

The Feasibility of Using $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ Values for Discriminating between Conventionally and Organically Fertilized Pepper (*Capsicum annuum* L.)

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A greenhouse experiment was conducted to determine the feasibility of using leaf and fruit $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values to discriminate between conventionally and organically fertilized peppers, when conventional management involves the application of organic amendment for soil preparation. All of the treatments involved adding horse manure to the soil before applying different rates of synthetic N fertilizers: 0 (T1 and T2), 150 (T3), and 300 kg ha⁻¹ (T4). The difference between T1 and T2 was that no synthetic fertilizer had been applied to plot T1 during the 5 years prior to the experiment. Significant differences were found in the $\delta^{15}\text{N}$ values of leaves and fruit from the plants grown under organic or mixed fertilization. The results indicate the possibility of using ^{15}N natural abundance as an indicator of fertilization management. On the other hand, $\delta^{13}\text{C}$ values did not contribute any additional information for discriminating between the organically and the synthetically and organically fertilized peppers.

KEYWORDS: $\delta^{15}\text{N}$; $\delta^{13}\text{C}$; nitrogen; fertilizer; organic; pepper; *Capsicum annuum* L.

INTRODUCTION

In general, organic agricultural production is considered to benefit the environment by using earth friendly agricultural methods and practices (1, 2). In recent years, there has been increasing demand for organic products because consumers consider them to be healthier and safer than conventional ones (3, 4). With this increasing interest in organic food comes the need for ways of confirming that organically labeled products have truly been grown with organic inputs. Synthetic pesticides are not allowed in organic systems, and in accordance with European Community Council Regulation No. 2092/91, analytical controls of organic agricultural products are based on the search for residues of these substances (5). As far as soil fertility management is concerned, only amendments and selected fertilizers of natural origin are allowed in organic systems. However, at present, no analytical controls of the fertilizer inputs are performed. Therefore, the fraudulent application of synthetic fertilizers to organic crops is difficult to detect.

To solve this problem and guarantee the authenticity of organic products, some authors have recently demonstrated that the stable isotope abundance of N ($\delta^{15}\text{N}$) may act as a potential tool for discriminating between organically and conventionally grown crops (6). This is based on the higher $\delta^{15}\text{N}$ values of composted manures as compared with those of synthetic fertilizers, due to preferential ammonium volatilization of the

lighter isotope (^{14}N) during the composting process (7, 8). Hence, the application of synthetic fertilizer to the crops will result in lower plant/fruit $\delta^{15}\text{N}$ values than observed in crops grown under organic regimes. In agreement with this hypothesis, Choi et al. (9) reported a significant ^{15}N enrichment of the soil and of plants of different species after long-term application of compost. Nakano et al. (10) found a good correlation between the $\delta^{15}\text{N}$ values of different fertilizers (organic or inorganic) and those of fruits, in an experiment performed with tomato plants. Also, differences in the $\delta^{15}\text{N}$ values of spinach (11) and oranges (12) have been reported when plants were grown with inorganic or organic fertilizers.

Most of this research work has been focused on totally organic or totally inorganic fertilization. However, the application of manure to the soil is not an agricultural practice exclusive to organic systems, and conventional management techniques may also involve organic amendment to improve soil quality and reduce pathogen populations (13). In particular, decomposing organic matter has been shown to inhibit soil-borne pests and diseases, and its use represents an alternative to using methyl bromide for soil pathogen control in glasshouse-grown horticultural crops (14). In conventional and low-input systems using organic amendments, additional applications of inorganic fertilizers at different rates are regularly performed, and as a consequence, the $\delta^{15}\text{N}$ values in soil and plants are presumably a mixture of the $\delta^{15}\text{N}$ values from the different sources (organic and inorganic). This combination could complicate interpretation

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Table 1. Soil Characterization of T1 Plot, before and after Biofumigation with Solarization^a

	before biofumigation	after biofumigation
water weight/soil weight	0.46	0.45
pH	7.59	7.64
EC (mmho cm ⁻¹)	6.54	10.79
soluble Na (mequiv/100 g)	1.17	1.97
Cl (mequiv/100 g)	1.12	1.67
SO ₄ (mequiv/100 g)	2.14	2.56
HCO ₃ (mequiv/100 g)	0.18	0.08
NO ₃ (mequiv/100 g)	8.04	27.69
available P (mequiv/100 g)	3.94	2.97
soluble K (mequiv/100 g)	0.53	0.55
soluble Ca (mequiv/100 g)	1.69	2.36
soluble Mg (mequiv/100 g)	1.29	2.01
total N (%)	0.19	0.21
organic matter (%)	2.26	2.28
C/N ratio	8.15	6.30

^a Biofumigation + solarization involved the application of fresh horse manure, mixing of the organic matter with the top soil layer, and subsequent soil covering with a plastic film for 2 months.

of the results when using the $\delta^{15}\text{N}$ values as an indicator of an agricultural regime.

The values of $\delta^{13}\text{C}$ have also been found to act as indicators of crop management practices (15). During photosynthesis, C isotope fractionation occurs as a result of Rubisco discrimination against the isotope ¹³C. Factors affecting stomatal conductance indirectly affect photosynthesis and, therefore, the isotope signal in the plant. In particular, fertilization has been shown to modify leaf C isotope composition due to its effects on stomatal conductance (16, 17). Fertilization performed in conventional systems involves higher N supply to the plant than in the case of organic fertilization and, therefore, may modify the C isotope signal. The aim of this study was to evaluate the feasibility of using N and C isotope composition for discriminating between conventionally (organic + synthetic) and organically fertilized peppers, when conventional management also involves the application of organic amendment for soil preparation.

MATERIALS AND METHODS

Plant Material and Cultivation. The experiment was conducted within a plastic greenhouse located in Murcia (SE Spain). The soil surface was divided into four different plots (7.8 m × 6.7 m). During the summer months before transplanting, biofumigation and solarization were performed. Biofumigation involved the uniform application of fresh horse manure at the rate of 4 kg m⁻² to all plots, mixing the organic matter with the top soil layer (10 cm). The soil was then covered with a plastic film, and the plastic greenhouse was closed from July to September (solarization). In **Table 1**, the initial soil characteristics of the T1 plot, before and after biofumigation with solarization, are presented.

Pepper seedlings (*Capsicum annuum* L. cv. Ribera) from the same seed lot and of the same age were transplanted on the same day. Plants were distributed in seven rows per plot, separated by 0.4 m within the rows and with 1 m between rows. Synthetic fertilizers, in the form of KNO₃ and Ca(NO₃)₂, were applied with each irrigation throughout the growing season at the different total rates shown in **Table 2**, which constituted the different treatments of the study. To avoid nutrients imbalance, P and Mg, in the form of H₃PO₄ and MgSO₄, respectively, were applied to the plots, maintaining a constant relationship among nutrients. All of the plots had a similar land-use history (crop sequences and management practices). Furthermore, they had received similar synthetic fertilizer inputs, except for T1, to which no synthetic fertilizer had been applied during the previous 5 years. This constituted the difference between treatments T1 and T2.

Table 2. Total Concentrations of Nutrients (kg ha⁻¹) Applied for the Different Treatments during the Cultivation: N, P, K, Ca, and Mg Were Applied in the Form of KNO₃ and Ca(NO₃)₂, H₃PO₄, and MgSO₄

treatments	N	P	K	Ca	Mg
T1					
T2					
T3	150	52	225	92	29
T4	300	103	451	185	59

All of the plots were similarly irrigated by drip irrigation at a rate based on FAO methodology (18) partially modified by Keller and Bliesner (19). Tensiometers were installed at depths of 15, 30, and 60 cm to monitor fluctuations in the soil moisture content. Soil water tension values were kept between 11 and 14 cb at 15 cm and over 20 cb at 60 cm.

Samplings. The $\delta^{15}\text{N}$ values were determined in the fresh horse manure and in the synthetic nitrogen fertilizers. Three replicates of manure and fertilizers were collected and then homogenized, oven-dried at 65 °C, and ground into very fine powders with a planetary monomill Pulverizette 6 grinding bowls (Fritsch) for posterior analysis.

One plot per treatment was allocated. Soil samples were collected from each plot, after biofumigation and solarization and before transplanting. Three replicates were sampled per plot, and each replicate was made by mixing four subsamples taken individually from the 0–20 cm depth zone.

For leaf and fruit sampling, each plot was divided into four subplots, perpendicularly to the rows. Samples were collected from the central rows of each subplot, considering each subplot as a replicate. Ten leaves and six fruits per replicate were harvested, randomly, from different plants. Leaf determinations were performed in the uppermost fully expanded leaves. Immature tissues were avoided, because the isotope fractionation effect, associated with NO₃⁻ assimilation, is more likely to be expressed in such tissues (20). Leaves were collected 53, 92, 116, and 158 days after transplanting (DAT). Fruits were harvested 150 DAT, at different stages of ripening (green, turning, or mature red). Fruits were washed with deionized water, and the seeds were removed. Leaves, fruit, and soil were oven-dried at 65 °C and ground into very fine powders for subsequent determination of $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values and leaf organic N and NO₃⁻ concentrations.

Soil Analysis. A representative soil sample was collected before and after biofumigation + solarization to determine the effect of this disinfection technique on soil properties. Soluble salts, pH, and EC were determined in the saturated paste extract according to Rhoades (21). Extractable phosphorus was determined by the Olsen method (22). Organic N was analyzed by the Kjeldahl procedure (23).

Leaf Organic N and NO₃⁻ Concentration. Leaf NO₃⁻ was extracted with water according to Abbas et al. (24). For this, 50 mg of dried plant material was extracted with deionized water by shaking for 30 min. The extracts were centrifuged (6000g) and filtered (0.45 μm), and NO₃⁻ was determined by using a Waters Capillary Ion Analyzer (Milford, New Hampshire). Organic N was determined from dried material by the Kjeldahl procedure (23).

N and C Isotope Composition. $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ were determined in duplicates of each sample, using a ThermoFinnigan FlashEA1112 Elemental Analyzer (Milan, Italy), connected to a Finnigan MAT DELTAplus Isotopic Mass Spectrometer (Bremen, Germany) with a Finnigan MAT ConFlo II connection interphase. Dried samples were weighed (2 mg) in tin capsules with a Sartorius XM 1000P microbalance. The values of the isotope ratio were expressed in ‰ according to the formula:

$$\delta\text{‰} = [(R_{\text{sample}} - R_{\text{standard}})/R_{\text{standard}}] \times 1000$$

where R is ¹⁵N/¹⁴N for $\delta^{15}\text{N}$ values and ¹³C/¹²C for $\delta^{13}\text{C}$ values. Following international convention, the standards used were the nitrogen isotope ratio in air and the carbon isotope ratio in VPDB (Vienna Pee Dee Belemnite Standard), for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$, respectively. For ¹⁵N_{air}, the samples were referenced against the following materials certified by the International Atomic Energy Agency: IAEA-N-1 [(NH₄)₂SO₄],

Table 3. Soil Isotope Composition ($\delta^{15}\text{N}$ and $\delta^{13}\text{C}$) of the Different Plots after Biofumigation and Solarization and before the Different Treatments with Inorganic Fertilizers

treatment	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$
T1	12.05 \pm 0.14 ^a	-11.65 \pm 0.36 ^a
T2	11.72 \pm 0.18	-11.52 \pm 0.31
T3	12.32 \pm 0.17	-11.81 \pm 0.52
T4	11.9 \pm 0.13	-11.73 \pm 0.18

^a Values are means \pm SE ($n = 3$).

Table 4. Isotope Composition ($\delta^{15}\text{N}$) of the Fertilizers and Manure Used in the Different Treatments

sample	$\delta^{15}\text{N}$
KNO ₃	1.23 \pm 0.05 ^a
Ca(NO ₃) ₂	-1.72 \pm 0.06
manure	7.16 \pm 0.07

^a Values are means \pm SE ($n = 3$).

IAEA-N-2 [(NH₄)₂SO₄], and IAEA-NO3 (KNO₃). For ¹³C_VPDB, the reference materials were NBS 19 (TS-limestone), IAEA-CH-6 (sucrose), and NBS22 (hydrocarbon oil). The data were analyzed statistically, using the SPSS 7.5 software package, by analysis of variance (ANOVA) and by Tukey's multiple range test to determine differences between means.

RESULTS AND DISCUSSION

$\delta^{15}\text{N}$ Values of Soil, Manure, and Synthetic Fertilizers.

Soil samples were collected before synthetic fertilizer application to T3 and T4. At this time, soil management for T2, T3, and T4 plots had been the same, and as a consequence, no significant differences in $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values between plots were detected (Table 3). In addition, there were no significant differences in N and C isotopic composition between these treatments and the T1 treatment, in which no synthetic fertilizer had been applied during the previous 5 years. This can be attributed to the negligible contribution of the fertilizers (applied to T2 plot during previous cultivations) to the soil N pool due to extraction by plants or loss by leaching.

From these results, it can be assumed that the isotope composition of the N source, before the different synthetic fertilizer applications, was similar for all of the treatments. In addition, the only difference between organic treatments (T1 and T2) and those with combined fertilization (T3 and T4) in isotopic composition of the N source would have been due to the synthetic fertilizers applied during cultivation in T3 and T4.

The $\delta^{15}\text{N}$ value of fresh horse manure was lower than that of the biofumigated soil (Table 4). Isotope fractionation of N during soil solarization could play a part in these differences. In spite of the horse manure applied for soil biofumigation, the soil organic matter content before biofumigation with solarization was similar to that found after application of this soil disinfection technique (Table 1). However, the soil NO₃⁻ concentration was greatly increased after soil biofumigation and solarization, which may be explained by the fact that soil solarization involves high rates of organic matter mineralization by increasing the soil temperature (25). It has been demonstrated that during the mineralization processes, organic matter is enriched in ¹⁵N due to preferential ammonium volatilization of the lighter isotope (¹⁴N) (7, 8). Thus, the higher $\delta^{15}\text{N}$ values in soil after manure application and solarization than in fresh horse manure are to be expected.

As far as synthetic N fertilizers are concerned, their $\delta^{15}\text{N}$ values are usually close to 0‰ since they are manufactured from

Table 5. Effect of Treatments and Sampling Date (DAT) on Leaf Organic N and NO₃⁻ Concentration (mmol kg⁻¹)^a

	Factorial ANOVA	
	F	significance level
	organic N	
treatment	19.1	***
sampling date	7.3	**
treatment \times sampling date ^b	1.7	NS
	NO ₃ ⁻	
treatment	23.0	***
sampling date	14.6	***
treatment \times sampling date	2.3	*
	Tukey Test ^c	
	organic N	NO ₃ ⁻
	treatment	
T1	2483.4 a	74.8 a
T2	2469.9 a	86.96 a
T3	2610.2 ab	147.3 b
T4	2851.6 b	198.0 c
	sampling date	
53 DAT	2920.4 c	193.8 c
92 DAT	2785.9 bc	128.7 ab
116 DAT	2614.6 b	159.5 bc
158 DAT	2263.6 a	86.2 a

^a Summary of factorial ANOVA and means comparison test (Tukey). ^b Asterisk (*) signifies interaction between the factors. ^c Different letters in the same column indicate significant differences between treatments and sampling dates, respectively, at the 5% level of probability.

atmospheric N ($\delta^{15}\text{N}_{\text{atm}} = 0\text{‰}$) by different industrial processes (26, 27). In our experiment, the $\delta^{15}\text{N}$ values of the KNO₃ and Ca(NO₃)₂ used for plant fertilization were significantly lower than in biofumigated soil (Table 4). These differences in the ¹⁵N signature between N sources are the basis for differentiating between crops grown with or without inputs of synthetic N fertilizers.

Leaf N Composition. Leaf organic N and NO₃⁻ concentrations decreased with time regardless of the treatment (Table 5). Both parameters were significantly ($p < 0.001$) affected by the treatments and reflected the increasing rates of N fertilization. Tukey's test did not identify significant differences in organic N or NO₃⁻ content between plants grown in soils to which no synthetic fertilizer had been applied during the previous 5 years (T1) and those of T2 (first year without receiving any synthetic fertilizer).

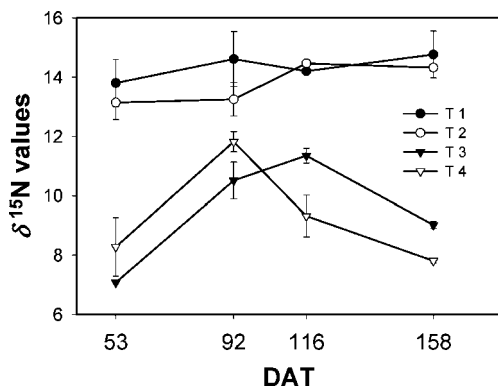
In T1 and T2, the leaf $\delta^{15}\text{N}$ values were about 2‰ higher than in the N source (biofumigated soil) (Tables 3 and 6). Differences in $\delta^{15}\text{N}$ between the plant and the original source can be attributed to isotope discrimination during physiological processes of N assimilation within the plant (28). Similar results were found in a previous study with tomato plants, where higher leaf $\delta^{15}\text{N}$ values than those of N source were observed due to NO₃⁻ reduction in both root and shoot (29). On the contrary, no intraplant variation was found when using NH₄⁺ as an N source in the above study. In addition, whole-plant fractionation was not observed due to the lack of N efflux back to the soil. In our study, the $\delta^{15}\text{N}$ of the different plant N pools was not measured. However, higher leaf $\delta^{15}\text{N}$ values in T1 and T2 plants with regard to N source can similarly be explained as a consequence of (i) the N efflux rate, (ii) assimilation patterns between NH₄⁺ and NO₃⁻, and (iii) partitioning of total reduction between root and shoot tissues.

The leaf $\delta^{15}\text{N}$ values of plants grown with synthetic fertilizers (T3 and T4) were significantly ($p < 0.001$) lower than in T1

Table 6. Effect of Treatments and Sampling Date (DAT) on Leaf $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ Values^a

Factorial ANOVA		
	F	significance level
$\delta^{15}\text{N}$		
treatment	10.7	***
sampling date	98.4	***
treatment \times sampling date ^b	4.7	**
$\delta^{13}\text{C}$		
treatment	2.9	NS
sampling date	45.0	***
treatment \times sampling date	0.7	NS
Tukey Test ^c		
	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$
treatment		
T1	14.35 b	-28.10
T2	13.79 b	-27.71
T3	9.49 a	-27.69
T4	9.31 a	-27.65
sampling date		
53 DAT	10.59 a	-28.91 a
92 DAT	12.55 b	-27.43 bc
116 DAT	12.32 b	-27.07 c
158 DAT	11.48 ab	-27.74 b

^a Summary of factorial ANOVA and means comparison test (Tukey). ^b Asterisk (*) signifies interaction between the factors. ^c Different letters in the same column indicate significant differences between treatments and sampling dates, respectively, at the 5% level of probability.

**Figure 1.** Evolution of leaf N isotope composition ($\delta^{15}\text{N}$) for plants grown under the different fertilization treatments. Bars are means \pm SE ($n = 4$).

and T2 plants (Table 6). The patterns of temporal changes in leaf $\delta^{15}\text{N}$ were different depending on the treatment (Figure 1). In T1 and T2, $\delta^{15}\text{N}$ values remained within a range of $\pm 1\%$ throughout the growing period. However, in T3 plants, the $\delta^{15}\text{N}$ values increased sharply between days 53 and 116 while the same occurred in the T4 plants between days 53 and 92, both decreasing afterward. This different variation in leaf $\delta^{15}\text{N}$ can explain the significant ($p < 0.01$) interaction between the effect of treatments and sampling date, detected by factorial ANOVA (Table 4). The isotope signature in T3 and T4 plants changed during the growth period, probably due to temporal variations in the availability of the two N sources (biofumigated soil and synthetic fertilizers). Similar variations with time were found by others authors who attributed increasing $\delta^{15}\text{N}$ values in fertilized crops to the decreasing availability of synthetic fertilizer with time due to uptake, losses, and immobilization (8, 30). At 53 and 92 DAT, the T4 plants showed higher $\delta^{15}\text{N}$ values than the T3 plants. According to Kohl (31), this increase in $\delta^{15}\text{N}$, in spite of increasing application rates of synthetic

fertilizers with low $\delta^{15}\text{N}$, reflects the loss of excess N from plots continuously fertilized at the higher rates.

In spite of these temporal variations, T3 and T4 plants showed lower leaf values than T1 and T2 plants throughout all of the growing periods, perhaps due to the continuous supply of synthetic fertilizer by fertigation. Unlike other methods, fertigation, which is the most usual method adopted for horticultural crops in greenhouse conditions, provides a supply of water-dissolved fertilizers throughout the growing season. Therefore, synthetic N fertilizers may contribute to plant $\delta^{15}\text{N}$ values during the entire course of cultivation. As a consequence, differences between organically and synthetically + organically fertilized peppers could be detected at all of the sampling dates. This difference in isotope signature could, potentially, be used for discriminating between organically and conventionally grown pepper plants, even if conventional management involves the application of organic amendment.

Reflecting the soil $\delta^{15}\text{N}$ values, there were no significant differences in leaf $\delta^{15}\text{N}$ values between T1 and T2 plants (Table 6). Similarly, overall significant differences between T3 and T4 plants were not observed although the fertilizer rates in T4 were two times greater than in T3. However, from 116 DAT, the isotope composition of T4 plants showed a decrease in $\delta^{15}\text{N}$ relative to T3 plants (Figure 1), probably due to the higher contribution of the synthetic fertilizers to plant values.

The carbon isotope composition of leaves did not differ significantly between treatments (Table 6). Changes in leaf $\delta^{13}\text{C}$ during the growth period were similar in all of the treatments, and no interaction between treatment and sampling date was detected by ANOVA. An increase of $\delta^{13}\text{C}$ values was observed in all of the treatments from 53 to 116 DAT, followed by a decrease between 116 and 158 DAT.

Factors affecting plant C isotope composition include atmospheric CO_2 concentration, soil moisture content, fertilization, and other environmental factors (32). In our experiment, the soil organic matter content, irrigation rates, and environmental conditions were the same for all of the treatments. Thus, factors affecting plant $\delta^{13}\text{C}$ values did not differ between treatments, with the exception of fertilization rates. Although our experiment involved increasing rates of N fertilization, $\delta^{13}\text{C}$ values did not reflect these differences and so cannot be used for discriminating between conventionally and organically fertilized peppers.

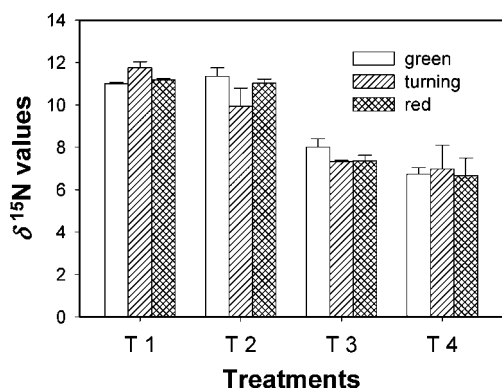
Fruit Isotope Composition. The $\delta^{15}\text{N}$ values in fruit were lower than leaf values (Table 7). The enzymatic reactions involved in N remobilization and redistribution generally result in products with lower $\delta^{15}\text{N}$ levels (33). Thus, N redistribution during reproductive growth from organs with a low N demand to fruits could lead to lower $\delta^{15}\text{N}$ values in fruits than in leaves. Treatments involving synthetic fertilization (T3 and T4) led to significantly ($p < 0.001$) lower fruit $\delta^{15}\text{N}$ values than the organic treatments (T1 and T2) (Table 7). No effect of fruit ripening stage on N isotope composition was observed. The effect of the different treatments on the N isotope composition of green, turning, and fully mature red peppers is represented in Figure 2 where differences of around 2–5‰ in fruit $\delta^{15}\text{N}$ values can be observed between the organic and the mixed-fertilization treatments. Although these differences are significant, more studies, using a greater range of inorganic fertilization rates and different types of organic fertilizer, are needed to assess the feasibility of using the N isotope signal as an indicator of fertilization management.

In spite of the slight increase in fruit $\delta^{13}\text{C}$ values with increasing fertilization rates (Table 7), the values did not differ significantly and so cannot be said to discriminate between the

Table 7. Effect of Treatments and Stage of Ripening (DAT) on Fruit $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ Values^a

Factorial ANOVA		
	F	significance level
$\delta^{15}\text{N}$		
treatment	47.7	***
stage of ripening	0.8	NS
treatment \times stage of ripening ^b	1.2	NS
$\delta^{13}\text{C}$		
treatment	2.2	NS
stage of ripening	0.3	NS
treatment \times stage of ripening	0.7	NS
Tukey Test ^c		
	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$
treatment		
T1	11.30 b	-27.49
T2	10.78 b	-27.28
T3	7.55 a	-27.18
T4	6.78 a	-26.80
stage of ripening		
green	9.60	-27.16
turning	9.07	-27.11
mature red	8.98	-27.29

^a Summary of factorial ANOVA and means comparison test (Tukey). ^b Asterisk (*) signifies interaction between the factors. ^c Different letters in the same column indicate significant differences between treatments and sampling dates, respectively, at the 5% level of probability.

**Figure 2.** $\delta^{15}\text{N}$ values of green, turning, and fully mature red peppers from plants grown under the different fertilization treatments. Bars are means \pm SE ($n = 4$).

organic and the conventional treatments. These results were consistent with those reported by Nakano et al. (10), who found similar $\delta^{13}\text{C}$ values in tomato fruit grown with organic and inorganic fertilization. However, higher $\delta^{13}\text{C}$ values in onion and Chinese cabbage were reported when comparing organic and integrated fertilization (34). In that study, the differences in C isotope composition were attributed to differences in soil respiration rates between organic and integrated treatments, as a consequence of green manure only being applied to the organic plots. However, in our study, all of the plots were subjected to the same management practices (except for the addition or not of synthetic fertilizers); therefore, no such differences were to be expected. Finally, fruit C isotope composition was not affected by the stage of ripening.

In summary, significant differences in the $\delta^{15}\text{N}$ values of leaves and fruits from plants grown under organic and mixed fertilization were found. The results point to the possibility of using the natural abundance of ^{15}N as an indicator of fertilization management (organic or conventional) in pepper when con-

ventional management also involves the application of organic matter to the soil. In general, $\delta^{15}\text{N}$ values of composted manure are significantly higher than those of synthetic fertilizers (9). This signature difference between sources is a condition required for the applicability of our results to other commonly used organic amendments. On the other hand, environmental factors, such as light, temperature, and air humidity, and their interaction with agricultural management practices can modify plant N isotope compositions (17, 35). Therefore, further studies, including these factors and also other organic sources with lower $\delta^{15}\text{N}$ values (e.g., soybean foliage), are necessary to ascertain a methodology for discrimination between organic and nonorganic crops.

Finally, there was no correlation between leaf or fruit $\delta^{13}\text{C}$ values and fertilization. Therefore, the C isotope composition did not contribute additional information for discriminating between organically and synthetically + organically fertilized pepper.

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