity.

Determination of Quality Parameters

The parameters of quality and bioactivity, firmness, SSC, total acidity (TA), pH, sensorial profile, and polyphenol concentration have been evaluated in the study. For firmness determination, the penetration force in kilograms was measured, using a fruit firmness tester (Model KM; Fruit Test Tech, Japan). The mean values of the firmness were expressed as newtons (N). The peeled fruits were homogenized and filtered through cheesecloth to obtain a clear juice for determination of SSC (°Brix), pH, and TA. The SSC was measured using a refractometer (Atago Co. Ltd, Tokyo, Japan), and pH with a pH meter. The TA was measured in 4 mL of juice, diluted to 20 mL of distilled water, and titrated with 0.1 N NaOH. The TA

was expressed as a percentage of citric acid. Twelve qualified panelists assessed sensory quality in a sensory laboratory. Taste quality was evaluated by affective test of appearance, taste (sweetness, sourness, flavor), and total acceptance in the Hedonic scale method, which was 1–5 rating scale (1 = severely bad, 2 = bad, 3 = moderate, 4 = good, 5 = excellent). Total polyphenols (TP) were extracted with methanol (concentration 20 mg/mL) during 1 h in a cooled ultrasonic bath (21). TP [mg gallic acid equivalents (GAE)/100 g FW)] were determined by the FC and Prussian Blue (PB) assays (PBA; 22, 23) with absorbance measurements at 765 and 700 nm, respectively, using a spectrophotometer (Model 8452A; Hewlett-Packard, Rockville, MD).

Fluorometric Measurements

Two-dimensional fluorescence (2D–FL) measurements for kiwifruit extracts at a concentration of 0.01 mg/mL were recorded on a Model FP-6500, Jasco spectrofluorometer (Serial No. N261332; Japan) equipped with 1.0 cm quartz cells and a thermostat bath. The 2D–FL was taken at emission wavelengths from 300 to 750 nm; and at excitation of 280 nm. The three-dimensional spectra (3D–FL) were collected with subsequent scanning emission spectra from 200 to 795 nm at 1.0 nm increments by varying the excitation wavelength from 200 to 500 nm at 10 nm increments. Measurement procedure and data evaluation are described in details in (19, 20).

Statistical Analysis

Two-way ANOVA (Friedman test) as well as multivariate statistical analysis were performed, using principal component analysis (PCA), canonical discriminant analysis (CDA), and factor analysis (FA) using the statistical package Unistat v. 6.0 (Unistat, London, United Kingdom), in which all the statistical calculations and data visualizations were performed for considering the effects of production conditions and ethylene treatment on selected physicochemical and sensory properties of the “Hayward” kiwifruit. Origin (Microcalc, Northampton, MA) was also used for data visualization in plots, if not specified otherwise.

Results and Discussion

Effect of Ethylene Treatment and Storage on Qualitative Indices of Kiwifruit

During the treatment with ethylene for 24 h and storage for 10 days, the following changes appeared in conventional, semiorganic, and organic kiwifruit. The firmness (N) of ethylene-treated samples and of samples stored for 10 days has changed from 27.84 ± 1.99 to 5.66 ± 0.93 ; from 30.6 ± 1.98 to 9.83 ± 0.3 ; and from 27.84 ± 1.99 to 7.74 ± 0.24 for conventional, semiorganic, and organic kiwifruit, respectively. After ethylene treatment and storage, the values of dry matter (expressed in %) have changed for conventional, semiorganic, and organic kiwifruit to 16.88 ± 0.44 ; 17.11 ± 0.62 ; and 16.32 ± 0.17 , respectively. The sensory value (score) at the beginning was from 1.83 ± 0.26 to 3.6 ± 0.53 ; from 2.4 ± 0.2 to 3.67 ± 0.29 ; and from 2.57 ± 0.12 to 3.42 ± 0.14 for conventional, semiorganic, and organic kiwifruit, respectively. After the treatment and storage, the values were 4.43 ± 0.12 ; 4.67 ± 0.29 , and 3.61 ± 0.53 for conventional, semiorganic and organic fruits, respectively.

Figure 1 shows the combined effect of ethylene treatment, growing conditions, and time on individual proximate characteristics of kiwifruit (firmness, dry matter, and sensorial properties). The soluble solid contents ($^{\circ}$ Brix) before the treatment for conventional, semiorganic, and organic fruits were from 8.89 ± 0.09 to 12.51 ± 0.02 ; from 8.32 ± 0.16 to 12.9 ± 0.1 , and from 8.6 ± 0.44 to 13.07 ± 0.12 , but after the treatment the values changed to 13.37 ± 0.15 ; 13.21 ± 0.37 ; and 13.51 ± 0.02 , respectively. The acidity (%) was from 1.63 ± 0.08 to 0.75 ± 0.06 ; from 1.54 ± 0.04 to 0.77 ± 0.06 ; and from 1.65 ± 0.14 to 0.73 ± 0.14 for conventional, semiorganic, and organic fruits, respectively. The values changed after the treatment to 0.55 ± 0.05 ; 0.56 ± 0.07 ; and 0.75 ± 0.06 , respectively. The pH values during the storage were from 3.22 ± 0.11 to 3.37 ± 0.09 ; from 3.23 ± 0.1 to 3.51 ± 0.01 ; and from 3.35 ± 0.1 to 3.6 ± 0.22 , and after treatment were 3.66 ± 0.12 ; 3.44 ± 0.05 ; and 3.37 ± 0.09 for conventional, semiorganic, and organic fruits, respectively (Table 1). The final results of total phenolics differed for conventional, semiorganic, and organic kiwifruit (Table 2). After treatment, polyphenols increased in all kiwifruit samples. The amount of polyphenols (mg GAE/100 g FW) was the highest in OG kiwifruits (163.11 ± 2.96) in comparison with the CG (154.18 ± 7.92). The values of polyphenols by PBA were slightly higher, but the proportion was the same as by the FC method (183.43 ± 10.71 versus 216.62 ± 3.60).

Our report (17) showed that when CG and OG “Hayward” kiwifruits were exposed to ethylene treatment for 24 h, followed by storage at 20°C for 10 days, firmness gradually decreased. Firmness depended on storage time, regardless of cultivation type. The rate of softening was slightly higher in OG than in CG fruits. The sensory value of kiwifruit increased with reduced firmness and did not vary among cultivation types. The rate of softening was slightly higher in OG than in CG fruits. The sensory value of kiwifruit increased with reduced firmness and did not vary among cultivation types. SSC increased with the storage time, whereas acidity decreased in all fruits. Significant differences between treated conventional and organic kiwifruits were found in polyphenols (189.98 ± 12.75 and 219.43 ± 15.73 mg GAE/100 g FW) in the last day of treatment.

Kiwifruit samples cultivated by conventional, semiorganic, and organic methods, treated with ethylene and stored 10 days at 20°C were examined for quality parameters: firmness, dry matter, sensorial parameter, SSC, acidity, pH, and total phenolics (FC; PBA). In the case of firmness, dry matter content, and sensorial evaluation, multiple comparisons by Friedman two-way ANOVA test denoted some significantly different pairs ($P < 0.05$) between kiwifruit related to different producing systems (Table 3). No significant differences among kiwifruit-producing systems were found in the case of the remaining characteristics. Comparison of ethylene-treated “Hayward” kiwifruit farmed differently in conditions of

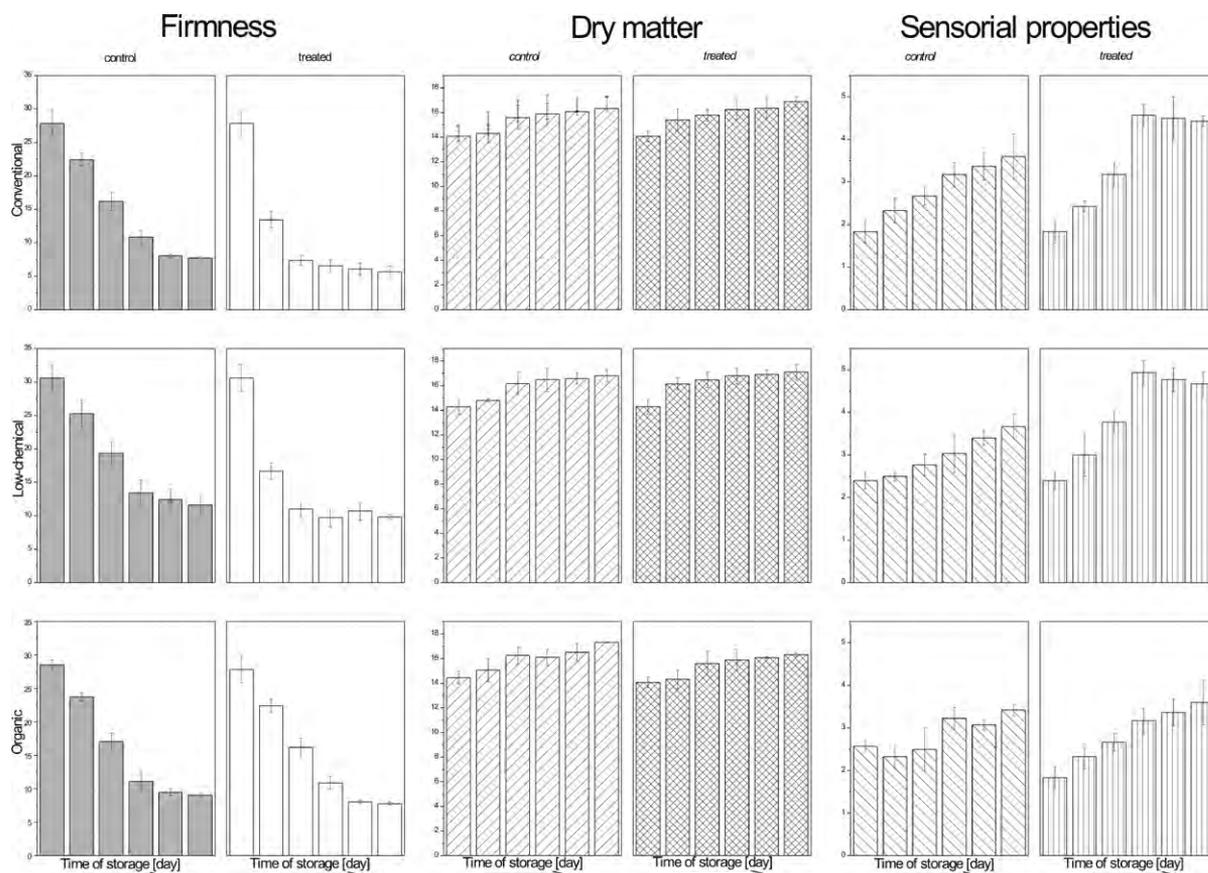


Figure 1. The effects of ethylene treatment (control versus treated), growing conditions (conventional versus semiorganic versus organic), and time of storage on selected proximate characteristics [firmness (N), dry matter (%), sensorial properties (score)] of “Hayward” kiwifruit.

Table 1. Changes in fruit SSC^a, acidity^b, and pH values of conventional, semiorganic, and organic “Hayward” kiwifruit during storage

Cultivar	Ethylene treatment	Days of storage, 20°C					
		0	2	4	6	8	10
		SSC ^c					
Conventional	Control	8.89 ± 0.09 ^{abA}	9.97 ± 0.23 ^{bbB}	11.8 ± 0.33 ^{ccC}	12.5 ± 0.32 ^{ddD}	12.23 ± 0.02 ^{cdD}	12.51 ± 0.02 ^{ddD}
	Treated	9.89 ± 0.09 ^{abA}	11.63 ± 0.23 ^{ccC}	11.93 ± 0.12 ^{ccC}	12.63 ± 0.15 ^{ddD}	12.87 ± 0.12 ^{ddD}	13.37 ± 0.15 ^{eeE}
Semiorganic	Control	8.32 ± 0.16 ^{abA}	9.52 ± 0.17 ^{bbB}	11.73 ± 0.12 ^{ccC}	12.13 ± 0.12 ^{ccC}	12.45 ± 0.18 ^{ddD}	12.91 ± 0.13 ^{bdD}
	Treated	9.02 ± 0.16 ^{abA}	11.93 ± 0.17 ^{ccC}	12.47 ± 0.06 ^{ddD}	12.73 ± 0.12 ^{ddD}	12.85 ± 0.05 ^{edD}	13.21 ± 0.37 ^{eeE}
Organic	Control	8.62 ± 0.44 ^{abA}	9.83 ± 0.25 ^{bbB}	11.9 ± 0.17 ^{ccC}	12.16 ± 0.08 ^{ccC}	13.02 ± 0.52 ^{eeE}	13.07 ± 0.12 ^{edD}
	Treated	8.79 ± 0.09 ^{abA}	9.93 ± 0.23 ^{bbB}	12.14 ± 0.31 ^{ccC}	12.52 ± 0.34 ^{ddD}	13.23 ± 0.02 ^{cdD}	13.51 ± 0.02 ^{ddD}
		Acidity					
Conventional	Control	1.63 ± 0.08 ^{abA}	1.44 ± 0.04 ^{bbB}	1.28 ± 0.06 ^{ccC}	1.14 ± 0.05 ^{ccC}	0.98 ± 0.07 ^{ccC}	0.75 ± 0.06 ^{ddD}
	Treated	1.43 ± 0.07 ^{abA}	1.24 ± 0.09 ^{ccC}	1.06 ± 0.08 ^{ccC}	0.75 ± 0.06 ^{ddD}	0.65 ± 0.04 ^{ddD}	0.55 ± 0.05 ^{eeE}
Semiorganic	Control	1.54 ± 0.04 ^{abA}	1.38 ± 0.04 ^{bbB}	1.34 ± 0.03 ^{bbB}	1.18 ± 0.06 ^{ccC}	0.97 ± 0.06 ^{ccC}	0.77 ± 0.06 ^{ddD}
	Treated	1.34 ± 0.14 ^{abA}	1.31 ± 0.14 ^{bbB}	1.15 ± 0.09 ^{ccC}	0.91 ± 0.07 ^{ddD}	0.76 ± 0.06 ^{ddD}	0.56 ± 0.07 ^{eeE}
Organic	Control	1.65 ± 0.14 ^{abA}	1.42 ± 0.17 ^{bbB}	1.35 ± 0.12 ^{bbB}	1.25 ± 0.12 ^{bbB}	0.93 ± 0.09 ^{ccC}	0.73 ± 0.14 ^{ddD}
	Treated	1.33 ± 0.08 ^{abA}	1.40 ± 0.04 ^{bbB}	1.28 ± 0.06 ^{bbB}	1.10 ± 0.05 ^{ccC}	0.88 ± 0.07 ^{ccC}	0.51 ± 0.06 ^{ddD}
		pH					
Conventional	Control	3.22 ± 0.11 ^{abA}	3.31 ± 0.19 ^{abA}	3.43 ± 0.03 ^{abA}	3.55 ± 0.08 ^{bbB}	3.72 ± 0.03 ^{ccC}	3.37 ± 0.09 ^{abA}
	Treated	3.25 ± 0.11 ^{abA}	3.35 ± 0.09 ^{abA}	3.44 ± 0.02 ^{abA}	3.65 ± 0.13 ^{bbB}	3.55 ± 0.06 ^{bbB}	3.66 ± 0.12 ^{ccC}
Semiorganic	Control	3.23 ± 0.13 ^{abA}	3.38 ± 0.07 ^{abA}	3.33 ± 0.03 ^{abA}	3.42 ± 0.08 ^{abA}	3.4 ± 0.13 ^{abA}	3.51 ± 0.01 ^{baA}
	Treated	3.24 ± 0.17 ^{abA}	3.43 ± 0.12 ^{abA}	3.25 ± 0.03 ^{abA}	3.55 ± 0.11 ^{bbB}	3.31 ± 0.05 ^{abA}	3.44 ± 0.05 ^{abA}
Organic	Control	3.35 ± 0.14 ^{4abA}	3.33 ± 0.07 ^{abA}	3.52 ± 0.04 ^{bbB}	3.43 ± 0.03 ^{abA}	3.72 ± 0.03 ^{ccC}	3.61 ± 0.22 ^{acC}
	Treated	3.43 ± 0.11 ^{abA}	3.41 ± 0.19 ^{abA}	3.43 ± 0.03 ^{abA}	3.55 ± 0.08 ^{bbB}	3.58 ± 0.03 ^{ccC}	3.67 ± 0.09 ^{abA}

^a SSC = Soluble solids content, °Brix;

^b Acidity = % of citric acid.

^c Lower case letters in superscripts within column denote statistically significant differences resulting from growing conditions/treatment, whereas capital letters in superscripts within rows denote statistically significant differences following from storage. Tests were performed by Duncan's ANOVA multiple-range test. Values of means not sharing the same letters are significantly different from one another ($P = 0.95$; $n = 10$).

conventional, semiorganic input, and organic production systems are graphically reported by canonical discrimination in Figure 2.

Two-Way ANOVA Friedman Test (Tukey's HSD)

The Friedman test is a nonparametric statistical test that was used to detect the differences in treatments for multiple test

attempts. It, therefore, can possess an answer regarding the significance (at $P < 0.05$) of the results (qualitative characteristics) between the control and the treated groups as well as a consideration of the significance of the differences resulting from storing or from the method of production. When the presented characteristics of quality and bioactive properties (BAP) of kiwifruit are evaluated in terms of the effect of the growing conditions (affiliation to respective production systems), significant ($P < 0.05$) and even

Table 2. Changes in fruit total phenol concentration expressed via Folin-Ciocalteu (FC) and *p*-hydroxybenzoic acid (PBA) assays, determined for conventional, semiorganic, and organic “Hayward” kiwifruit during storage

Cultivar	Ethylene treatment	Days of storage, 20°C					
		0	2	4	6	8	10
		FC ^a					
Conventional	Control	125.72 ± 4.22 ^{abA}	126.42 ± 3.68 ^{abA}	127.14 ± 3.31 ^{abA}	130.35 ± 4.41 ^{abA}	131.34 ± 2.21 ^{abA}	132.25 ± 2.96 ^{abA}
	Treated	137.14 ± 4.22 ^{abA}	138.45 ± 1.68 ^{bbB}	140.15 ± 7.50 ^{abB}	143.72 ± 7.68 ^{cbB}	152.14 ± 7.93 ^{dcD}	154.18 ± 7.92 ^{dcD}
Semiorganic	Control	129.47 ± 6.00 ^{abA}	130.06 ± 10.64 ^{abA}	131.74 ± 2.57 ^{abA}	133.06 ± 1.37 ^{abA}	134.25 ± 2.89 ^{abA}	136.12 ± 9.50 ^{baA}
	Treated	141.13 ± 6.00 ^{abA}	145.34 ± 8.58 ^{baA}	150.81 ± 12.82 ^{abA}	154.71 ± 1.58 ^{baA}	155.38 ± 10.49 ^{dcD}	158.12 ± 13.67 ^{dcD}
Organic	Control	133.61 ± 10.38 ^{abA}	134.18 ± 2.98 ^{baA}	135.31 ± 0.72 ^{abA}	136.45 ± 3.02 ^{cbB}	137.02 ± 2.53 ^{abA}	140.29 ± 5.04 ^{abA}
	Treated	145.56 ± 4.22 ^{abA}	151.14 ± 3.68 ^{baA}	155.11 ± 3.31 ^{abA}	158.21 ± 4.41 ^{abA}	161.24 ± 2.21 ^{abA}	163.11 ± 2.96 ^{abA}
		PBA					
Conventional	Control	146.62 ± 2.53 ^{abA}	147.82 ± 4.14 ^{abA}	149.85 ± 4.00 ^{abA}	151.95 ± 6.03 ^{abA}	152.26 ± 1.36 ^{abA}	158.45 ± 3.6 ^{abB}
	Treated	159.82 ± 2.53 ^{abA}	162.24 ± 7.46 ^{abA}	169.14 ± 10.07 ^{abA}	176.14 ± 6.56 ^{ccC}	178.60 ± 7.37 ^{ddD}	183.43 ± 10.71 ^{deE}
Semiorganic	Control	153.44 ± 2.06 ^{abA}	154.25 ± 6.05 ^{abA}	155.25 ± 4.08 ^{abA}	157.14 ± 5.04 ^{abA}	159.18 ± 6.91 ^{abA}	161.11 ± 5.45 ^{abA}
	Treated	167.72 ± 2.06 ^{abA}	172.18 ± 4.18 ^{abA}	175.42 ± 7.10 ^{bbB}	183.25 ± 10.04 ^{ccC}	194.43 ± 7.59 ^{ddD}	204.61 ± 7.00 ^{ddD}
Organic	Control	161.28 ± 3.11 ^{baA}	162.81 ± 3.09 ^{baA}	163.33 ± 7.10 ^{abA}	165.24 ± 5.08 ^{abA}	168.12 ± 8.62 ^{abA}	170.95 ± 1.05 ^{abB}
	Treated	175.79 ± 2.53 ^{abA}	180.41 ± 4.14 ^{abA}	186.48 ± 4.00 ^{abA}	192.23 ± 6.03 ^{abA}	204.41 ± 1.36 ^{abA}	216.62 ± 3.60 ^{abB}

^a In both cases, the results are expressed as mg GAE/100g FW (gallic acid equivalents); Different lower case letters in superscripts within columns denote statistically significant differences following from growing conditions/treatment; whereas different capital letters in superscripts within rows denote statistically significant differences following from storage. Tests were performed by Duncan's ANOVA multiple range test. Values of means not sharing the same letters are significantly different from one another ($P = 0.95$; $n = 10$).

Table 3. Statistically significant ($P < 0.05$) differences between kiwifruit's production systems performed by Friedman two-way ANOVA Tukey's honest significant difference (HSD) test

(a) Without respect of treatment	
Parameter	Significantly different pairs, $P < 0.05^a$
Firmness	SO-C, O-C
Dry matter content	SO-C
Sensorial parameter	SO-O
(b) Control (nontreated) and treated samples assessed separately	
Dry matter content	SOO
Sensorial parameter	SO-O
Firmness	SO-C
Acidity	O-C
Total phenols (FC)	O-C
Total phenols (TBA)	O-C

^a Producing systems: SO = Semiorganic; O = organic; C = conventional.

very significant ($P < 0.001$) differences were confirmed with the exception of dry matter content, firmness, and sensorial characteristics. If the datasets are considered separately for treated and nontreated (control) samples in terms of differences between the characteristics of samples of the different production system, then also significant differences between the CG, semiorganic, and OG kiwifruit samples were confirmed for dry matter content, sensorial properties, firmness, acidity, and total phenols assessed by both FC and PBA assays. In addition to the effects of the growing conditions, the effects of ethylene treatment and storage on the monitored characteristics were also evaluated by ANOVA and Duncan's multiple range test, as is indicated in Tables 1–3.

Processing of Data by Multivariate Statistics: Canonical Discriminant Analysis

Effect of ethylene treatment.—The effect of ethylene treatment on sample properties cannot be visualized, as there is only one discriminant function (DF; separating treated and nontreated samples). However, in spite of this, the overall differentiation of treated and nontreated samples reached 72% with a total of 10 misclassified samples (4 control samples classified as treated, while 6 treated samples were classified as control), which indicates that the properties of samples are meaningfully affected by the ethylene treatment; however, because of the diversity of the results (disparity of the values of individual characteristics used for discrimination), the classification score is not absolute (Figure 2). Based on the values of standardized canonical correlation coefficients, dry matter content, sensorial characteristics, and soluble solids content with coefficients (scores) of 2.5, 1.4, and 1.2 played the dominant role in differentiation (discrimination function construction).

Effect of the growing conditions.—In the case of discrimination according to the growing conditions, the discrimination score reached almost 81%, indicating partially successful differentiation, as is finally apparent also from the plot of discrimination functions presented in Figure 2a.

To discriminate three different kinds of samples (organic, low-chemical and conventional), 2 discriminant functions were created. Function 1 may be represented by the highest discrimination power of firmness (values of canonical discrimination coefficient, 4.8), followed by sensorial values and PBA (3.4 and 3.2, respectively). On the other hand, function 2 is characterized by the dominant role of the dry matter content, SSC, and PBA (Figure 2a).

Effect of storage.—Results of the discriminant analysis focused on the effects of the long-term storage of kiwifruit samples (Figure 2b), which indicated that the kiwifruit samples are affected by the storage in a way comparable to the growing conditions. The overall discrimination score based on eight qualitative characteristics (firmness, dry matter content, sensorial characteristics, SSC values, acidity, pH, and content of polyphenols expressed either via FC or PBA assays) reached 86.2%, with 5 misclassifications of 36 cases. As it is obvious from the plot of the discriminant scores, absolute classification was reached for fresh samples (0) as well as for samples stored for 10 days. In two other cases, the discrimination was lower, reaching 50% (6 days) and 66.6% (2 days), respectively. The role of individual characteristics in the discrimination process, acidity, and the concentration of polyphenols assessed by FC assay had the dominant role in the first DF construction, whereas in the second DF, acidity followed by sensorial characteristics showed the highest standardized canonical coefficient values. However, it is apparent from the comparison of all three discriminations (ethylene treatment, growing conditions, and storage) that storage up to 10 days has the most significant impact on samples' qualitative properties, even more significant than the ethylene treatment or the growing conditions.

Principal Component Analysis

PCA was used to explain the variability of the dataset of individual experimental characteristics related to the quality of kiwifruit and to visualize the hidden relationships between the individual characteristics from the way of production, ethylene treatment, and the effects of storing conditions. From the results of the analysis (data not depicted), it clearly follows that there is only a partial grouping tendency according to the affiliation of individual samples to the production system or treatment, typically observed for samples stored for 8 and 10 days. However, according to the results of CDA presented earlier (Figure 2a and b), the tendency toward grouping the PCs according to the time of storage is apparent. First, two of a total of eight PCs cumulatively explain approximately 89% of the overall variability; the addition of the third PC would increase the score to 94.5%. Regarding the role of individual characteristics in the variability explanation, component 1 arises dominantly from acidity, dry matter content, and sensorial parameters (eigenvectors coefficients at 0.4 each), whereas in the second component, the variability is mainly described by the concentration of polyphenols (FC or PBA assays, eigenvectors coefficients 0.58 and 0.52, respectively), and in the third by pH values with eigenvector of 0.82. Separation of individual components into two main groups of vectors indicates that, from a time-of-storage viewpoint, properties of samples freshly prepared and those stored for up to 4 days are comparable (although gradual changes from samples stored for

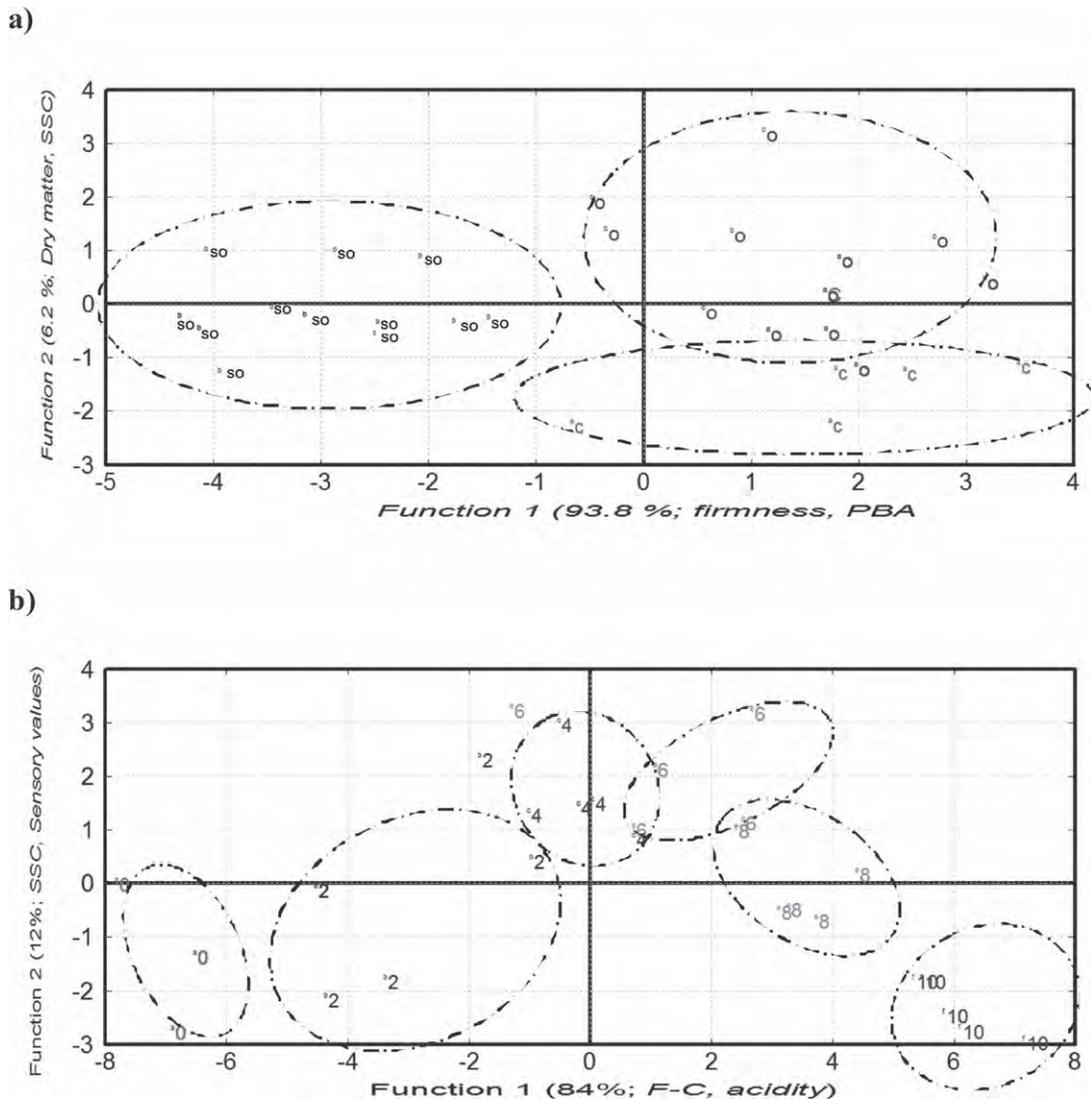


Figure 2. CDA of “Hayward” kiwifruit samples according to the (a) growing conditions—conventional (C), semiorganic (SO), and organic (O); (b) time of storage for up to 10 days. The entire dataset of eight qualitative characteristics, comprising firmness, dry matter content, sensorial characteristics, soluble solids content (SSC), values, acidity, pH, and content of polyphenols (expressed via Folin-Ciocalteu and PBA assays) were used for discrimination. % in brackets and parameters explain overall discrimination score of individual function and the dominant role of parameters in individual discrimination functions, respectively.

0–2–4 days are apparent), and that the significant change in this trend could be identified in fourth to sixth day of storage, after which the samples are relatively similar in their properties, or they change practically independently on time of continuous storage. It is also evident that in the initial phase of storage (0–3 days), there is practically no difference between samples in terms of the growing conditions. Part of PCs belonging to both treated and nontreated samples (not depicted) of organic origin stored for 4–10 days differs from the rest by their position in the opposite quadrants of the plot. In other cases, differences among the samples resulting from the growing conditions could be assigned as statistical.

Principal Component Factoring (PCF)

The influence of individual experimental characteristics and its contribution to the overall variability in terms of correlation (noncorrelation) with the other parameters was assessed by PCF analysis. PCF was performed in Equimax orthogonal

rotation, which minimizes the number of variables that have high loadings and also minimizes the number of factors needed to explain a variable (data not depicted). From the results of this analysis, it is apparent that there exist two main groups of factors, forming partially independent clusters. The first cluster is formed by the firmness and acidity with the highest discrimination potential. The second cluster comprises of the rest of the characteristics, with partially separated polyphenolic assay characteristics. It is apparent from the analysis of these five aspects that, as a result of treatment, the physicochemical and sensorial properties are the most affected. According to the results of ANOVA analysis, PCP and SP are dominantly affected by the time of cold storage, whereas the other factors did not lead to significant differences. In cases of BAP, antioxidant properties (AP), and binding properties (BP), ethylene treatment was identified as key for the differences in these characteristics. Only in the case of AP, the significant influence of the growing conditions was evident (Figure 3). Differentiation of kiwifruits on the basis of five parameters and

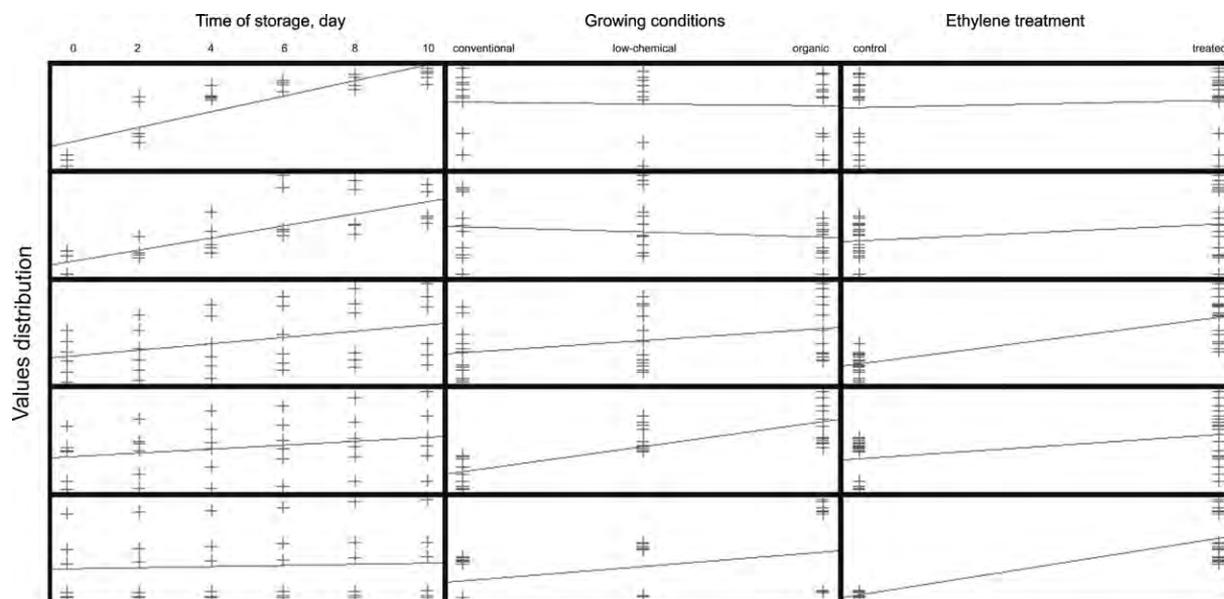


Figure 3. Changes of physicochemical, sensorial, BAP, AP, and BP of “Hayward” kiwifruit resulting from three independent factors: way of production, time of cold storage, and ethylene treatment.

three factors by means of CDA fully correspond with the results of ANOVA analysis. When differentiated according to the time of storage, only moderate, 67% correctness of classification was reached, with dominant roles of PCP in the discrimination function. The discrimination of samples according to the way of production increased the correctness to 97.5%, identifying AP as the most important characteristic (20). In fact, only one in 30 cases was misclassified as being low chemical instead of organic. Discrimination of samples on the basis of ethylene treatment was absolute, with the dominant role of all three factors identified by ANOVA, of which BP was recognized as the most important. The analysis performed by PCA also fully supported the hypothesis, that the effects of treatment on individual characteristics and so, on kiwifruit properties is as follows: Ethylene treatment →→ Growing conditions → Time of storage. AP are most affected parameter by these three factors, do not reflecting sufficiently only the time of storage.

Fluorescence Studies

Two-dimensional fluorescence spectra correlated with the results described in the following text. Three-dimensional fluorescence of conventional, semiorganic, and organic kiwifruit shows the differences in the main peaks and their fluorescence intensities (FIs; Figures 4 and 5). At emission of the wavelengths from 327 nm to 338 nm, spectra of organic treated kiwifruit with the highest FI were recorded [Table 4A, Figure 5A, 333, 83, 54, 61, and 67 arbitrary units (A.U.), the first line from the top]. The lowest FI was in the conventional control sample (Table 4A, Figure 4A with 151, 59, 36, and 36 A.U., the lowest line). The change in the FI of the sample is in direct correlation with the BP of HSA before and after its interaction with extracted polyphenols. The FIs of organically treated to conventionally treated samples was about 1.68 times (lines 1, 3, Figure 4A) and for organic control to conventional control (lines 4, 5, Figure 4A) was 1.08 times. Similar correlations in the FI between organically treated and conventionally treated samples (about

1.67 times) were also found in three-dimensional fluorescence peak a (Table 4B, Figures 4B, 4Bb, and 5E, 5Ee). Such correlation between organic control to conventional control was 1.19 times (Table 4B, Figures 4C, 4Cc, and 5F, 5Ff). The main two peaks a and b were in organic treated sample of 255 and 283 A.U. with the highest FI (Table 4B, Figure 4B, 4Bb). The lowest FI was for conventional control (Table 4B, Figure 5F, 5Ff with 112 and 138 A.U.). Caffeic acid as a standard corresponded with similar peaks of organically treated samples. The changes in the FI of treated and untreated samples corresponded with the changes in the amount of polyphenols (Table 3). The present results differ from those discussed for the harvest of 2012 (17). The observed peaks by three-dimensional fluorescence showed differences in the position of the main peaks and their FI for conventional and organic kiwifruits in comparison with non-ethylene-treated samples.

In conclusion, ethylene treatment increased the bioactivity of organic, semiorganic, and conventional kiwifruits (17). In recent years, there has been growing interest in the influence of sustainable cultivation systems on the biochemical quality of vegetables and fruits. The changes in physiological parameters of “Qinmei” kiwifruit with the extension of storage time under the condition of storage at 20°C are in line with our results (24). The firmness and TA of fruits was increased. Other reports showed that harvested kiwifruits of a new variety of *Actinidia eriantha Benth* “Walter” were stored at 20°C. The fruit firmness and TA decreased sharply in the first 6 days and then maintained a relatively stable level. The SSC increased at the early period of storage and then decreased gradually (25). The skin and flesh firmness was higher in organic kiwifruits than in conventional ones. Results from this study showed that the physicochemical properties and color characteristics of organic and conventional kiwifruits were affected by storage periods (26). Polyphenols were significantly higher in ethylene-treated samples than in nontreated, and were significantly higher in organic samples than in conventional, except for some data from the sixth and eighth days of treatment (Table 2).

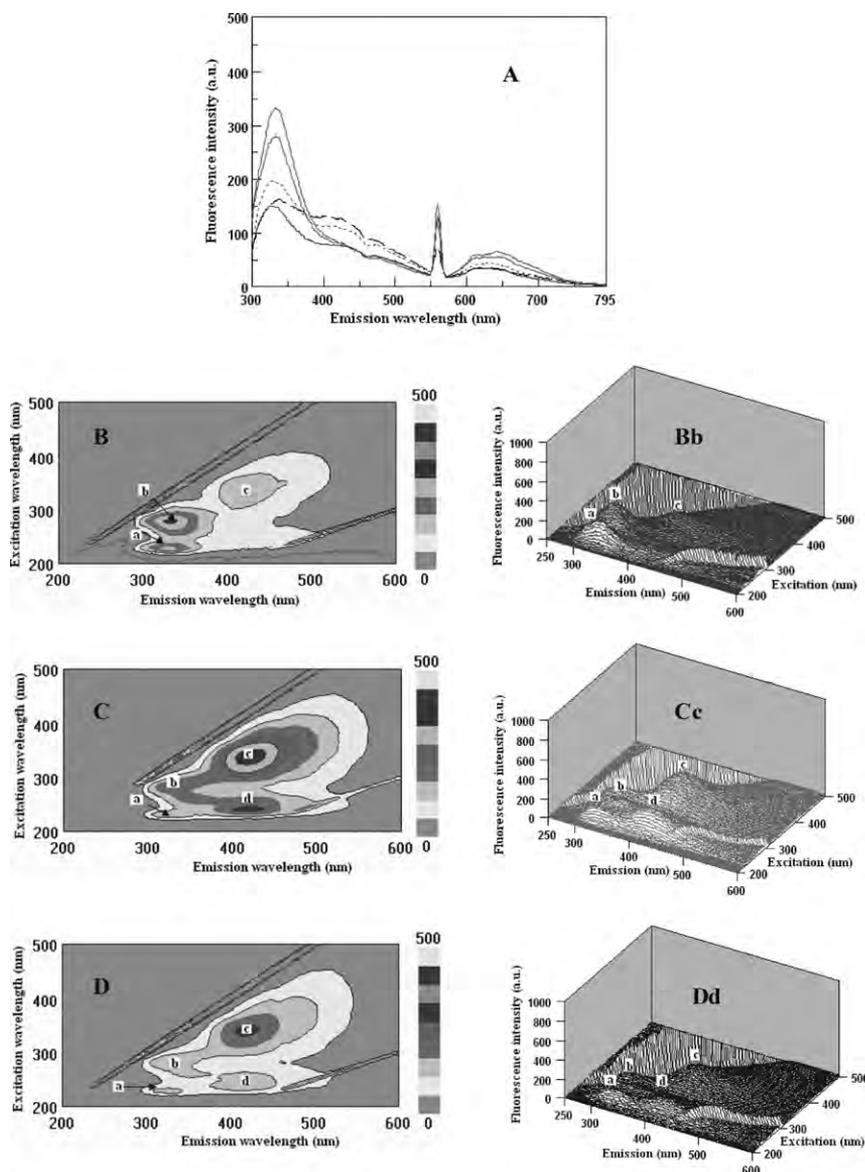


Figure 4. Spectral studies of kiwifruit methanol extracts. (A) Emission spectra of kiwifruit methanol polyphenol extracts at λ_{ex} 280 nm and λ_{em} 300 nm: lines from the top: OrgTr; SOrgTr; ConvTr; OrgC; ConvC. (B–D) Three-dimensional spectrum of OrgTr; SOrgTr; OrgC; Bb, Cc, Dd, cross spectrum of OrgTr; SOrgTr; OrgC. (Color images are available online at <http://aoac.publisher.ingentaconnect.com/content/aoac/jaoac>)

The amounts of polyphenols were similar to the information given in other reports (21). In spite of many variables studied in this report, the data showed the bioactivity of organic kiwifruit during two seasons was significantly higher than in conventional (Tables 1–3). We can only compare the physicochemical status of fruits treated with different substances. The treatment with ethylene was less evident in the literature than with other substances. The doses of spermine at 1.5 mM and spermidine at 2.0 mM showed the best results in extending the shelf life (2). Our tested parameters such as firmness changes, SSC, TA, reducing sugar, ascorbic acid, total phenols, and respiration rate during a 120-day storage period are in accordance with other results (8). The results showed that under conditions of approximately 2 to 5% O_2 , 3% CO_2 , and 2C, the kiwifruit could be stored for more than 120 days. Treatment with 1-MCP significantly slowed the decrease of flesh firmness and TA and delayed the increase of SSC in

kiwifruits during cold storage (6). Our results were in line with those of Sharma et al. (3), indicating that 2.0 $\mu\text{L/L}$ 1-MCP was the most effective treatment to delay softening and ripening in “Allison” kiwifruit. The fruits began to ripen only after 12 days in storage with the lowest weight loss ($9.8 \pm 0.2\%$). The untreated fruit started ripening on the sixth day of storage. The treated fruits showed the following indices: total phenolics ($24.3 \pm 0.3 \text{ mg } 100 \text{ g}^{-1}$), TA ($1.33 \pm 0.3\%$), and lower SSC ($8.33^\circ \pm 0.2^\circ \text{ Brix}$) than untreated kiwifruit (TA, $1.0 \pm 0.2\%$; SSC, $13.7^\circ \pm 0.3^\circ \text{ Brix}$). Fruits with the 1-MCP treatment could be stored for up to 5 weeks by maintaining higher fruit firmness, ascorbic acid, and total phenolic contents compared to the control. These results suggest that the application of 1-MCP at harvest effectively delayed the ripening process of the fruits. Fruit extracts had beneficial effects for the prevention of human cancer growth (7). At the end of storage, in comparison with the control, the treatment of $0.9 \mu\text{L} \times \text{L}^{-1}$

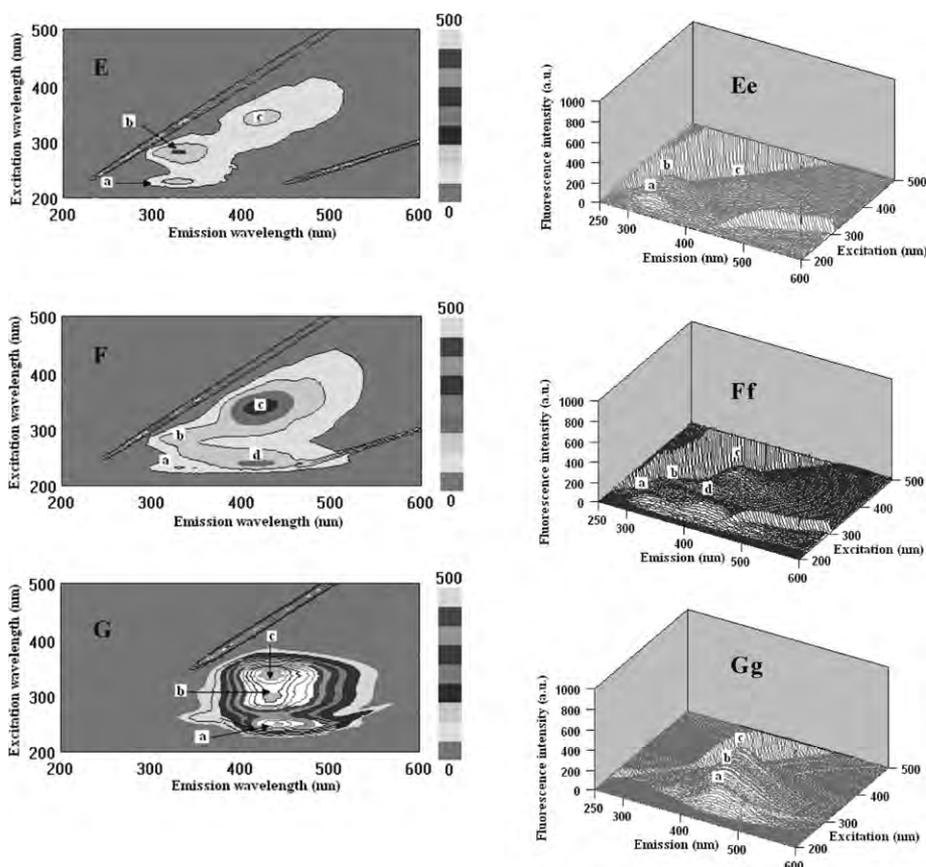


Figure 5. (E–G) Three-dimensional spectrum of ConvTr; ConvC; Caffeic acid; cross spectrum of ConvTr; ConvC; Caffeic acid. ConvC = Conventional control; OrgC = organic control; ConvTr = conventional treated; SOrgTr = semiorganic treated; OrgTr = organic treated; FI = fluorescence intensity; a.u. = arbitrary units. Concentration of ConvC = 5.6 µg/mL; OrgC = 5.7 µg/mL; ConvTr = 9.35 µg/mL; LCChemTr = 10.26 µg/mL; OrgTr = 17.16 µg/mL; Caffeic acid = 0.05 mM. (Color images are available online at <http://aoac.publisher.ingentaconnect.com/content/aoac/jaoac>)

1-MCP significantly improved “Xuxiang” quality TA content in 0.17% (27). Firmness abruptly decreased in fruits nontreated with NO, in contradiction to kiwifruit exposed to 200 µL/L of NO; indicating thus that NO can be used effectively for prolonging shelf life and maintaining fruit quality during distribution after cold storage (10). There are also contradictory

results indicating that “Hayward” kiwis of the three origins (conventional, integrated, and organic production systems) differed significantly in regard to panel ratings, sweetness, and juiciness. Similar results (28) were found in the comparison of postharvest-quality of CG and OG “Washington Navel” oranges. CG kiwis were larger and heavier than the others, had

Table 4. Fluorescence data of investigated samples of kiwifruit^a

(A) Two-dimensional fluorescence								
Line number	Samples	Quantitative parameters, λ _{em} /FI, nm/A.U.						
1	O-treated	333/333.26	425/83.27	473/54.33	615/60.53	642/67.21		
2	LC-treated	334/281.38	463/57.49	473/58.17	614/55.53	625/56.89		
3	C-treated	330/198.21	407/114.09	475/79.70	627/45.57	—		
4	O-control	338/163.91	400/132.33	424/127.90	467/90.04	621/36.17		
5	C-control	327/151.29	468/59.20	618/36.94	635/36.30	—		

(B) Three-dimensional fluorescence								
Samples	Peak a		Peak b		Peak c		Peak d	
	λ _{ex} /λ _{em} , nm/nm	FI A.U.	λ _{ex} /λ _{em} , nm/nm	FI A.U.	λ _{ex} /λ _{em} , nm/nm	FI A.U.	λ _{ex} /λ _{em} , nm/nm	FI A.U.
O-treated	228/344	225.09	278/333	283.10	337/421	164.08	—	—
LC-treated	231/329	140.00	282/329	206.45	240/419	241.54	339/421	363.01
C-treated	229/349	134.99	282/332	169.79	340/421	125.38	—	—
O-control	231/327	132.97	285/332	152.03	340/417	232.05	238/417	161.73
C-control	233/334	111.92	290/345	138.30	339/422	247.29	238/419	178.63
Caffeic acid	249/444	232.59	295/437	284.92	337/436	340.98	—	—

^a Kiwifruit samples of O-treated, LC-treated, C-treated, O-control, C-control, organic treated; low-chemical treated; conventionally treated; organic control and conventional control. Concentrations of all methanol extracts are 0.83 mg/mL at emission mode, at excitation = 280 nm; emission = 300 nm. FI = Fluorescence intensity; A.U. = arbitrary units (see Figure 5A).

the greatest SSC, and were sensorially sweeter and more juicy than those grown by integrated farming methods (29). Organic farming did not result in a clear superiority of the mineral quality of fruit and did not provide fruit that was free of toxic elements (18). There is no significant difference for moisture content of OG and CG fruits. OG fruits showed significantly higher content of total phenol, flavonoid, and total antioxidant activities than CG fruits. A positive relationship between total phenol-flavonoids, total phenol-total antioxidant capacity, and flavonoids-total antioxidant capacity among OG fruits was found. OG fruits contain higher amounts of nutritional as well as AP. Our recent results (20) with and without interaction with human serum albumin confirmed that the bioactivity mostly depends on the amount of polyphenols. Supplementation of OG fruits in the daily diet could lead to positive health effects. Organic limes and oranges showed higher mean values of acidity, at 4.5 and 34.8%, respectively (14), when compared to conventional fruit, which is in accordance with our results. SSC of organic limes were 25.8% higher than conventional fruits. In relation to the apple samples, there is no significant difference between organic and conventional cultivars. The antioxidant activities for limes and oranges were higher at 18.4 and 22.2%, respectively, in comparison to conventional fruit. Oranges from the conventional and certified organic citrus orchards were harvested at common maturity and kept at 4°C for 5 months. Compared to CG oranges, OG oranges had lower TA and citric acid, but better taste scores because they attained a higher TSS/TA ratio at harvest and during storage. The taste of CG and OG oranges was rated as acceptable throughout the storage period (18). Juices from CG and OG blood oranges were investigated for quality parameters and antioxidant capacity (30). The OG fruits contained higher sugar content and a lower acidity. CG fruit was found to have an increase in antioxidant capacity, in the concentration of phenolic acids, and in most flavonoids. OG fruit contained an increased concentration of hesperidin, which has been observed to possess biological activities associated with a healthy life (30). Tomato fruit from conventional greenhouse production contained higher levels of SSC on average than tomatoes grown organically (31).

Our results are in line with the previously published observations, that organic fruits tend to have higher hydrolysable polyphenols than conventional ones, with values from 11.5% in orange peels to 72.6% in papaya peels. Organic agriculture results in food products with similar or slightly higher polyphenol content and antioxidant capacity (32). The results shown here make it necessary to apply new analytical methods to find the differences in the quality of differently cultivated fruits. Our applied methods are in line with other reports (33), in which a combination of analytical and chemometric methods are useful tools for the characterization of extra virgin and other edible virgin oils. Applied methods showed slight differences among the grown fruits.

Conclusions

Based on the performed experiments and data evaluation, the following conclusions are formulated: (1) Ethylene treatment and then ripening improved the quality and bioactivity of kiwifruit. The bioactivity of organic kiwifruit was significantly higher than of semiorganic and conventional. Still, the scientific reports and our present and previous results are contradictory

regarding organic, semiorganic, and conventional fruits, as many variables are included in such comparisons.

(2) For a majority of the evaluated kiwifruit characteristics, it is apparent that their properties are affected in a specific order: treatment → growing conditions → time of storage that was also proved by statistical evaluation.

(3) The suggested methods for the improvement of kiwifruit quality can be applied for any fruit. The high quality of bioactive compounds in three cultivation systems makes kiwifruit even more important for daily consumption.

(4) All these aspects indicate the complexity of the problem of kiwifruit quality. We also note that the variability of the data, typical for measurements with samples of natural origin, indicates that some trends are unambiguous or, in some aspects, the differences are not as significant as expected.

Acknowledgments

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