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Changes in red pepper antioxidants as affected by UV-C treatments and storage at chilling temperatures

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ABSTRACT

Chilling injury (CI) is one of the main factors limiting refrigeration in several horticultural commodities of subtropical and tropical origin such as pepper. Short UV-C treatments before low temperature storage have been shown to reduce CI. In this work we wanted to test whether or not the reduced susceptibility to CI in UV-C treated fruits was associated with increased levels of antioxidant compounds and enzymes. Red peppers (*Capsicum annuum* L.) were treated with UV-C radiation (10 kJ/m^2) and stored at 0 °C for 21 d. During storage we analyzed chilling injury development, ascorbic (AA) and dehydroascorbic acids and DPPH radical scavenging capacity. We also followed superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX) and guaiacol peroxidase (GPX) activities. CI increased rapidly when the fruit was stored for longer than 14 d, but was significantly lower in UV-C treated peppers. Exposure to UV-C did not alter fruit color but reduced weight loss. Although AA and DPPH radical scavenging capacity were lower in the control, this occurred towards end of storage, when CI was already advanced. In contrast, SOD, CAT and APX activities were higher in UV-C treated fruits during the first 2 weeks of storage when the symptoms became visible. Results show that UV-C exposure prevents CI and weight loss in red pepper and suggest that this might be related to increased activity of antioxidant enzymes.

1. Introduction

Chilling injury (CI) includes a series of disorders associated with storage of fruits and vegetables at low temperature. The physiological and biochemical alterations occurring during CI include inactivation of enzymes, membrane dysfunction caused by modifications in plasma membrane fluidity and permeability, stimulation of respiration and ethylene production and ultimately an alteration in whole cell machinery (Concellón, Añón, & Chaves, 2005, 2007; Wang, 2000). Metabolic alterations can be reversible in a first phase occurring well before visible symptoms are manifested (Balandran-Quintana et al., 2002) but if exposure to low temperatures persists severe damage occurs.

Pepper fruits are susceptible to CI when stored at temperatures lower than 7 °C (González-Aguilar et al., 2000). Symptoms of the disorder include seed browning, tissue discoloration and depressions in the pericarp which evolve into scalds at advanced stages. After transfer to ripening temperatures, rapid softening occurs and the fruit becomes highly susceptible to opportunistic pathogens such as *Alternaria alternata*.

Several treatments have been tested in fruits and vegetables to reduce CI. In peppers, heat treatments (González-Aguilar et al., 2000), methyl jasmonate (MeJa) and methyl salicylate (MeSa) vapors (Fung, Wang, Smith, Gross, & Tian, 2004) have shown positive results. Postharvest UV-C treatments are mainly used to control pathogens (Allende & Artes, 2003; Erkan, Wang, & Krizek, 2001). However, they can also modulate ripening (Maharaj, Arul, & Nadeau, 1999), delay senescence (Costa, Vicente, Civello, Chaves, & Martínez, 2006), induce the accumulation of bioactive compounds and reduce some physiological disorders (Erkan, Wang, & Wang, 2008; González-Aguilar, Wang, & Buta, 2004). Short UV-C treatments (7 kJ/m^2) reduced the severity and incidence of CI of bell peppers (Vicente et al., 2005). The mechanisms determining tolerance to low temperatures are largely unknown and given that the term CI includes alterations having

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a common effector, but with different targets, symptoms and manifestations it might be dependant on the commodity considered.

Reactive oxygen species (ROS) such as singlet and triplet oxygen, $O^{\bullet}{}_{-2}$, $OH^{\bullet}$, H_2O_2 are generated as a part of normal metabolism. However, their production could increase dramatically under stress conditions (Mittler, 2002). Exacerbated production of ROS has been observed at early phases of CI (Fuller, Hopwood, Anson, & Sala, 1998). In pepper, overproduction of mitochondrial superoxide is detected when the fruit is stored at chilling temperatures (Purvis, Shewfelt, & Gegogeine, 1995). Fung et al. (2004) found that treatments with MeJA and MeSa decreased CI and induced the expression of alternative oxidase (AOX) which has been suggested to prevent ROS production (Maxwell, Wang, & McIntosh, 1999). However, in a survey of different cultivars no correlation was found between endogenous AOX transcript levels and susceptibility to CI, suggesting that other processes might be involved (Smith, Stommel, Fung, Wang, & Whitaker, 2006). In order to cope with ROS, plant cells have a series of antioxidant compounds and enzymes such as superoxide dismutases, peroxidases and catalases (Apel & Hirt, 2004). The activation of antioxidant defenses has been suggested to participate in the acclimation of plants to stress (Cao, Zheng, Wang, Jin, & Rui, 2009). Changes in the antioxidative defense system were reported in tomatoes and strawberries subjected to UV-C radiation (Barka, Kalantari, Makhlouf, & Arul, 2000; Erkan et al., 2008). The aim of this work was to evaluate the changes in antioxidant enzymes and compounds in UV-C treated bell peppers during storage at chilling temperatures.

2. Materials and methods

2.1. Plant material and selection of UV-C treatment

Peppers (*Capsicum annum* L. cv Cornago) grown in greenhouses in La Plata, Buenos Aires (Argentina), having at least 80% surface red color estimated by visual inspection were harvested and immediately taken to the laboratory. Fruits free of damage were selected and washed with 100 mg/L sodium hypochlorite. In order to determine the most suitable experimental conditions, different UV-C light doses (peak emission at 254 nm) were applied. Fruit was carefully placed in plastic trays under a bank of four germicidal UV-C lamps (TUV G30T8, 30W, Philips, Bs. As., Argentine) and irradiated at a distance of 30 cm to obtain doses of 10 or 20 kJ/m^2. The UV-C treatment demands only a few minutes and the different doses were obtained by altering the duration of the exposure at the fixed distance. The radiation is not penetrating so we rotate 3 times the fruit to irradiate each of their 3 sides and ensure uniform surface exposure to UV light. The radiation intensity was measured with an UV digital radiometer (Model WLX3W, Cole-Palmer Instrument Company, Vernon Hills, IL, USA). Once the treatments were finished, irradiated fruit was placed in plastic trays, covered with perforated PVC film and stored at 0 °C for 21 d. Corresponding controls without UV-C treatment were packed and stored in the same conditions. Selection of the appropriate dose was done based on CI index and weight loss. The whole experiment was repeated three times.

2.2. Effect of UV-C treatment on physico-chemical quality at 0 °C

Once the proper UV-C dose was selected, red pepper fruit was harvested (> 80% surface red color) and treated with UV-C light (10 kJ/m^2) as previously described. After that, fruit was put in plastic trays, covered with perforated PVC and stored at 0 °C for 0, 7, 14 and 21 d. Corresponding controls without treatments were directly packed and stored in the same conditions. On each sampling day, fruit was analyzed immediately or otherwise cut, frozen in liquid nitrogen and stored at −20 °C until use. The whole experiment was repeated three times.

2.2.1. Chilling injury (CI)

Fruit was evaluated weekly for internal and external CI symptoms (color development, softness, water soaking, pitting, decay, surface depressions and seed browning). Twenty seven fruits were used for each treatment and sampling day. A scale ranging from 1 to 4 was used, being: 1 = no damage; 2 = low damage; 3 = moderate damage and 4 = severe damage. The CI index was calculated according to the following equation:

$$\text{CI index} = \frac{\sum(\text{injury level} \times \text{number of fruits in this level})}{\text{Total number of fruits in the treatment}}$$

2.2.2. Weight loss

Fruit was weighed individually at the beginning of the experiment and weekly during storage. Twenty seven fruits were used for each treatment and sampling day. Results were expressed as grams of weight loss per 100 g of fresh fruit relative to the initial value.

2.2.3. Surface color

Surface color was evaluated with a colorimeter Konica Minolta Model CR-400 (Minolta, Osaka, Japan) by measuring the Hue angle in three zones of each fruit (basal, equatorial and apical zones). Two determinations for each fruit and zone were done and values were averaged. Twenty seven fruits were analyzed for each treatment and storage time.

2.2.4. DDPH radical scavenging capacity

For sample preparation frozen fruit tissue was ground in a mill (IKA Model A11; IKA Works Inc., SP Brazil) and 1 g of the resultant powder was extracted 60 min in 15 mL of ethanol. The mixture was centrifuged at 9000×*g* for 15 min at 4 °C and the supernatant was brought to 30 mL with ethanol. Free radical scavenging capacity of pepper was tested according to the method reported by Brand-Williams, Cuvelier, and Berset (1995) based on the reduction of the radical 2,2-diphenyl-1-picrylhydrazyl ($DPPH^{\bullet}$) in the presence of hydrogen-donating antioxidants. Several aliquots of the ethanolic extracts (50–250 μL) were mixed with 1 mL of 40 mg/L $DPPH^{\bullet}$ in ethanol prepared daily. The absorbance at 515 nm was measured at different times until the reaction reached a plateau (10 min). The percentage of remaining $DPPH^{\bullet}$ against the volume of extract was then plotted to obtain the amount of extract necessary to decrease the initial $DPPH^{\bullet}$ concentration by 50%, which was defined as EC_{50}. Results were expressed as $1/EC_{50}$ in 1/g of fresh tissue. Two extracts were prepared for each treatment and storage time and measurements were done in triplicate.

2.2.5. Ascorbic acid (AA) and dehydroascorbic acid (DHA)

Frozen tissue was ground in a mill and 1 g of the resultant powder was extracted for 10 min in 6 mL of cold 3 g/100 mL citric acid in an ice-bath with stirring and protected from light. The suspension was centrifuged at 14,000×*g* for 10 min at 4 °C and the supernatant was filtered through a 0.45 μm pore membrane. The filtrate was used for AA and DHA analysis. Measurements were done in a Waters HPLC, Model 6000A (Milford, MA, USA) equipped with an UV–vis detector. The samples were separated by using an Altex Ultrasphere™ –ODS-C18 column (4.6 × 250 mm) operated at room temperature. A flow rate of 0.8 mL/min was used with 0.2 mol/L KH_2PO_4, adjusted to pH 2.2 as mobile phase. Detection was done at 254 nm for AA and 220 nm for DHA. Two extractions were done for each treatment and

storage time, and measurements were done in duplicate. Results were expressed as μg per gram of fresh tissue.

2.2.6. Extraction of antioxidant enzymes

Frozen pericarp tissue was ground in a mill and 2 g of the resulting powder were extracted in 15 mL of the corresponding buffer (Table 1) for 60 min with stirring at 4 °C. The suspension was then centrifuged at 13,000×*g* for 15 min and the supernatant was saved and used to determine the enzymatic activities. All steps during sample preparation were carried out at 0–4 °C. Two independent extracts were done for each storage time and treatment.

2.2.7. Enzyme activity assays

For superoxide dismutase (SOD, EC 1.15.1.1) activity, the reaction mixture contained 100 mmol/L phosphate buffer pH 7.8; 13 mmol/L methionine, 75 μmol/L nitroblue tetrazolium (NBT), 6 μmol/L riboflavine, 0.1 mmol/L EDTA and 200 μL of enzymatic extract in a total volume of 3 mL. The reaction mixture was prepared in glass test tubes which were put under four 30 W fluorescent lamps at 20 °C for 15 min. A sample without enzymatic extract was used as control. A non-irradiated reaction mixture was used as a blank. The absorbance at 560 was measured and one enzymatic activity unit (EAU) was defined as the amount of enzyme required to produce a 10% inhibition of NBT oxidation under the assay conditions.

For guaiacol peroxidase (GPX, EC 1.11.1.7) assay the reaction mixture (2 mL) contained 2.5 g/L guaiacol, 8 mmol/L H_2O_2, 0.3 mL of extract and 0.1 mol/L phosphate buffer (pH 7.0). Enzyme activity was determined by monitoring the increase in the absorbance at 470 nm. The unit of GPX activity was defined as the ΔOD in a min.

Catalase (CAT, EC 1.11.1.6) activity was determined by measuring the residual H_2O_2 in the reaction mixture with $TiCl_4$. The reaction mixture (1 mL) contained phosphate buffer (0.1 mol/L, pH 7.0), 15 mmol/L H_2O_2 and 250 μL of enzymatic extract. Aliquots of 150 μL of the reaction mixture were taken at 0, 0.5, 1, 2, 3 and 5 min and added to test tubes containing 300 μL of 0.2 mol/L $TiCl_4$, 200 μL of 98 g/100 g H_2SO_4 and 1.35 mL of H_2O. The absorbance of the samples at 410 nm was measured. One unit of CAT activity was defined as the amount of enzyme consuming 1 μmol of H_2O_2 in a min under the assay conditions.

For ascorbate peroxidase (APX, EC 1.11.1.11) activity the reaction mixture contained 100 mmol/L phosphate buffer (pH 7.0), 0.1 mmol/L EDTA, 0.5 mmol/L sodium ascorbate, 0.1 mmol/L H_2O_2 and 300 μL of enzymatic extract. The activity was determined by monitoring the change in absorbance at 290 nm due to oxidation of ascorbate and the enzymatic activity unit (EAU) was defined as the amount of enzyme oxidizing 1 μmol of sodium ascorbate in a min under the assay conditions.

In all cases protein content was determined according to the method of Bradford (1976), measurements were done in triplicate and results were expressed in EAU per mg of protein.

Table 1
Extraction buffers used for superoxide dismutase (SOD), guaiacol peroxidase (GPX), ascorbate peroxidase (APX) and catalase (CAT) extraction.

Enzyme	Extraction buffer
SOD	Phosphate buffer (100 mmol/L, pH 7.8) containing 1 mmol/L phenyl methyl sulfonyl fluoride (PMSF), 0.1 mmol/L EDTA and 10 g/L polyvinylpolypyrrolidone (PVPP).
GPX and CAT	Phosphate buffer (100 mmol/L pH 7.0) containing 1 mmol/L PMSF, 0.1 mmol/L EDTA, 0.1 mmol/100 mL Triton X-100 and 10 g/L PVPP.
APX	Phosphate buffer (100 mmol/L, pH 7.0) containing 1 mmol/L PMSF, 0.1 mmol/L EDTA, 0.1 mL/100 mL Triton X-100, 5 mmol/L ascorbic acid and 10 g/L PVPP.

2.3. Statistical analysis

Experiments were performed according to a factorial design. Analysis of variance (ANOVA) was performed and means were compared by a least significant difference (LSD) test at $P < 0.05$.

3. Results and discussion

3.1. UV-C dose optimization

Previous work reported a reduction in CI in red peppers treated with a UV-C radiation (Vicente et al., 2005). In this study, the dose used (7 kJ/m^2) was initially selected based on decay control of fruit stored at 10 °C and it was then shown that the treatment was also useful to reduce CI. Here, we initially evaluated if a higher dose might was even more beneficial. Red peppers were subjected to UV-C treatments of 10 and 20 kJ/m^2 and then stored at 0 °C. Control peppers were more susceptible to chilling injury than fruit treated with 10 or 20 kJ/m^2 of UV-C (Table 2). The least weight loss was found in fruit treated with an UV-C dose of 10 kJ/m^2 (1.3%). Given both the reduced weight loss and the lower exposure time of the 10 kJ/m^2 treatment to obtain the same benefit on CI prevention, this condition was selected for further studies.

3.2. Effects of UV-C treatment on chilling injury color and weight loss

In a second set of experiments, fruit was treated with 10 kJ/m^2 and stored at 0 °C. Control fruit showed incipient CI symptoms after 7 d (Fig. 1 A). The manifestation of CI rose markedly between 14 and 21 d at 0 °C, reaching a CI index = 3.7, corresponding to a moderate-severe damage (Fig. 1B). UV-C treated fruit showed low-moderate CI index (2.5) after 21 d. The UV-C treatment did not cause negative changes in fruit surface color (Fig. 2A) and reduced weight loss significantly (Fig. 2B). This might be associated with the reduced damage caused by exposure to UV-C. The results obtained in the present study show that 10 kJ/m^2 UV-C treatments can reduce CI of peppers cv. Cornago. The existence of cross resistance against different stress conditions suggests a crosstalk in the physiological responses (Apel & Hirt, 2004). Actually common transcription factors involved in the tolerance against drought, chilling, and oxidative stress have been identified (Yamaguchi-Shinozaki & Shinozaki, 2006). A reduction of chilling injury by UV-C treatments has been reported in stone fruits (González-Aguilar et al., 2004). While in peaches and nectarines the mealy symptoms accompanying CI are associated with alterations in cell wall metabolism (Lurie & Crisosto, 2005) the basis of CI in peppers is not completely understood.

3.3. Effect of UV-C treatments on the antioxidant system

Overproduction of ROS leads to irreversible alterations of cellular homeostasis. CI has been associated with oxidative damage.

Table 2
Chilling injury (CI index) and weight loss, in control and UV-C treated fruit (10 and 20 kJ/m^2) during storage at 0 °C.

		21 d at 0 °C
CI index	Control	3.3 ± 0.2 a
	10 kJ/m^2	2.3 ± 0.3 b
	20 kJ/m^2	2.3 ± 0.2 b
Weight loss (g/100 g FW)	Control	1.7 ± 0.1 b
	10 kJ/m^2	1.4 ± 0.2 c
	20 kJ/m^2	2.2 ± 0.3 a

Data are the mean ± SD of twenty seven values and different letters show significant differences in a least significant difference (LSD) test at $P < 0.05$.

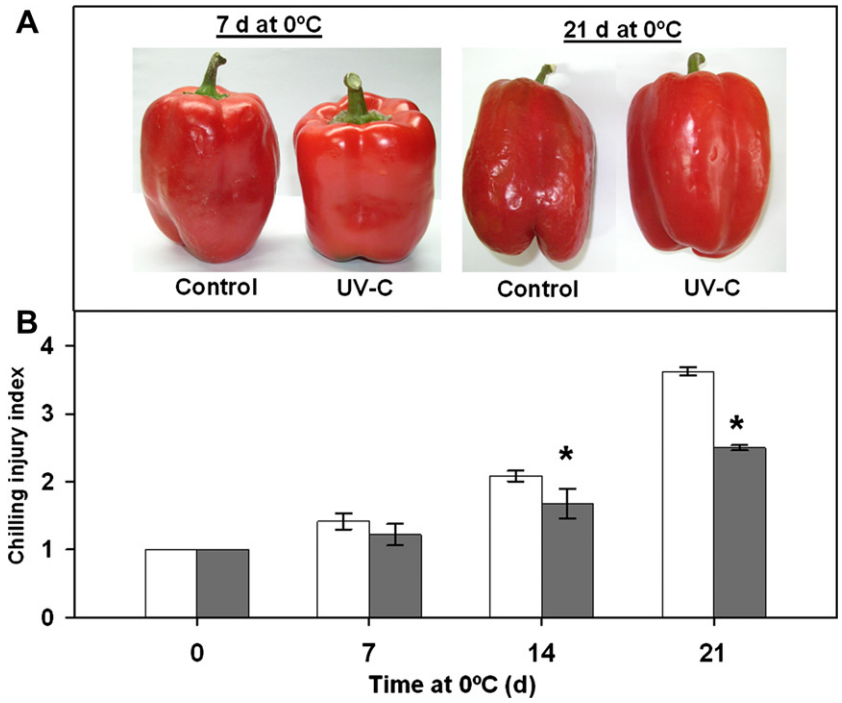

Fig. 1. A) Appearance and B) chilling injury index in control (□) and UV-C treated (10 kJ/m^2) peppers (■) during storage at 0 °C. Data are the mean of twenty seven values and the asterisk shows significant differences from the corresponding control at $P < 0.05$ according to a least significant difference (LSD) test.

In pepper, Purvis et al. (1995) found that superoxide production in mitochondria is detected a few days after storing fruit at chilling conditions. Treatments with antioxidants such as diphenylamine have been useful to reduce chilling injury suggesting the involvement of oxidative reactions in the disorder (Purvis, 2002). Consequently we decided to analyze if the reduction of CI development in UV-C treated peppers was associated with modifications in antioxidants levels.

3.3.1. Ascorbic acid (AA), dehydroascorbic acid (DHA) and DPPH radical scavenging capacity

Red peppers are an excellent source of AA (Deepa, Charanjit-Kaur, Singh, & Kapoor, 2007) with a concentration even higher than orange and kiwifruit. Despite of the nutritional relevance of that, AA might contribute to prevent oxidative damage in the commodity. The effect of UV-C treatment on AA and DHA contents in fruits has not been thoroughly studied. In seedlings, previous

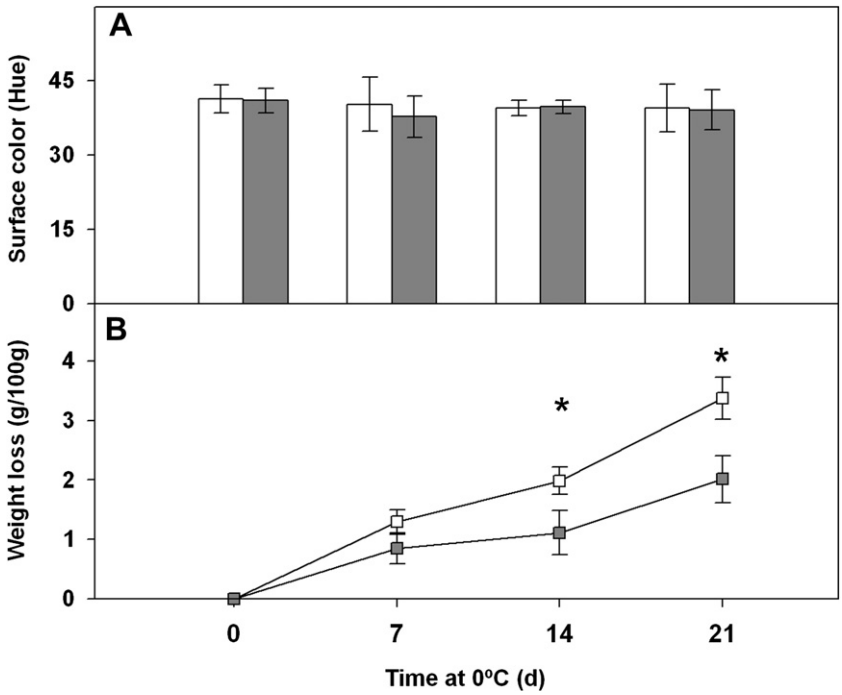

Fig. 2. A) Surface color (Hue) and B) weight loss in control (□) and UV-C treated (■) peppers (10 kJ/m^2) during storage at 0 °C. Data are the mean of twenty seven values and the asterisk shows significant differences from the corresponding control at $P < 0.05$ according to a least significant difference (LSD) test.

work found that exposure to UV-B and UV-C radiation resulted in an increased accumulation of AA and DHA (Nasibi & M-Kalantari, 2005). In the present study, at the beginning of the storage period AA content was close to 800 μg/g in both control and UV-C treated peppers (Table 3). During the first week of storage, AA content increased slightly in both control and UV-C treated fruit. AA has been reported to increase during bell pepper development (Martínez, López, González-Raurich, & Bernardo Alvarez, 2005). Even though the fruit was held at 0 °C, the 5% increase observed during the first 7 d of storage could be associated with a slight progression of fruit development. Later on, no variations in AA were detected in UV-C treated fruits, while a reduction was found in the control after 21 d of storage. Since AA is highly prone to oxidation upon wounding, the higher reduction in control fruit might be related to the increased injury found (Fig. 1A). DHA content represented less than 0.5% of the vitamin C pool and did not show great variations during storage (Table 3). At the end of the storage period even though AA content decreased in control fruit, a concomitant accumulation of DHA was not found suggesting that further degradation rapidly occurred.

On analyzing the DPPH radical scavenging capacity, immediately after UV-C treatment or after 1 week at 0 °C no differences were detected between control and treated fruits (Table 3). In contrast, Costa et al. (2006) and González-Aguilar, Zavaleta-Gatica, and Tiznado-Hernández (2007) found that UV-C treatment increased total antioxidants in broccoli and fresh-cut mango. After 14 or 21 d of storage slightly higher DPPH radical scavenging capacity was found in UV-C treated fruit (Table 3). UV-C treatments did not cause marked modifications in AA or DPPH radical scavenging capacity at early stages of CI development. UV-C treated peppers maintained slightly higher AA and DPPH radical scavenging capacity mainly when the manifestation of the disorder was already advanced.

3.3.2. SOD, GPX, APX and CAT activities

To determine if UV-C exposure was altering fruit redox metabolism we evaluated superoxide dismutase (SOD), guaiacol peroxidase (GPX), catalase (CAT) and ascorbate peroxidase (APX) activities. In plant cells, O_2^- might be consumed by SOD while enzymes such as GPX and APX or CAT may be involved in hydrogen peroxide removal. Given that the goal was to identify whether or not these enzymes were associated with the change in susceptibility to CI caused by UV treatments, the analysis was performed during the initial 14 d of storage in which the disorder became visible. Initially both control and UV-C treated fruit showed no differences in SOD activity. However after 7 d of storage SOD activity was significantly higher in the irradiated peppers (Table 4). Afterwards SOD decreased and no differences were found. Erkan et al. (2008) found that UV-C irradiation induced SOD activity in strawberry. Cao et al. (2009) reported that methyl jasmonate treatments, which reduced CI in loquat, reduced O_2^- and H_2O_2 production and increased SOD.

Table 4
Superoxide dismutase (SOD), guaiacol peroxidase (GPX), ascorbate peroxidase (APX) and catalase (CAT) activity in control and UV-C treated peppers (10 kJ/m^2) during storage at 0 °C for 14 d.

		Time at 0 °C (d)		
		0	7	14
SOD (EAU/mg protein)	Control	105.4 ± 5.9	113.9 ± . 9.1	90.9 ± 1.2
	UV-C	109.8 ± 0.9	138.7 ± 10.5*	92.9 ± 2.6
GPX (EAU/mg protein)	Control	15.9 ± 0.3	23.9 ± 1.3	13.9 ± 0.6
	UV-C	18.2 ± 0.5	18.0 ± 0.1*	12.5 ± 0.1
APX (EAU/mg protein)	Control	1.04 ± 0.03	1.20 ± 0.05	0.82 ± 0.05
	UV-C	1.31 ± 0.02*	1.37 ± 0.06*	1.00 ± 0.04*
CAT (EAU/mg protein)	Control	23.1 ± .1.0	32.2 ± 0.8	13.7 ± .1.5
	UV-C	44.2 ± 1.7*	38.9 ± 2.2*	23.2 ± .0.2*

ND = Non determined.
Data are the mean ± SD of six values issued from at least two different extractions of fresh tissue and the asterisk shows significant differences from the corresponding control in a least significant difference (LSD) test at $P < 0.05$.

Phenol peroxidases (PODs) are involved in lignin biosynthesis and wound healing. In the present study no differences in GPX were found after the UV-C treatment (Table 4). GPX increased in control fruit after 7 d at 0 °C showing higher activity than UV-C treated fruits. Later, GPX activity decreased and no difference was detected between control and UV-C treated peppers. These changes agree with those reported in UV-C irradiated tomatoes (Barka, 2001).

Regarding APX, higher activity was found in treated peppers after 0 or 7 d of storage at 0 °C. After 2 weeks APX decreased markedly in both control and treated fruit, but the latter still maintained higher activity (Table 4). Barka (2001) and Erkan et al. (2008) also found increased APX activity in UV-C treated tomato and strawberry. This enzyme is a member of the ascorbate–glutathione pathway, also known as the Halliwell-Asada cycle and is considered an important player in the scavenging of H_2O_2 in the chloroplast, cytosol and apoplast (Mittler, 2002). It is worth mentioning that the higher *in vitro* APX activity found in UV-C treated fruit did not correlate with changes in the levels of AA and DHA (Table 3). This might imply either that this increment in the enzymatic activity did not result in higher peroxide removal in *vivo* with the consequent accumulation of DHA or that the AA regenerating system was highly active. It has been reported that UV-C treatments enhanced the monodehydroascorbate reductase (MDAR), and dehydroascorbate reductase (DHAR) activities (Erkan et al., 2008).

Similar to APX, immediately after the treatment, UV-C treated fruit showed higher CAT activity (Table 4). The differences remained after 7 d at 0 °C. Finally as it was found for all the

Table 3
Ascorbic acid (AA), dehydroascorbic acid (DHA) and DPPH radical scavenging capacity in control and UV-C treated peppers (10 kJ/m^2) during storage at 0 °C.

		Time at 0 °C (d)			
		0	7	14	21
AA (μg/g FW)	Control	806 ± 18	847 ± 22	ND	763 ± 25
	UV-C	810 ± 21	847 ± 20	ND	838 ± 30*
DHA (μg/g FW)	Control	2.45 ± 0.3	2.50 ± 0.2	ND	2.39 ± 0.2
	UV-C	2.33 ± 0.5	2.63 ± 0.2	ND	3.01 ± 0.3*
DPPH radical scavenging capacity (1/g FW)	Control	189 ± 4.8	250 ± 3.6	232 ± 9.3	240 ± 5.8
	UV-C	195 ± 2.8	236 ± 1.1	251 ± 6.1*	259 ± 7.8*

ND = Non determined.
Data are the mean ± SD of four values for AA and DHA and six values for DPPH radical scavenging capacity, issued from at least two different extractions of fresh tissue and the asterisks show significant differences from the corresponding control in a least significant difference (LSD) test at $P < 0.05$. 195 ± 2.8.

enzymes studied the activity decreased after 14 d at low temperature. However, UV-C treated fruits still maintained higher CAT activity than control fruit. CAT is present in peroxisomes and it has been described that UV-C radiation can induce the proliferation of these organelles (Mittler, 2002). However, the dramatic and rapid increase in CAT seems more likely to have resulted from activation of pre-existing enzyme. Yang and Poovaiah (2002) reported that some plant catalases could be activated by the ubiquitous calcium-binding protein calmodulin and suggested that this could be important in responses to environmental stimuli. Sala and Lafuente (1999) found that changes in catalase induced by pre-storage heat treatments, correlated with reduced CI damage during storage in citrus. Cao et al. (2009) recently showed that methyl jasmonate treatments reduced CI, resulted in increased activity of redox scavenging enzymes. In strawberry, it has been shown that UV radiation can modulate several antioxidative enzymes (Erkan et al., 2008). Overall, the present work shows that exposure to 10 kJ/m^2 UV-C radiation can reduce CI in red bell peppers. The treatments do not cause marked modifications in DPPH radical scavenging capacity or AA content. In contrast UV-C treatments increase the activity of enzymes involved in the detoxification of superoxide and hydrogen peroxide such as SOD, CAT and APX during early storage at chilling temperatures.

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