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THE INFLUENCE OF COLORED SHADE NETS ON PEPPER QUALITY AFTER HARVEST – A POSSIBLE MODE-OF-ACTION

SUMMARY

In previous studies, we have found that Pearl and Yellow shade nets significantly reduced pepper fruit rot development after harvest. The significant low decay incidence in fruit harvested under Pearl and Yellow shade nets was explained by the low inoculum level of Alternaria spp. in the field, and inhibition of fungal sporulation, and/or by slowing the fruit ripening during its growth, which reduces fruit susceptibility to fungal infection in the field due to the scattered light, its quality and the ratio between the light spectrum, under those two shade nets (Goren *et al*., 2011). Therefore, the goal of this research was to shed more light on the influence of Pearl shade net on postharvest quality of two different commercial red pepper bell cultivars (cv. Romans and Vergasa). Evaluation of fruit quality was based on chlorophyll, carotenoids and antioxidant content after prolonged storage and marketing simulation. No significant differences were observed in fruit weight loss, firmness and sugar content of the two cultivars under the different nettings. However, some differences were observed between the cultivars. Fruit from the two cultivars which were grown under Pearl shade net had significantly lower decay incidence compared to fruit harvested under commercial black net, particularly cv. Vergasa. Fruit chlorophyll content under Pearl net was higher than in fruit grown under the black shade net, and was significantly higher in Vergasa than in Romans. The carotenoids content was significantly higher under the commercial black net than under the Pearl net, and was significantly higher in Vergasa than in Romans immediately after harvest, storage and shelf life simulation. Immediately after harvest and after storage and marketing simulation, the antioxidant (AOX) activity in Vergasa was significantly higher than Romans in fruit harvested under both shade nets. A significant increase in AOX activity was measured in Vergasa fruit harvested under Pearl net after storage and marketing simulation in comparison to black net and all Romans fruit harvested. In conclusion, Pearl net's influence on ripening inhibition, in higher AOX activity and carotenoids content, could reduce decay in pepper fruit and maintain better fruit quality after harvest.

Keywords: *Capsicum annuum*, postharvest, shelf life, storage

INTRODUCTION

Sweet bell pepper (*Capsicum annuum* L.) is an important export commodity in Israel, and more than 130,000 tons are harvested per year (Goren

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et al., 2011). Because of a growing consumer demand for peppers with improved flavor and nutritional characteristics, such as carotenoids, ascorbic acid and phenols (Deepa *et al.*, 2007), bell peppers are increasingly harvested at full color to achieve higher nutritional levels and market price (Frank *et al.*, 2001).

In Israel, in order ensure pepper quality during the summer and early fall; peppers are traditionally grown in semi-arid regions under black nets with 35%-40% shading to reduce sun radiation (Fallik *et al.*, 2009). In the past decade, development of photo-selective coloured shade nets provide better light filtration, and have their effects have been studied on pre-harvest physiological, entomological and pathological aspects in several horticultural crops (Shahak *et al.*, 2004; Shahak, 2008; Shahak *et al.*, 2009). The photo-selective coloured shade nets, depending on the thread pigmentation and knitting design, provide diverse mixtures of natural, unmodified light, along with spectrally-modified scattered light (Shahak *et al.*, 2004; Rajapakse and Shahak, 2007).

Goren *et al.* (2011) and Kong *et al.* (2013) reported that Pearl shade net significantly maintained sweet red pepper fruit quality after prolonged storage and shelf life simulation, compared with use of commercial black shade nets. The improved quality was due to low inoculum level of *Alternaria* spp. in the field, and inhibition of fungal sporulation. In addition, the shade net slowed fruit ripening during its growth as shown by external colour, which could reduce fruit susceptibility to fungal infection in the field (Goren *et al.*, 2011). The goals of this research were to evaluate the influence of Pearl shade net, compared to black shade net, on two main commercial red bell pepper cultivars as it was reported that cultivars' quality can be differed in their susceptibility to physiological and pathological deterioration after harvest (Maalekuu *et al.*, 2003, 2004; Smith *et al.*, 2006), and to shed more light on the fruit quality maintenance based on antioxidants contents after prolonged storage and marketing simulation.

MATERIAL AND METHODS

Plant materials and experimental design. Red sweet bell pepper (*Capsicum annuum* L.) cvs. ‘Romans’(as a reference for the 2007-2008 year) and ‘Vargasa’, were grown at the B’sor Experimental Station in the south-west of Israel(31.271° N; 34.389° E), using commercial cultivation practices, under two different colored shade-nets, as follows: Pearl and black (commercial shade net) with 35% relative shading (in PAR) (Polysack Plastics Industries, Nir-Yitzhak, Israel under the trade mark ChromaticNet), in four random replicates of 18 x 18 m each, all within one large horizontal net-house, 2.5 m high. Plants were planted toward the end of May, 2010, and harvested as described below. Fruits (~190 ± 10 g) without defects or diseases were harvested four times, between September to December, at ~80-85% red colour without calyx. Fruit were then treated as described by Fallik *et al.* (1999) and packed in three – 6.5 kg corrugated new carton. Fruit were stored at 7°C and RH 94% for 16 d + 3 d at 20°C (RH 70%)(Storage and shelf life simulation).

Quality parameters. Fruit quality parameters were evaluated at the end of the simulation as follows: *Weight loss* was expressed as percentage of weight loss from the initial weigh of ten fruits. Ten fruits were measured for *firmness* as described by Hamson (1952). Each fruit was placed horizontally between two flat surfaces and a 2 kg weight was loaded on top of the flat surface. A dial fixed to a graduated plate recorded the deformation of the fruit in millimetres. Full deformation was measured 15 s after exerting the force on the fruit, then the weight was removed and the residual deformation was measured 15 s later. Fruit was considered very firm with 0 – 1.5 mm deformation; firm = 1.6 – 3 mm deformation; soft = 3.1 – 4.5 mm deformation; very soft = above 4.6 mm deformation.

Total soluble solids (TSS) were measured on the same ten fruits tested for firmness, by squeezing out juice from fruits onto an Atago digital refractometer (Atago, Tokyo, Japan) and taking readings.

Decay incidence was considered once fungal mycelia appeared on fruit pericarp and/or calyx. Decay was expressed as a percentage of the total initial fruit number.

Antioxidants, chlorophyll & carotenoids - extraction and determination.

Nine fruits were taken from each treatment and three fruit from each treatment were sliced and pooled (three replicates). Samples were weighed, frozen in liquid N_2, lyophilized and kept at -80°C until further analysis. The lyophilized tissues were extracted according to Vinokur & Rodov (2006).

Antioxidant activity was evaluated by decolorization of the $ABTS^+$ radical cation. The radical was generated in acidified ethanol medium in order to allow the activities of both hydrophilic and lipophilic antioxidants. The medium comprised 16 mg of 2,2'-azino-bis-(3-ethylbenzothiazoline-6-sulphonic acid), diammonium salt ($ABTS^+$) and 4mg $K_2O_8S_2$ in acidified ethanol. Incubation of the reaction mixture at 45ºC for 60 minutes was sufficient for $ABTS^+$ generation. The decolorization test was performed in plastic cuvettes by adding 10 μL of test sample to 1 mL of acidified ethanol solution of $ABTS^+$ and comparing the optical density at 734 nm after 15 min of incubation at room temperature with that of a blank sample. The 1 mM solution of 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox, a water-soluble derivative of the vitamin E) was used as a standard, and the radical-scavenging activity of samples was expressed as Trolox equivalent antioxidant capacity (TEAC). The analyses were performed in triplicate (nine readings).

Carotenoids and chlorophyll were determined from the lipophilic fraction (Vinokur and Rodov, 2006). Carotenoids and chlorophyll were extracted four times with 3 ml of hexane (i.e., total of 12 ml), which were dried out under nitrogen. The sample was resuspended with 2 ml of 100% acetone and was measured in a spectrophotometer at 470, 645 and 662 nm. Chlorophyll A and B, and total carotenoids were calculated according to Lichtenthaler (1987) and expressed as mg/100 g FW.

The reagents were purchased from Sigma-Aldrich Co., Israel. The analyses were performed in triplicate (nine readings).

Statistical analysis. Four experiments (harvest) were conducted. All data were subjected to one or two-way Anova analysis at P = 0.05 using JMP6 Statistical Analysis Software Program (SAS Institute Inc., Cary, NC, USA) and the mean values for all data are presented (Sall *et al.*, 2001). Correlation coefficient values between decay development and AOX, carotenoids and chlorophyll contents were performedusing P value determined by Student t-test..

RESULTS

Decay incidence. Pepper fruit from the two cultivars that were grown under Pearl shade net had significantly lower percentage of decay incidence than pepper fruit that were grown under commercial black net (Fig. 1). Vergasa was found to have the lowest decay incidence (10% and 15% under Pearl and black nets, respectively, 33% decay reduction from the black net), compared to Romans (14% and 18% under Pearl and black nets, respectively, 22% decay reduction) (Fig. 1).

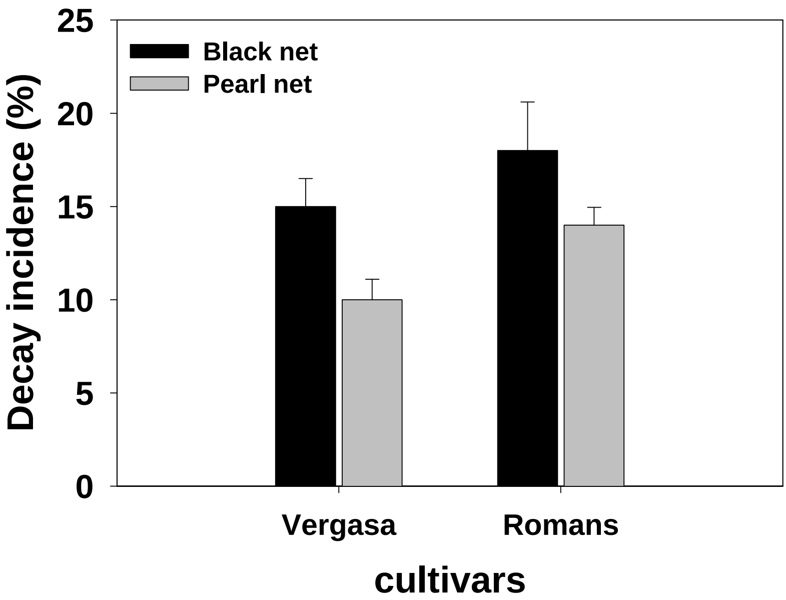

Figure 1. Effect of coloured shade nets on decay incidence on fruit of two pepper cultivars. Percentage of decay incidence was measured after 16 d at 7°C plus 3 d at 20°C (Means of four experiments ± SE, with three - 6.5 kg box per treatment).

Chlorophyll and carotenoids. Significant differences were obtained in total chlorophyll and carotenoids content between the commercial black net and Pearl net (Figs. 2 and 3). Immediately after harvest the chlorophyll content of pepper fruits grown under Pearl net was significantly higher than chlorophyll of fruit grown under the black net, and chlorophyll content in Vergasa was significantly higher than in Romans (1.35 and 0.65 mg/ 100 g FW, respectively)

(Fig. 2). After 16 d at 7°C and 3 d at 20°C, chlorophyll content significantly decreased in the two cultivars. In addition, the chlorophyll content under Pearl net was higher than in fruit grown under the black shade net, and significantly higher in Vergasa than in Romans (Fig. 2).

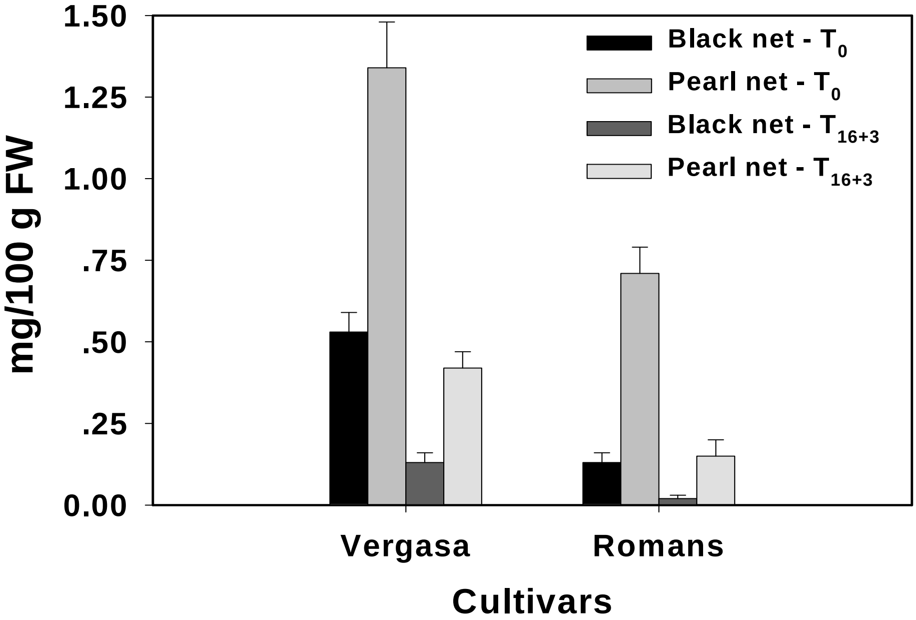

Figure 2. The influence of coloured shade nets on Chlorophyll A and B in fruit of two pepper cultivars immediately after harvest, and after 16 d at 7°C plus 3 d at 20°C (Means of four experiments ± S.E).

Carotenoids content was negatively correlated to chlorophyll content in the treated fruits (Fig. 3). Carotenoids content increased in the two red cultivars during storage and shelf life simulation, under black and Pearl nets. However, content was significantly higher under the commercial black net than Pearl net. Carotenoids content in Vergasa was significantly higher than in Romans immediately after harvest (35 and 30 mg/100 g FW, respectively) and after storage and shelf life simulation (53 and 50 mg/100 g FW, respectively) (Fig. 3).

Antioxidant activity (AOX). Immediately after harvest and after storage and marketing simulation, the antioxidant (AOX) activity in Vergasa was significantly higher than Romans, both in fruit harvested under commercial black net and Pearl net (Fig. 4). A significant increase in AOX activity in Vergasa fruit harvested under Pearl net after storage and marketing simulation, when compared to Vergasa fruit harvested under black net, or Romans fruit harvested under both nets (121, 105 and 82, 84 μM TE/g, respectively). There was only a slight increase in AOX activity in Romans after storage and marketing simulation.

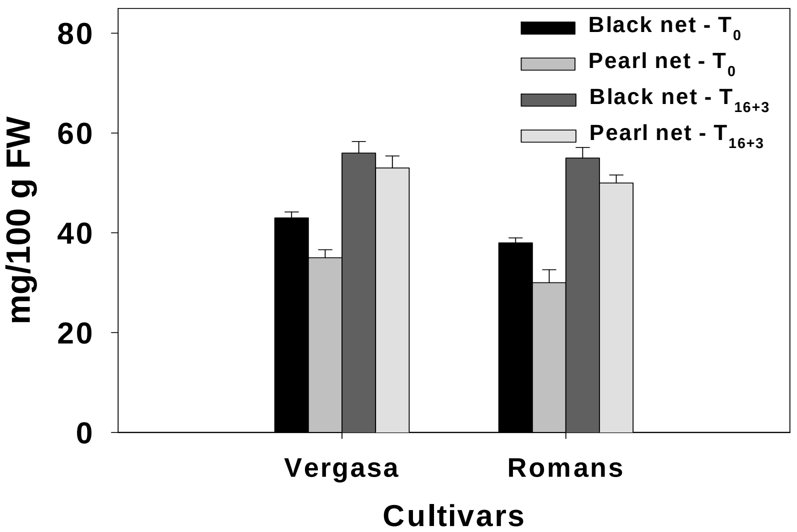

Figure 3. The influence of coloured shade nets on carotenoids content in fruit of two pepper cultivars immediately after harvest, and after 16 d at 7°C plus 3 d at 20°C (Means of four experiments ± S.E).

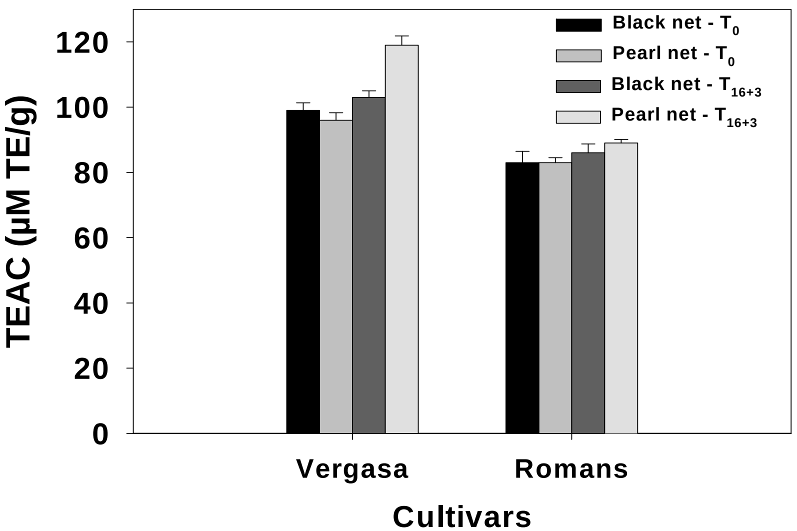

Figure 4. The influence of coloured shade nets on total antioxidants activity in fruit of three pepper cultivars immediately after harvest and after 16 d at 7°C plus 3 d at 20°C (Means of four experiments ± S.E)

Regression analysis. Regression analysis of decay development and AOX in both Vergasa and Romans grown under Pearl net showed strong positive association (r = 0.99 and r = 0.98, P = 0.007 and 0.01 for Vergasa and Romans, respectively) (Table 1). There was no significant correlation between decay development and the amount of chlorophyll, carotenoids and AOX in the two cultivars grown under the black shade net. A moderate correlation, yet not significant, was found between decay development and carotenoids in Vergasa grown under the Pearl shade net (r = 0.54, P = 0.45) (Table 1).

Table 1. Correlation coefficients of decay development and chlorophyll (Chlo), carotenoids (Caro) and antioxidants (AOX). P value determined by Student's t-test (n = 4)

Cultivar	Vergasa						Romans					
Shade net	Black			Pearl			Black			Pearl		
	Chlo	Caro	AOX	Chlo	Caro	AOX	Chlo	Caro	AOX	Chlo	Caro	AOX
r	-0.09	0.19	0.29	0.05	0.54	0.99	-0.65	-0.80	-0.34	0.23	-0.72	0.98
P value	0.91	0.81	0.71	0.94	0.45	0.007	0.34	0.19	0.66	0.76	0.28	0.019

Quality traits. Overall, netting colour did not present significant differences in fruit quality (weight loss, firmness and sugar content [TSS]) after 16 d storage and 3 additional d at 20°C (Table 2). There were, however, significant differences between the two cultivars. Vergasa had better quality traits, when compared to Romans, based on weight loss and firmness (Table 2).

Table 2. The influence of Pearl and black color shade nets on weight loss, firmness and TSS of three pepper fruit cultivars after shipping simulation of 16 days at 7°C plus 3 days at 20°C (means of four experiments)

Shade net /	Weight loss (%)[a]		Firmness (mm)[b]		TSS (%)[c]	
cultivar	Vergasa	Romans	Vergasa	Romans	Vergasa	Romans
Black	2.7 Ab[z]	3.5 Aa	2.4 Ab	2.9 Aa	7.2 Aa	6.8 Ab
Pearl	2.8 Ab	3.6 Aa	2.5 Ab	3.0 Aa	7.1 Aa	6.6 Ab

aWeight loss (%)
bFirmness – millimeter deformation (flexibility)
cPercent total soluble solids
zDifferent upper-case letters represent significant differences between the colored shade nets, and lower-case different letters represent significant differences between the two cultivars at P=0.05.

DISCUSSION

During the last decade, coloured shade nets (photo-selective shade nets) designed specifically for manipulating plant development and growth have become available. These nets can provide physical protection (birds, hail, insects, excessive radiation), affect environmental modification (humidity, shade, temperature) (Perez *et al*., 2006) and increase the relative proportion of diffuse (scattered) light, while absorbing various spectral bands, thereby affecting light quality (Shahak *et al*., 2004). These effects can influence crops as well as the organisms associated with them. The effect of coloured shade net on plant development in crops, foliage crops, fruit trees and vegetables had been studied in the past years (Oren-Shamir etal., 2001; Nissim-Levi *et al*., 2008; Shahak *et al*., 2008). Recently, King *et al*. (2013) reported that red pepper quality, which was grown under Pearl shade net, had significantly prolonged storage and shelf life, compared with the same pepper grown under the black net. They concluded that the lower decay incidence under the Pearl net was due to the scattered light spectrum and the highratio between the Red/Far Red and Blue/UV under the Pearl net. The light fragment alterationreduced the inoculum level of *Alternaria* spp. in the field, inhibited fungal sporulation and inhibited fruit ripeningbased on skin color development, which has probably led to significant reduction in decay development after harvest, while maintaining the fruit quality (Goren *et al*., 2011). The goals of this study were to further investigate the mode-of-action of Pearl shade net in maintaining postharvest quality of two commercial red pepper varieties in relation to carotenoids content and antioxidant activity, which are involved, in maintaining the quality of fresh produce.

Bell pepper decay reduction could also be influenced by the fruit ripening process, as shown by colour development, and antioxidant contentsthat are affected by the light quality (Liu *et al*., 2004; Giliberto *et al*., 2005). Light quality and different wave lengthswere reported to affect fruit color and maturation in several different crops (Shahak *et al*., 2004; Lopez *et al*., 2007; Solomakhinand Blanme, 2010). In this study, fruits harvested under the Pearl had a lighter red color compared to fruitsgrown under the black shade net, which was associated with higher chlorophyll content.(Fig. 2). To the naked eye, fruit pigmentation between the two nets was nearly identical (Goren *et al.* 2011), however; chlorophyll values were higher under Pearl net, while carotenoid values were higher under black netting. The higher chlorophyll content in the fruit harvested under the Pearl compared to the black shade net is evidence of ripening inhibition, as reported in peach colour development under Pearl net (Shahak *et al*., 2004). Ripening inhibition can be associated with less fruit susceptibility to decay (Barkai-Golan, 2001). Vergasa was found to be less susceptible to decay in the field and decay development during storage and shelf life than Romans (Fig. 1).

Wang *et al*. (2008) reported that an increase in the antioxidant capacity and free radical scavenging activity reduced the physiological deterioration and enhanced the resistance of tissue against microbial invasion and reduced the spoilage of blueberries fruit. The antioxidant activity and carotenoids content in Vergasa fruit, under Pearl net, were significantly higher than the antioxidant activity and carotenoids content measured in Romans. Filtrated light has been shown to be the most important environmental factor that influences antioxidants as well as carotenoids biosynthesis in plants (Luthria *et al*., 2006; Pizarro andStange, 2009). Therefore, the higher antioxidant activity found in Vergasa under Pearl net, could be associated with low decay incidence, which was also reported in Erkan *et al*. (2008) for strawberries. These results could be strengthened by the strong correlation that was found between low decay development and high AOX, especially in Vergasa grown under Pearl net (Table 2). Such reaction mechanisms require further investigation.

CONCLUSIONS

Pearl shade net significantly inhibited fruit maturity and ripening, compared to the commercial black shade net. Significant differences in fruit quality were found between the two cultivars; Vergasa and Romans. Vergasa was found to have better quality traits than Romans. AOX activity and carotenoids content in Vergasa harvested under pearl net were higher in the fruit, while decay incidence was lower. Therefore, ripening and sporulation inhibition due to low ratio between FAR/FAR-red and high ratio between Blue/UV, together with higher AOX activity and carotenoids in the fruit could play an important role in decay reduction in pepper fruit after harvest. It is important to note that improved decay tolerance under Pearl net may be gene related, according to the variety. Further analysis of pepper varieties under coloured shade nets are required in order to develop optimal growing protocols for summer pepper crops.

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UTICAJ BOJENIH MREŽA SJENILA NA KVALITET PAPRIKE NAKON BERBE – MOGUĆI NAČIN RADA

SAŽETAK

U prethodnim studijama otkrili smo da su mreže sjenila biserne i žute boje značajno smanjile razvoj truleži paprike nakon berbe. Značajno niža pojava propadanja kod plodova ubranih ispod mreže sjenila biserne i žute boje objašnjena je niskim nivoom inokuluma Alternaria spp. na njivi i inhibicijom sporulacije gljiva, i/ili usporavanjem zrijenja plodova tokom rasta, što smanjuje podložnost ploda gljivičnim infekcijama na njivi zbog difuznog svjetla, njegovog kvaliteta i odnosa između spektra svjetlosti pod ove dvije mreže sjenila (Goren *et al*., 2011). Stoga je cilj ovog istraživanja bila da se dobije više informacija o uticaju mreže sjenila biserne boje na kvalitet plodova nakon berbe dva različita komercijalna kultivara crvene paprike (cv. Romans and Vergasa). Procjena kvaliteta ploda zasnovana je na sadržaju hlorofila, karotenoida i antikosidanasa nakon produženog skladištenja i simulacije plasmana na tržište. Nijesu zapažene značajne razlike u kaliranju ploda, čvrstini i sadržaju šećera ova dva kultivara pod različitim mrežama. Međutim, zapažene su neke razlike među kultivarima. Plodovi ova dva kultivara koji su rasli ispod mreže sjenila biserne boje su imali značajno nižu incidencu propadanja u odnosu na plodove ubrane pod komercijalnim crnim mrežama, naročito cv. Vergasa. Sadržaj hlorofila u plodu ispod mreže biserne boje bio je viši nego kod plodova uzgajanih pod crnom mrežom sjenila, a značajno viši kod Vergasa nego kod Romans. Sadržaj karotenoida je bio značajno viši ispod komercijalne crne mreže nego ispod mreže biserne boje, a značajno viši kod Vergasa nego kod Romans odmah nakon branja, skladištenja i simulacije roka trajanja. Odmah nakon branja i simulacije skladištenja i stavljanja na promet, aktivnost antioksidanasa (AOX) kod Vergasa bio je značajno viši nego kod Romans kod plodova ubranih ispod obje mreže sjenila. Značajno povećanje aktivnosti AOX je izmjereno kod plodova Vergasa ubranih ispod mreže biserne boje nakon simulacije skladištenja i stavljanja u promet u odnosu na crnu mrežu i svih ubranih plodova Romans. Zaključak je da bi uticaj mreže biserne boje na inhibiciju zrijenja, povećanu aktivnost AOX i sadržaj karotenoida mogao da smanji propadanje plodova paprike i održi bolji kvalitet plodova nakon branja.

Ključne riječi: *Capsicum annuum*, period nakon berbe, rok trajanja, skladištenje