

# The effect of harpin on shelf life of peppers inoculated with *Botrytis cinerea*

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**Abstract** The preservation methods as an alternative to chemical control to prevent postharvest quality losses of peppers were examined. The efficacy of harpin treatments on peppers (*Capsicum annuum* L. cvs. ‘Demre’, ‘Yalova Charleston’ and ‘Sari Sivri’) was tested in the same conditions in two different years. Peppers grown in greenhouse were applied with four treatments consisting of harpin, *Botrytis cinerea*, harpin+*B. cinerea* and control. The harpin in *B. cinerea* treatments reduced the percentage of rotten fruit in cv. ‘Demre’ from 42.68% to 22.85%, in cv. ‘Yalova Charleston’ from 60.87% to 26.59% and in cv. ‘Sari Sivri’ from 32.83% to 12.82%. The harpin and harpin +*B. cinerea* peppers had a better overall appearance at the end of shelf-life. Changes in percentage of red fruit and fruit color at the end of shelf life proceeded more slowly in the harpin treated fruit. The treatments of harpin gave the best results in all three cultivars. Moreover, the values obtained from fruits subjected to harpin+*B. cinerea* were better than those of the fruits picked from the plants only subjected to *B. cinerea*. In the trials, harpin slowed down the changes leading to quality loss in fruits, in all cultivars. Thus, the positive effect of harpin was revealed more clearly especially in the fruits picked from the inoculated plants.

**Keywords** *Capsicum annuum* L · Colour · Gray mold · Messenger · Shelf life · Quality

## Introduction

Pepper (*Capsicum annuum* L.) is an important crop grown extensively both in the field and in the greenhouses with high consumption rate due to its importance in rich vitamin contents for human diet. High rates of yield and quality losses occur in pepper because of a number of diseases encountered during growth (Zitter 2011). Gray mold caused by *Botrytis cinerea*, a ubiquitous fungus distributed worldwide and reported to be a pathogen of plants in more than 200 genera, including *Capsicum* (Pernezny et al. 2003). The disease is common in pepper. It is generally more severe in plants grown in enclosures that maintain high relative humidity, such as plant beds or greenhouses, but it is also a threat to field-grown pepper crops. Furthermore, *B. cinerea* causes a postharvest decay of pepper (Pernezny et al. 2003).

Use of plant activators, which have been developed against plant diseases in recent years, has become common with the developments in biological agriculture practices (Terry and Joyce 2004). One of these bioactivators is the messenger consisting of harpin. Harpin is a new bioactivator that may be used as an alternative to control insects and fungi as well as to increase yield and quality (Gang and Liu 1999; Mayer et al. 2001). It is isolated from *Erwinia amylovora*, the bacterial pathogen that causes fire blight disease. The ability of harpin to activate the growth and defense system of plants has encouraged the use of this product in Integrated Pest Management programs (Wei et al. 1992; Anonymous 2000). Previous studies show that harpin modulates genes expression involved in cell wall modifi-

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cations, stress response, signal transduction, membrane transporting and photosynthesis system (Boccaro et al. 2007; Chuang et al. 2010; Livaja et al. 2008). When harpin is applied to a plant, it binds to plant receptors. This binding process induces a cascade of responses that activate the expression of hundreds of genes, stimulating several of many genes that stimulate distinct biochemical pathways responsible for the enhancement of growth and pest resistance within the plant (Hunt and Ryals 1996; Ryals et al. 1996). Harpin may also aid in the suppression of insects and diseases as well as for the improvement of vigor, growth and increasing plant height, biomass fruit size, fruit quality and overall yield and stress tolerance (Copping and Menn 2000; Bednarz et al. 2002; Anonymous 2004).

Since the moisture content of fresh fruits and vegetables is more than 80%, they are classified as highly perishable commodities (Orsat et al. 2006). Keeping the product fresh is the best way to maintain its nutritional value, but most storage techniques require low temperatures, which are difficult to maintain throughout the distribution chain (Sagar and Kumar 2010). To regulate the marketing and to get higher remuneration, it is necessary to prolong shelf-life fruits and vegetables (Singh et al. 2010). The objective of this study was to determine the effect of harpin treatments on the shelf life and quality of the fruit picked from *B. cinerea* infected pepper plants.

## Materials and methods

*Plant materials, growth medium and growth conditions* The study was conducted at the Uludag University, Research and Training Greenhouse (glasshouses with automatically temperature and humidity control) and Cold Storage Unit in the same conditions in two different years. Seedlings produced from surface disinfected seeds were grown in 1.5 L capacity pots (one seedling per pot) containing soil and sand mixture (in 4/1 ratio) in greenhouse with a day/night temperature regime of  $22\pm 2/18\pm 2$  °C and 16 h photoperiod. Hand-harvest was started when fruit reached maturity, 136 days after pot transplanting in first year (15 May) and 140 days after transplanting in second year (19 May). Fruit were harvested at 15-day intervals (total five harvests). Fruit were sorted for uniform size, without wounds or rots.

The pepper cultivars ‘Demre’ (dark green color, thick fruit wall, sweet, long type pepper), ‘Yalova Charleston’ (yellow-green color, thick fruit wall, sweet, charleston type pepper) and ‘Sari Sivri’ (yellow-green color, sweet, long type pepper) (Anonymous 2003) which are widely grown in Turkey under protected cultivation were used in the trials. There were four treatments for each cultivar in the trials: 1) Harpin: The plants treated only with harpin, 2)

*B. cinerea*: The plants inoculated with only artificial pathogen, 3) Harpin+*B. cinerea*: The plants treated both with harpin and artificial pathogen inoculum, 4) Control: The plants treated only with water.

*Harpin treatments* In the trials, foliar applications of the harpin were made three times per season, at 50 g (3% a.i.) 100 L<sup>-1</sup> water. First application was applied when the plants were 15 days old, and the second and the third applications were made at 14-day intervals at a rate of 6–6.5 mg plant<sup>-1</sup> a.i. (Akbulduk et al. 2004).

*Inoculation of the plants with B. cinerea* One pathogenic isolate of *B. cinerea* obtained from pepper was cultured in petri plates containing PDA medium at 25 °C for 10 days. Leaves were inoculated with 10 mL of a conidial suspension (10<sup>6</sup> conidia m L<sup>-1</sup>) per plant using a small-calibrated hand sprayer (1.5 L capacity). Two-month-old plants (harpin-treated, non-treated) were inoculated with *B. cinerea*. After inoculation, the plants were held in closed polyethylene bags for 1 day and the disease caused by the pathogen progressed over a 2-month period.

*Investigated quality parameters* The following changes were examined in the fruit samples under shelf life conditions ( $22\pm 2$  °C and  $60\pm 5\%$  RH). Weight loss during shelf life was determined by weighing samples after harvest and at 5-day intervals during shelf life on a precision balance (Sartorius Co., Göttingen, Germany). Fruit were kept for 30 days under shelf life conditions following harvest in order to determine the percentage of rotten fruits. Number of decayed fruits was determined in comparison to the total number of fruits as a result of the measurements made during shelf life and the decay ratios were calculated in percentages. Changes in the external appearances during shelf life were evaluated by a jury of five persons (as 1–2 unusable, 3–4 unsalable, 5–6 salable, 7–8 good, 9–10 very good). The fruits were kept for 30 days under shelf life conditions in order to determine the percentage of red fruit. Samples that were more than 50% red area were classified as red. Number of fruits with red color was determined as a result of the measurements made at 5-day intervals during shelf life and the red fruit rates were determined in percentages. The colors of pepper fruits were determined by two readings on the two symmetrical faces of the fruit in each replicate, using a colorimeter (CR300, Konica-Minolta, Osaka, Japan) calibrated with a white standard tile.

*Experimental design* Anova procedures for a randomized complete block design were used to test for experimental factors (block and treatment). Treatment differences in weight loss, percentage of rotten fruit, overall appearance, percentage of red fruit, and fruit color were gauged by LSD

( $P < 0.05$ ). The analyses were carried out in 3 replicates so that there would be 30 pepper fruits in each replicate. Besides, analyses were carried out in the fruits taken at 5-day intervals during shelf life. Fruits were kept under shelf life conditions for 30 days after every harvest period. Pepper cultivars used in the study were evaluated independently. The experiment was repeated in the same conditions in two different years.

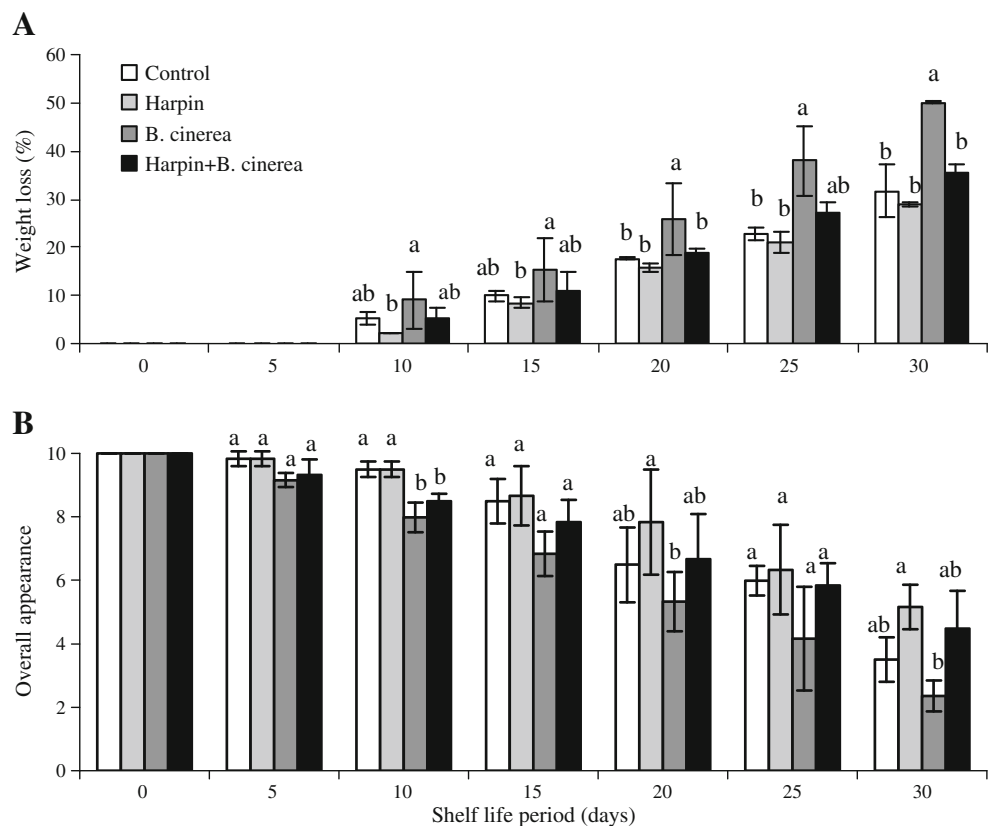
**Results and discussion**

**Weight loss** Fruit weight losses increased in parallel to the prolongation of shelf life period (Figs. 1a, 2a and 3a), especially after 10 days-shelf life in all treatments. The lowest weight loss was recorded in fruits picked from plants treated with harpin, whereas the highest weight loss was observed in *B. cinerea* treated fruits in three cultivars. Moreover, the weight losses in peppers treated with harpin+*B. cinerea* (35.28% in ‘Demre’, 25.45% in ‘Yalova Charleston’) were similar to control (31.68% in ‘Demre’, 28.45% in ‘Yalova Charleston’) in cvs. ‘Demre’, ‘Yalova Charleston’ at the end of shelf life. In the same period, in cv. ‘Sari Sivri’, harpin+*B. cinerea* (33.98%) showed less weight losses than control treatment (43.53%). Fruits treated with preharvest harpin are believed to have reduced water loss due to low rotten fruit

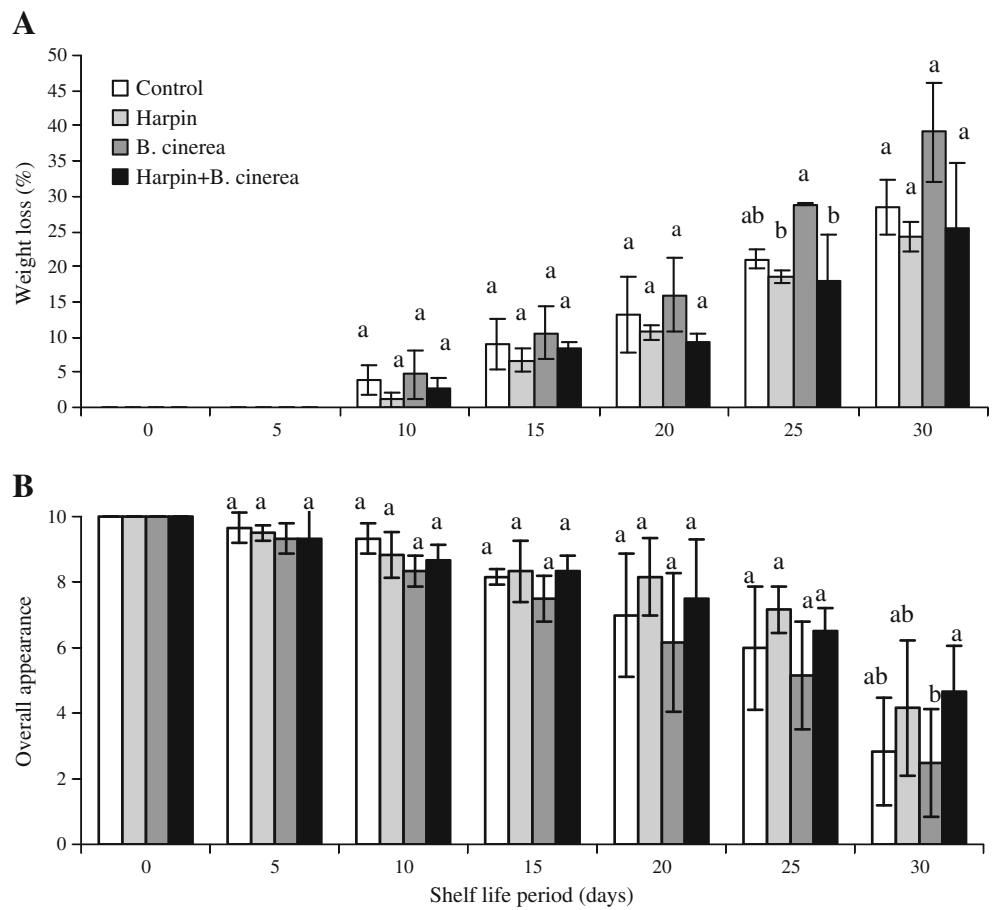
during the shelf life period. Weight loss and fruit decay proceeded more slowly in the fruits exposed to harpin. The suppression of fungal growth by harpin, reduced fruit weight losses. Thompson (1998) observed weight losses up to 20–25% in peppers stored under normal atmosphere conditions, whereas losses did not exceed 3–5% in other storage techniques. Therefore, the weight loss during shelf life period may be higher because of high temperature and low humidity. Heavy water loss occurred in the fruits subjected to *B. cinerea* at the end of day 30. In these treatments, the overall appearance and quality values of pepper fruits diminished with the prolonging shelf life, probably depending on the water loss in fruits. This finding suggested increasing water in the fruits treated with *B. cinerea*. Our results are similar to those of Jaxsens et al. (2002). In addition, Ozden and Bayindirli (2002) and Ayrancı and Tunc (2004) have used various treatments such as semperfresh (sucrose ester) which inhibited the weight loss in peppers, and they claimed that more successful results could be obtained by combining these treatments with controlled atmosphere storage.

**Percentage of rotten fruit** Percentage of rotten fruit increased towards the end of shelf life. Given the percentages of rotten fruit at 30 days after harvest, the highest percentage of rotten fruit was recorded from the fruits infected with *B. cinerea* (42.68% in cv. ‘Demre’, 60.87% in cv. ‘Yalova Charleston’ and 32.83% in cv. ‘Sari

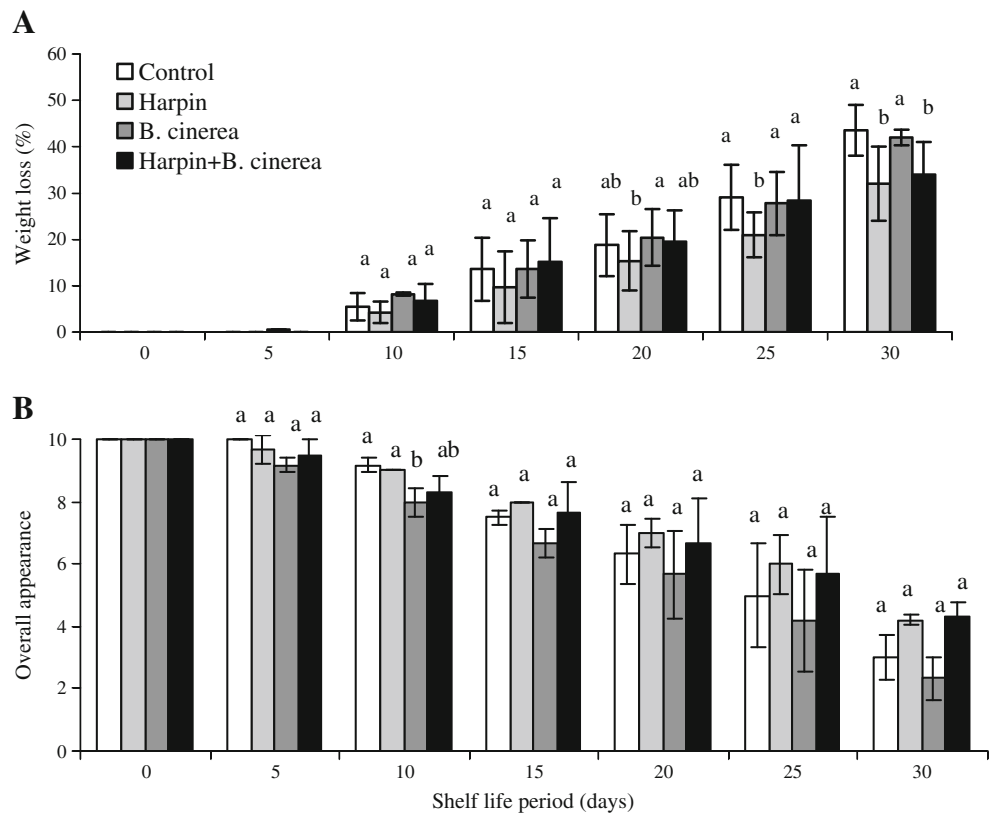
**Fig. 1** Changes in weight loss (a) and overall appearance (b) during shelf life of pepper fruits cv. ‘Demre’ treated with harpin and *B. cinerea* (Different letters in the same column indicate significant differences  $p \leq 0.05$  LSD, least significant difference ( $n=30$ ) average of 2 years)



**Fig. 2** Changes in weight loss (a) and overall appearance (b) during shelf life of pepper fruits cv. ‘Yalova Charleston’ treated with harpin and *B. cinerea* (Different letters in the same column indicate significant differences  $p \leq 0.05$  LSD, least significant difference ( $n=30$ ) average of 2 years)



**Fig. 3** Changes in weight loss (a) and overall appearance (b) during shelf life of pepper fruits cv. ‘Sari Sivri’ treated with harpin and *B. cinerea* (Different letters in the same column indicate significant differences  $p \leq 0.05$  LSD, least significant difference ( $n=30$ ) average of 2 years)



Sivri’) (Tables 1, 2 and 3). Rotting commenced at the end of shelf life in cv. ‘Sari Sivri’ fruits inoculated with *B. cinerea*, whereas rotting appeared in cvs. ‘Demre’ and ‘Yalova Charleston’ fruits beginning from the 20th day of shelf life. Furthermore, the rotten ratio was lower in cv. ‘Sari Sivri’ fruits compared to cvs. ‘Demre’ and ‘Yalova Charleston’. In general, harpin (18.81% in cv. ‘Demre’, 31.05% in cv. ‘Yalova Charleston’, 7.90% in cv. ‘Sari Sivri’) and harpin +*B. cinerea* (24.37 and 21.33% in ‘Demre’, 23.06 and

30.11% in ‘Yalova Charleston’, 20.00 and 5.65% in ‘Sari Sivri’) treatments resulted in lowest percentage of rotten fruits and the peppers had a better overall appearance at the end of shelf life. The severity of rotting in the fruit was significantly reduced especially when the fruit was treated with harpin. It is possible that resistance mechanisms triggered by harpin during the preharvest treatments remained active during the long storage period due to the effect low temperature on slowing cell metabolism, conse-

**Table 1** Changes in percentage of rotten fruit, percentage of red fruit and fruit colour properties during shelf life of pepper fruits cv. ‘Demre’ treated with harpin and *B. cinerea* (average of 2 years)

Shelf life period (days)	Treatment	Percentage of rotten fruit (%)	Percentage of red fruit (%)	Fruit colour		
				L (brightness)	a (greenness)	b (yellowness)
0	Control	0.0	0.0	41.37 a	-13.31 a	18.59 a
	Harpin	0.0	0.0	40.41 a	-13.68 a	18.48 a
	<i>B. cinerea</i>	0.0	0.0	41.61 a	-12.01 a	18.80 a
	Harpin+ <i>B. cinerea</i>	0.0	0.0	41.50 a	-12.67 a	18.33 a
LSD		–	–	5.36	3.77	5.57
5	Control	0.0	0.0	40.04 a	-13.15 a	19.62 a
	Harpin	0.0	0.0	39.85 a	-12.97 a	17.91 a
	<i>B. cinerea</i>	0.0	0.0	39.94 a	-11.98 a	19.28 a
	Harpin+ <i>B. cinerea</i>	0.0	0.0	39.49 a	-12.15 a	18.61 a
LSD		–	–	3.42	3.67	5.96
10	Control	0.0	0.0 b	39.12 a	-12.90 a	20.44 a
	Harpin	0.0	0.0 b	39.58 a	-12.76 a	17.81 a
	<i>B. cinerea</i>	0.0	29.37 a	38.47 a	-10.57 a	20.98 a
	Harpin+ <i>B. cinerea</i>	0.0	19.34 ab	38.45 a	-10.30 a	18.24 a
LSD		–	28.04	4.48	6.32	8.51
15	Control	0.0	22.45 ab	38.54 a	-12.20 b	20.72 a
	Harpin	0.0	0.0 b	39.16 a	-11.85 ab	17.66 a
	<i>B. cinerea</i>	0.0	35.59 a	37.95 a	-10.28 ab	21.36 a
	Harpin+ <i>B. cinerea</i>	0.0	31.86 ab	39.55 a	-8.73 a	18.32 a
LSD		–	32.14	3.89	3.30	9.25
20	Control	0.0 b <sup>a</sup>	28.04 ab	37.95 a	-7.97 ab	21.31 a
	Harpin	0.0 b	0.0 b	38.88 a	-11.24 b	17.43 a
	<i>B. cinerea</i>	5.69 a	52.72 a	37.33 a	-5.49 a	21.74 a
	Harpin+ <i>B. cinerea</i>	1.61 b	42.20 a	39.14 a	-7.76 ab	18.46 a
LSD		2.84	30.92	5.08	3.56	9.57
25	Control	3.92 b	35.93 ab	38.66 b	-1.02 bc	23.88 a
	Harpin	5.12 b	17.33 b	43.35 a	-7.33 c	18.79 a
	<i>B. cinerea</i>	14.03 a	58.23 a	35.83 b	8.53 a	24.75 a
	Harpin+ <i>B. cinerea</i>	4.96 b	66.07 a	42.27 a	2.94 ab	18.59 a
LSD		7.17	39.95	3.35	8.25	12.35
30	Control	25.10 b	64.91 ab	40.02 ab	5.08 b	25.47 a
	Harpin	18.81 c	33.07 b	44.02 a	-4.54 c	18.93 a
	<i>B. cinerea</i>	42.68 a	74.26 a	34.93 b	13.01 a	26.69 a
	Harpin+ <i>B. cinerea</i>	22.85 bc	78.49 a	39.25 ab	6.08 b	19.88 a
LSD		5.52	35.45	8.80	2.80	12.35

<sup>a</sup> Different letters in the same column indicate significant differences  $p \leq 0.05$  LSD, least significant difference ( $n=30$ )

**Table 2** Changes in percentage of rotten fruit, percentage of red fruit and fruit colour properties during shelf life of pepper fruits cv. ‘Yalova Charleston’ treated with harpin and *B. cinerea* (average of 2 years)

Shelf life period (days)	Treatment	Percentage of rotten fruit (%)	Percentage of red fruit (%)	Fruit colour		
				L (brightness)	a (greenness)	b (yellowness)
0	Control	0.0	0.0	55.54 a	-12.64 a	23.68 a
	Harpin	0.0	0.0	57.19 a	-13.53 a	24.54 a
	<i>B. cinerea</i>	0.0	0.0	59.02 a	-12.94 a	25.65 a
	Harpin+ <i>B. cinerea</i>	0.0	0.0	57.18 a	-12.85 a	25.50 a
LSD		–	–	3.87	3.89	4.30
5	Control	0.0	0.0	55.50 a	-12.24 a	24.29 a
	Harpin	0.0	0.0	57.11 a	-13.24 a	27.26 a
	<i>B. cinerea</i>	0.0	0.0	56.68 a	-11.58 a	26.21 a
	Harpin+ <i>B. cinerea</i>	0.0	0.0	56.28 a	-12.00 a	26.04 a
LSD		–	–	3.33	3.12	3.04
10	Control	0.0	0.0 b	55.65 a	-11.34 b	24.88 a
	Harpin	0.0	0.0 b	56.21 a	-12.65 b	23.96 a
	<i>B. cinerea</i>	0.0	8.97 a	56.81 a	-2.85 a	25.78 a
	Harpin+ <i>B. cinerea</i>	0.0	14.56 a	56.85 a	-6.84 ab	25.68 a
LSD		–	6.39	3.11	6.45	4.63
15	Control	0.0	0.0 b	54.85 ab	-11.16 b	25.64 a
	Harpin	0.0	0.0 b	56.72 a	-12.00 b	24.53 a
	<i>B. cinerea</i>	0.0	14.70 a	53.74 b	-1.01 a	25.93 a
	Harpin+ <i>B. cinerea</i>	0.0	20.11 a	56.14 ab	-3.14 a	25.76 a
LSD		–	7.75	2.48	7.54	4.13
20	Control	12.61 a <sup>a</sup>	10.70 b	53.75 b	-10.52 b	25.92 a
	Harpin	2.78 b	0.0 c	57.57 a	-11.18 b	25.12 a
	<i>B. cinerea</i>	15.12 a	18.46 a	54.85 ab	-4.62 a	26.42 a
	Harpin+ <i>B. cinerea</i>	4.78 b	21.54 a	56.52 ab	-4.31 a	26.24 a
LSD		4.01	4.28	3.00	4.35	5.48
25	Control	29.41 b	23.89 b	54.41 bc	2.21 b	26.28 a
	Harpin	10.07 d	14.22 c	59.26 a	-2.90 c	26.00 a
	<i>B. cinerea</i>	44.77 a	27.05 b	52.37 c	20.67 a	27.38 a
	Harpin+ <i>B. cinerea</i>	19.85 c	36.36 a	56.25 ab	17.91 a	25.47 a
LSD		4.64	4.31	3.62	4.23	4.05
30	Control	45.63 b	32.27 c	55.04 b	7.21 c	26.64 a
	Harpin	31.05 c	36.63 c	61.66 a	4.51 c	26.46 a
	<i>B. cinerea</i>	60.87 a	96.17 a	50.31 c	24.32 a	28.13 a
	Harpin+ <i>B. cinerea</i>	26.59 c	76.64 b	53.63 bc	19.02 b	26.40 a
LSD		5.19	10.01	3.68	4.05	4.88

<sup>a</sup> Different letters in the same column indicate significant differences  $p \leq 0.05$  LSD, least significant difference ( $n=30$ )

quently reducing the degradation of resistance (Chen et al. 1998; Capdeville et al. 2003; Leon et al. 2007). Our results are in accordance with the previous studies. The spoilage ratios of the fruits taken from harpin+*B. cinerea*-treated plants were much lower compared with those picked from *B. cinerea*-treated plants. Capdeville et al. (2002) successfully reduced the disease progress curve of blue mold by applying harpin on apples and similar effect was also

reported on controlled atmosphere stored fruit. In another study, spraying apple trees with harpin a few day before harvest was a promising strategy for reducing blue mold decay in stored apples (Capdeville et al. 2003). In California, Tubajika et al. (2007) showed that plants treated with harpin were less likely to become infected with *X. fastidiosa* and had a lower incidence of PD symptoms than untreated control plants.



**Table 3** Changes in percentage of rotten fruit percentage of red fruit and fruit colour properties during shelf life of pepper fruits cv. ‘Sari Sivri’ treated with harpin and *B. cinerea* (average of 2 years)

Shelf life period (days)	Treatment	Percentage of rotten fruit (%)	Percentage of red fruit (%)	Fruit colour		
				L (brightness)	a (greenness)	b (yellowness)
0	Control	0.0	0.0	59.62 a	−12.80 a	26.34 a
	Harpin	0.0	0.0	60.21 a	−12.97 a	26.06 a
	<i>B. cinerea</i>	0.0	0.0	59.55 a	−11.72 a	26.56 a
	Harpin+ <i>B. cinerea</i>	0.0	0.0	60.30 a	−12.15 a	27.32 a
LSD		–	–	2.96	2.91	6.18
5	Control	0.0	0.0	58.87 a	−12.29 a	27.08 a
	Harpin	0.0	0.0	58.72 a	−12.74 a	26.61 a
	<i>B. cinerea</i>	0.0	0.0	58.37 a	−11.33 a	27.93 a
	Harpin+ <i>B. cinerea</i>	0.0	0.0	59.61 a	−12.25 a	28.52 a
LSD		–	–	5.92	1.84	2.99
10	Control	0.0	0.0	58.15 a	−11.85 a	28.29 a
	Harpin	0.0	0.0	58.44 a	−12.41 a	25.58 a
	<i>B. cinerea</i>	0.0	0.0	58.16 a	−10.37 a	28.52 a
	Harpin+ <i>B. cinerea</i>	0.0	0.0	58.89 a	−12.24 a	26.18 a
LSD		–	–	4.60	2.29	5.60
15	Control	0.0	0.0 c	57.60 a	−10.93 b	28.36 a
	Harpin	0.0	0.0 c	58.05 a	−11.37 b	25.12 b
	<i>B. cinerea</i>	0.0	19.92 a	56.24 a	−4.99 a	28.80 a
	Harpin+ <i>B. cinerea</i>	0.0	13.45 b	58.55 a	−11.09 b	26.56 ab
LSD		–	5.05	4.96	5.60	2.97
20	Control	0.0	7.90 d	55.91 a	−8.61 b	28.98 a
	Harpin	0.0	12.73 c	57.93 a	−8.72 b	25.74 a
	<i>B. cinerea</i>	0.0	26.89 a	54.93 a	3.24 a	28.97 a
	Harpin+ <i>B. cinerea</i>	0.0	20.45 b	56.59 a	−4.45 ab	26.53 a
LSD		–	2.79	5.58	8.17	3.50
25	Control	4.39 b <sup>a</sup>	15.81 b	54.95 a	−4.29 b	29.74 a
	Harpin	2.62 b	15.42 b	57.36 a	−4.23 b	26.45 b
	<i>B. cinerea</i>	10.09 a	33.78 a	53.16 a	7.20 a	26.47 b
	Harpin+ <i>B. cinerea</i>	2.08 b	29.99 a	56.02 a	−2.38 b	26.63 b
LSD		3.87	3.96	6.74	7.78	2.99
30	Control	13.84 b	28.06 c	55.73 ab	0.74 b	27.57 a
	Harpin	7.90 c	26.49 c	57.28 a	0.87 b	27.24 a
	<i>B. cinerea</i>	32.83 a	63.65 a	50.72 b	13.65 a	27.20 a
	Harpin+ <i>B. cinerea</i>	12.82 b	42.86 b	55.23 ab	−0.17 b	28.37 a
LSD		4.63	5.85	5.60	7.85	4.92

<sup>a</sup> Different letters in the same column indicate significant differences  $p \leq 0.05$  LSD, least significant difference ( $n=30$ )

**Overall appearance** In general, the overall appearance values of fruits along the shelf life diminished in parallel to the percentages of rotten and red fruits. These reductions proceeded faster in cv. ‘Demre’ (3.50 in control, 5.16 in harpin, 2.35 in *B. cinerea*, 4.49 in harpin+*B. cinerea*) (Fig. 1b) partially, compared with the other cultivars (2.83 in control, 4.16 in harpin, 2.49 in *B. cinerea*, 4.66 in harpin+*B. cinerea* for ‘Yalova Charleston’; 3.00 in control, 4.21 in harpin, 2.33 in *B. cinerea*, 4.33 in harpin+*B. cinerea* for

‘Sari Sivri’ (Figs. 2b and 3b). This cultivar was followed by cv. ‘Sari Sivri’ (Fig. 3b). The fact that harpin, one of the most commonly used bio activators developed using biotechnological techniques, increased the resistance against plant pests and diseases has been supported by previous studies (Gang and Liu 1999; Bednarz et al. 2002). In a different study, peppers were subjected to hot water treatment ( $55 \pm 1$  °C,  $12 \pm 2$  s), in this way fungi development was retarded and the overall appearance of peppers

was improved (Fallik et al. 1999). Some treatments like harpin retarded the fungi development, reduced the percentage of decayed fruits and improved the overall appearance values in peppers, as well as in some other crops.

**Percentage of red fruit** The chlorophyll evolution in the pepper varieties throughout their ripening is a subject which has also been studied by several authors due to the importance of their gradual transformation into carotenoids, responsible for color (Camara and Brangeon 1981; Ziegler et al. 1983; Carde et al. 1988). We determined that percentage of red fruit from inoculated with *B. cinerea* (74.26% for *B. cinerea*, 78.49% for harpin+*B. cinerea* in ‘Demre’, 96.17% for *B. cinerea*, 76.64% for harpin+*B. cinerea* in ‘Yalova Charleston’, 63.65% for *B. cinerea*, 42.86% for harpin+*B. cinerea* in ‘Sari Sivri’) were higher than non-inoculated with *B. cinerea* in the three cultivars (Tables 1, 2 and 3). Moreover, the lowest percentage of red fruit value was obtained from cv. ‘Sari Sivri’. *B. cinerea* inoculation increased red color change. Fruits treated with harpin treatment turned red more slowly than those treated with *B. cinerea*. Thus, harpin treatments without *B. cinerea* delayed fruit maturation and reduced fruit quality losses.

**Fruit color** The results of color parameters obtained from the trials are presented in Tables 1, 2 and 3 for L (brightness), a (greenness), b (yellowness) values, respectively. Fruit color parameters significantly varied among treatments. Increases and reductions in L values were observed during shelf life of peppers; the lightest color was obtained with harpin treatment after the shelf life. The decreased color change observed in *B. cinerea* was greater compared with the other treatments. Color change progressed (became dark, low L value) more rapidly in peppers subjected to *B. cinerea*. Moreover, significant L value losses occurred in ‘Demre’ compared with the other cvs. In harpin treated fruits, the highest loss of green color (*a*) was determined in *B. cinerea* (13.01 in ‘Demre’, 24.32 in ‘Yalova Charleston’, 13.65 in ‘Sari Sivri’) (Tables 1, 2 and 3). The *b* (yellow) values, which indicate the yellow color quantity, were lower in the fruits of plants subjected to harpin treatment compared with the control fruits in all cvs. The loss of green and yellow color was highest in *B. cinerea* treatment. Harpin application had less effect on color change when compared with other treatments. However, this value was high in inoculated with *B. cinerea* fruits. In all cultivars, *B. cinerea* caused a negative effect on color. On the other hand, harpin may be affected enzymatic degradation of chlorophyll. Accordingly, harpin treatments inhibited negative effect of *B. cinerea* inoculation. The color values of pepper fruits were affected by different criteria as previously reported by Gomez et al. (1998).

Furthermore, Martinez et al. (2003) applied potassium treatments at low rates during the preharvest period in order to retard the color changes.

Changes occurred in the physiological structure of plants following the harpin may positively affect the plant health and hence the fruit quality. Significant differences were determined in the changes in fruit quality criteria of the plants subjected to harpin or *B. cinerea*, compared with the fruit of the plants that were only inoculated, and harpin treatments were found to suppress the injury of *B. cinerea* fungus in the fruits. When the values related to the three cultivars used in the study were examined, cv. ‘Yalova Charleston’ was determined as the cultivars, which showed better response to the harpin treatment.

## Conclusions

Harpin treatments not only retarded the changes in quality criteria by affecting the plant physiologically, but also improved the resistance of peppers to diseases and pests. According to our results, optimum shelf life period in peppers could be prolonged and may be longer than a shelf life of 20 days, as suggested by Gonzalez-Aguilar and Tiznado (1993), considering the changes in the other quality parameters. However, the efficacy of other systemic acquired resistance (SAR) inducers in prolonging shelf life is still unknown. Further research is needed to determine the effects of such inducers on the fruit quality.

## References

- Akbadak N, Seniz V, Tezcan H (2004) Effect of harpin on yield and fruit quality of pepper grown in greenhouse conditions. *Acta Hortic* 729:267–270
- Anonymous (2000) United States Environmental Protection Agency (EPA). The messenger; a promising risk biopesticide. Pesticide Environmental Stewardship Program Update, 3 p
- Anonymous (2003) Agromar. [http://www.agromar.com.tr/tr/acik\\_tarla\\_sebze\\_tohumlari/biber.php](http://www.agromar.com.tr/tr/acik_tarla_sebze_tohumlari/biber.php) (Accessed 12 November 2010)
- Anonymous (2004) Biopesticides harpin. [http://www.epa.gov/opp00001/biopesticides/ingredients/factsheets/factsheet\\_006477.htm](http://www.epa.gov/opp00001/biopesticides/ingredients/factsheets/factsheet_006477.htm) (Accessed 12 November 2010)
- Ayrancı E, Tunc S (2004) The effect of edible coatings on water and vitamin C loss of apricots (*Armeniaca vulgaris* Lam.) and green peppers (*Capsicum annuum* L.). *Food Chem* 87:339–342
- Bednarz CW, Brown SN, Flanders TJ, Tankersley TB, Brown SM (2002) Effects of foliar applied harpin protein on cotton lint yield, fiber quality, and crop maturity. *Commun Soil Sci Plant Anal* 33:933–945
- Boccaro M, Schwartz W, Guiot E, Vidal G, Paepe RDe, Dubois A, Boccaro AC (2007) Early chloroplastic alterations analysed by optical coherence tomography during a harpin-induced hypersensitive response. *Plant J* 50:338–346



- Camara B, Brangeon J (1981) Carotenoid metabolism during chloroplast to chromoplast transformation in *Capsicum annuum* fruit. *Planta* 151:359–364
- Capdeville G, Wilson C, Beer SV, Aist JR (2002) Alternative disease control agents in harvest harpin treatments of apples induce resistance to blue mold. *J Phytopathol* 92:900–908
- Capdeville G, Beer SV, Watkins BC, Wilson C, Tedeschi OL, Aist JR (2003) Pre- and Post- Alternative disease control agents induce resistance to blue mold in harvested ‘Red Delicious’ apple fruit. *Plant Dis* 87:39–44
- Carde JP, Camara B, Cheniclet C (1988) Absence of ribosomes in *Capsicum* chromoplasts. *Planta* 173:1–11
- Chen CH, Lin HJ, Feng TY (1998) An amphipathic protein from sweet pepper can dissociate harpinPss multimeric forms and intensify the harpinPss-mediated hypersensitive response. *Physiol Mol Plant Pathol* 52:139–149
- Chuang H, Hamrak A, Chen YC, Hsu CM (2010) A harpin-induced ethylene-responsive factor regulates plant growth and responses to biotic and abiotic stresses. *Biochem and Bioph Res Co* 402:414–420
- Copping LG, Menn JJ (2000) Biopesticides: a review of their action, applications and efficacy. *Pest Manag Sci* 56:651–676
- Fallik E, Grinberg S, Alkalai S, Yekutieli O, Wiseblum A, Repev R, Beres H, Bar-Lev E (1999) A unique rapid hot water treatment to improve storage quality of sweet pepper. *Postharvest Biol and Technol* 15:25–32
- Gang LR, Liu FY (1999) Reduction of lesion growth rate of late blight plant disease in transgenic potato expressing harpin protein. *Sci China Ser C-Life Sci* 42:96–101
- Gomez R, Pardo EJ, Navarro F, Varon R (1998) Colour differences in paprika varieties (*Capsicum annuum* L.) cultivated in a greenhouse and in the open air. *J Sci Food Agric* 77:268–272
- Gonzalez-Aguilar G, Tiznado M (1993) Postharvest physiology of bell peppers stored in low density polyethylene bags. *LWT-Food Sci Technol* 26:450–455
- Hunt M, Ryals J (1996) Systemic acquired resistance signal transduction. *Crit Rev Plant Sci* 15:583–606
- Jacxsens L, Devlieghere F, Debevere J (2002) Temperature dependence of shelf-life as affected by microbial proliferation and sensory quality of equilibrium modified atmosphere packaged fresh produce. *Postharvest Biol Technol* 26:59–73
- Leon IP, Oliver JP, Castro A, Gaggero C, Bentancor M, Vidal S (2007) *Erwinia carotovora* elicitors and *Botrytis cinerea* activate defense responses in *Physcomitrella patens*. *BMC Plant Biol* 7:1–11
- Livaja M, Zeidler D, von Rad U, Durner J (2008) Transcriptional responses of *Arabidopsis thaliana* to the bacteria-derived PAMPs harpin and lipopolysaccharide. *Immunobiology* 213:161–171
- Martinez Y, Diaz L, Manzano J (2003) Influences of nitrogen and potassium fertilizer on the quality of ‘Jubiter’ pepper (*Capsicum annuum*) under storage. *Acta Hort* 628:135–140
- Mayer R, Weathersbee A, Moshe I, Hamed D (2001) In vivo modulation of a citrus B-1, 3-endoglucanase by benzo (1,2,3 thiadiazole-7-carbothioic acid s-methyl ester) (actigard). <http://www.ars.usda.gov> (Accessed 15 May 2006)
- Orsat V, Changrue V, Raghavan GSV (2006) Microwave drying of fruits and vegetables. *Stewart Post-Harvest Rev* 6:4–9
- Ozden C, Bayindirli L (2002) Effects of combinational use of controlled atmosphere, cold storage and edible coating applications on shelf life and quality attributes of green peppers. *Eur Food Res Technol* 214:320–326
- Pernezny K, Roberts PD, Murphy JF, Goldberg NP (2003) Compendium of pepper diseases. APS Press, 2003, 63 p
- Ryals JA, Neuenschwander UH, Willits MG, Molina A, Steiner HY, Hunt MD (1996) Systemic acquired resistance. *Plant Cell* 8:1809–1819
- Sagar VS, Kumar SP (2010) Recent advances in drying and dehydration of fruits and vegetables: a review. *J Food Sci Technol* 47:15–26
- Singh S, Singh AK, Joshi HK, Lata K, Bagle BG, More TA (2010) Effect of zero energy cool chamber and post-harvest treatments on shelf-life of fruits under semi-arid environment of Western India. Part 1. Ber fruits. *J Food Sci Technol* 47:446–449
- Terry AL, Joyce DC (2004) Elicitors of induced disease resistance in postharvest horticultural crops: a brief review. *Postharvest Biol Technol* 32:1–13
- Thompson AK (1998) Controlled atmosphere storage of fruits and vegetables. CABI Publishing, Wallingford, Oxon, OX10 8DE, UK, 1998, 278 p
- Tubajika KM, Civerolo EV, Puterka GJ, Hashim JM, Luvisi DM (2007) The effects of kaolin, harpin, and imidacloprid on development of Pierce’s disease in grape. *Crop Prot* 26:92–99
- Wei Z, Laby R, Zumoff C, Bauer D, Ho SY, Collmer A, Beer S (1992) Harpin, elicitor of the hypersensitive response produced by the plant pathogen *Erwinia amylovora*. *Science* 257:85–87
- Ziegler H, Shafer E, Schneider M (1983) Some metabolic changes during chloroplast-chromoplast transition in *Capsicum annuum*. *Physiol Veg* 21:485–494
- Zitter TA (2011) Pepper disease control—It starts with the seed. [http://vegetablemndonline.ppath.cornell.edu/NewsArticles/PepDisease\\_Con.htm](http://vegetablemndonline.ppath.cornell.edu/NewsArticles/PepDisease_Con.htm) (Accessed 03 March 2011)