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Cultivars Differences in Keeping Quality and Bioactive Constituents of Bell Pepper Fruit during Prolonged Storage

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ABSTRACT

The aims of this research were to evaluate the keeping quality (water loss, firmness, decay incidence) and bioactive constituents (vitamin C, antioxidant activity) of three commercial bell pepper cultivars (Selika-red, Dynamo-yellow and Sympathy-orange color) held for 21 days (d) storage at 7 ^{o}C and market simulation (3 d at 20 ^{o}C). Cultivars type significantly influenced fruit quality in bell pepper after prolonged storage. The low skin wax content (0.6 ng·cm^{-2}) and thin pericarp tissue (6.5 mm) in 'Sympathy' appeared to have contributed greatly to the high weight loss (5.2%), resulting in very soft fruits (4 mm), high decay incidence (20%) and lowest value in general appearance (2.5) among to the three cultivars. After 21 d storage at 7 ^{o}C plus 3 d at 20 ^{o}C content of ascorbic acid showed a insignificant increase in all cultivars. Hydrophilic antioxidant activity (HAA) remains practically unchanged after 3 weeks at 7 ^{o}C (between 2 and 5%) from the initial value. Total antioxidant activity (TAA) increases only in red pepper 'Selika' (6.07 µmol TE/g fr.wt.) and this increase is mainly because of changes in the lipophilic antioxidant activity (LAA - from 0.84 in harvest time increased to 2.31 µmol TE/g fr.wt. after storage).

Indexing Terms/Keywords

Pepper, Cultivars, Storage, Quality, Antioxidant .

Academic Discipline and Sub-Disciplines

Postharvest.

Subject Classification

Food quality.

Type (Method/Approach)

Quasi-Experimental.

INTRODUCTION

Sweet bell pepper (*Capsicum annuum* L.) is an important vegetable crop worldwide and can be consumed in many colors green (unripe), red, yellow, orange, or brown when ripe. Bell peppers are rich in both hydrophilic antioxidants, such as vitamin C, and lipophilic ones, such as carotenoids and vitamin E [1] with potential health-promoting properties [2]. Bell pepper have exceptionally high ascorbic acid (0.15 to 2.0 $mg \cdot g^{-1}$ fresh weight) compared to other fruit and vegetables. The major postharvest limiting factor in bell pepper is its relatively short shelf life of only one to two weeks which requires the use of air rather than sea transport for export of peppers to lucrative distant markets [3].

The storage life of pepper fruit is limited by physical decay and rapid senescence [4], water loss [5], and susceptibility to chilling injury [6; 7]. Pepper fruit cannot be stored at the low temperatures necessary to slow physiological activities (rate of respiration, ripening, etc.), because of chilling injury development. Extensive work has been done on various storage methods of bell peppers, ranging from CA (controlled atmosphere storage) [8], MAP (modified atmosphere packaging) [9], to the use of individual shrink packaging [10]. The most effective method of maintaining quality and controlling decay of peppers is by rapid cooling after harvest followed by storage at optimum temperature (7–10 ^oC) with a high relative humidity 95–98% [11]. Hot water rinsing while brushing (HWRB) bell peppers immediately after harvest at 55 ^oC for 12 s significantly reduced decay incidence, while maintaining fruit quality, compared with untreated fruit [12]. Yellow and orange pepper cultivars were found to be more susceptible to physiological and pathological deterioration than the red cultivars after prolonged storage [13]. These results are similar to the work of Ilić et al*.* [14] who found that yellow and orange mini pepper are in general more susceptible than red cultivars to physiological degradation, high decay incidence and the lowest value in external appearance (below marketable limit) after storage. Bell pepper quality attributes such as firmness, texture, decay incidence and general appearance (characterized by size, shape, uniform color, calyx freshness and absence of defects) are important criteria for determining market quality and consumer acceptance. The antioxidant activity (AA) of peppers depends on several factors including genetics, environmental conditions, production techniques used, stage of maturity, and postharvest storage conditions. Pigments in the red pepper, carotenoids and flavonoids, contribute significantly to its AA, while the green pepper lacks these pigments [1].

The aims of this research were to evaluate the keeping quality and antioxidans activity of different color bell pepper cultivars held for three weeks storage and shelf life period.

MATERIAL AND METHODS

Plant Material

Sweet bell pepper fruits (*Capsicum annuum L.*) 'Selika' red , 'Dynamo' yellow and 'Sympathy' orange color of uniform size (about 190±10 g each) and without defects or diseases were harvested from February in winter production season from plastic house in Arava (south Israel). For colored peppers (red, yellow and orange) an additional harvesting criterion is to have a minimum 80% coloration. Climate condition during the experimental season were average temperature (min 15 ^oC, max 27 ^oC), with 12 ^{hr} day length and average relative humidity (RH) 20%. The soil condition was well drained and sandy, and drip irrigation was used. Cultural practices, such as land preparation, planting and plant protection for the crop, were as is the standard in this area. The experimental design used was the randomized complete-block design. Data were collected from ten fruits in each block.

After harvest, fruits were placed on commercial equipment for cleaning and disinfecting. A unique and rapid method HWRB - Hot Water Rinsing and Brushing for simultaneously rinsing and disinfecting sweet peppers using hot water with 55 ± 1 ^oC for 12 ± 2 s was used [12].

Quality Traits

Quality parameters were evaluated immediately after harvest and at the end of a 21-day storage at 7 ^oC and a relative humidity (RH) of ~95%, plus 3 days at 20 ^oC and 70% RH (marketing simulation) as follows :

Loss of weight was assumed to be a loss of water, expressed as an average from the initial to the final weight of the fruit.

Fruit firmness was determined by compression [15] and linked as fruit deformation after application of a load of 2 kg to the equatorial region of the fruit. Lower deformation values indicated higher fruit firmness. At the end of storage, a hand firmness evaluation was also made, on a 5-grade scale (1-3: very firm to firm, 3-5: soft to very soft).

General appearance (GA) was evaluated visually considering fruit freshness, decay, softness, blemishes and other defects on a scale of 1-5 with : 1-poor, 3-good, 5-excellent.

Total soluble solids concentration (TSS %) in the juice was measured refractometrically (determined by an ATAGO, Brand Digital portable refractometer) on the 5 same fruits used for firmness.

Total *dry matter* was determined by drying procedure at the temperature of 105 °C up to the level of constant mass.

Wax content ($ng \cdot cm^{-2}$): six 1 cm diameter pericarp discs (ca.5-6g fresh wt./fruit) were excised from around the equator of each fruit from a random sample of 10 individual fruit of each cultivar on the day of harvest, frozen in liquid nitrogen, lyophilized, and stored at -80 ^oC prior to total wax extraction. Epidermal tissue of liophilized discs was peeled from pericarpdiscs with a sharp razor, scraped lightly to remove any residual pericarp tissue, and shaken in 5 ml hexane at room temperature for 30 min. The hexane extract was collected and filtered througth a Whatman GF/C glass microfiber filter. Remaining tissue residue was washed with additional hexane, filtered, and the extracts were combined. Hexane extracts were dried under nitrogen and the dry weights of the insoluble wax residue recorded.

Fruit was considered *dacayed* (D) once fungal mycelia apperead on the peel or calyx. Decay was expressed as a percentage of the total initial fruit number.

Ascorbic Acid (AA)

Ascorbic acid was analyzed with Tillman's method (PN-90 A -75101/11). The ascorbic acid quantification was carried out through the 2, 6-dichlorophenol indophenol method 967.21 (AOAC, 1990). 5 g from the sample along with 50 ml of oxalic acid (0.5%) were homogenized. A 5 ml aliquot was titrated with Tillman solution (0.02% of 2, 6-dichlorophenol indophenol Merk®), until a pink color was gotten. The concentration of the ascorbic acid was determined with the help of a pattern curve that was generated by titrating samples with known concentrations of ascorbic acid. The results are reported as mg of ascorbic acid per 100 g of pulp.

Antioxidants – Extraction And Determination

The antioxidant activity was measured using a modified version of the 2,2'-azinobis (3-ethylbenzothiazoline-6-sulphonate) (ABTS) discoloration method [16]. Hydrophilic and lipophilic fractions were isolated from fresh samples without preliminary drying by stepwise extraction with acetate buffer, acetone and hexane, and repeated partition of water-soluble and water-insoluble portions. Antioxidant activity was evaluated by decolorisation of the $ABTS^+$ radical cation. The radical was generated in acidified ethanol medium to allow the activities of both hydrophilic and lipophilic antioxidants. The final reaction mixture contained 150 µmol of ABTS and 1.77 mm of 2,2'-azobis (2- amidinopropane) dihydrochloride in acidified ethanol (249 mL ethanol 99.9% plus 250 µL H_2SO_4). Incubation of the reaction mixture at 45 ^oC for 60 min was sufficient for $ABTS^+$ generation. The obtained stock solution of $ABTS^+$ can be stored for at least 2 days at 4 ^oC without significant loss of its properties. The decolorization test was performed in plastic cuvettes by adding 10 µL of test sample to 1 mL of acidified ethanolic 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. Final results were calculated using the comparison between the absorbance of the samples and the absorbance of the standard (±)-6-Hydroxy-2,5,7,8- tetramethylchromane-2-carboxylic acid (Trolox). The antioxidant levels in the samples were determined as Trolox equivalents (TE) according to the formula :

$$TE\ (A_{sample} - A_{blank}) / (A_{standard} - A_{blank}) \times C_{standard},$$

where A is the absorbance at 734 mM and C is the concentration of Trolox (mM). To calculate the TE antioxidant capacity (TEAC) per weight of plant tissue, we used the formula:

$$TEAC\ (mmolTE/mg) = (TE \times V) / (1000 \times M),$$

where V is the final extract volume and M is the amount of tissue extracted.

Statistical Analysis

Each treatment consisted of four export cartons with 5 kg of fruit. All data were subjected to one- or two-way statistical analysis at P = 0.05 using JMP6 Statistical Analysis Software Program (SAS Institute Inc. Cary, NC, USA).

RESULTS

Total soluble solids and dry matter are known to increase fruit quality. No change in TSS was found after prolonged storage in all bell peppers cultivar (Table 1). Results of fruit quality evaluation showed that Selika (3.9%) and Dynamo (4.2%) fruits were significantly lower weight loss than Sympathy (5.2%) after storage and shelf life. It is possible than thin pericarp tissue (6.5 mm) and low epicuticular wax content (0.6 ng cm^{-2}) (Tab.1) increased weight loss (5.2%) and decreased fruit firmness (4.0 mm) (Tab.2) in 'Sympathy' pepper fruits after prolonged storage.

Table 1. Fruit composition of bell pepper cultivars

Cultivar	Dry matter %	TSS (%)	Pericarp thickness mm	Skin wax (ng cm^{-2})
Selika	11.4a	8.3 a$^{(v)}$	7.4± 0.08a	0.9± 0.04a
Dynamo	11.2a	8.4 a	7.2±0.06 a	0.7±0.03b
Sympathy	10.8b	8.1 a	6.5±0.05b	0.6±0.03b

TSS- percent of total soluble solids

A significant difference in decay incidence was observed in all three cultivars after shelf life. Orange pepper 'Sympathy' were found to be more susceptible to pathological deterioration (20.0%) that yellow 'Dynamo' (12.5%) and red 'Selika' (6.0%) cultivars after prolonged storage (Tab.2).

Table 2. Quality parameters of sweet pepper after 21 days at 7 °C + 3 days at 20 °C

Cultivar	Weight loss %	Firmness mm (*)	Decay incidence(**)	General appearance(***)
Selika	3.9±0.12b	2.2 c	6.0 c	3.4 a
Dynamo	4.2±0.08b	2.9 b	12.5 b	3.0 b
Sympathy	5.2±0.10a	4.0 a	20.0 a	2.5 c

(*) Fruit Firmness : very firm= 0-1.5mm residual deformation, firm 1,6-3mm, 3,1-4,5soft , more than 4,6-very soft

(**)Decay – Percentage of decayed fruit from total number of fruits.

(***) General appearance : 5-excellent; 3-good, 1-poor <3 –unmarketable

Values followed by the same letter are not significantly different at P=0,05 according to Duncan's multiple range test.

The highest content of ascorbic acid was recorded in cultivar 'Selika' (145 mg·100g^{-1}) followed by 'Sympathy' (136 mg·100g^{-1}) and the lowest in 'Dynamo' (78 mg·100g^{-1}). Storage at 7 °C for 21 d and shelf life simulation content of ascorbic acid showed a insignificant decrease in all cultivars (Table 3). However, stored red peppers showed a significant loss in vitamin C content, around 15%.

Table 3. Vitamin C content at harvest time and after 21 days storage at 7 °C + 3 days at 20 °C

Cultivar	Content of vitamin C (mg·100g^{-1})	
	Harvest time - To	After 21d 7 °C +3d 20 °C
Selika	147 ± 2.5 a	125 ± 2.6 a
Dynamo	78 ± 1.4 b	76 ± 1.6 b
Sympathy	136± 2.1 a	128± 2.3 a

TAA in red bell pepper 'Selika' immediately after harvest was 4.89 (0.84 lipophilic-LAA and 4.05 hydrophili-HAA) µmol TE/g fr.wt. Yellow pepper 'Dynamo' obtained 3.53 µmol TE/g fr.wt. while at same time, orange pepper fruit 'Sympathy' has 5.95 µmol TE/g fr. w. After 3 weeks storage at 7 °C and shelf life (3days at 20 °C) 'Selika' had the highest content of antioxidant activity (6.07) compared to 'Dynamo' (3.34) and 'Sympathy' (5.83 TEAC µmol TE/g fr.wt), Table 4.

Table 4. Antioxidant activity at harvest time and after 21 days storage at 7 °C + 3 days at 20 °C

	Antioxidant activity (AA) - µmol TE/g fresh weigh		
	Lipophylic - LAA	**Hydrophylic - HAA**	**Total - TAA**
Harvest time - T_o			
Selika	0.84±0.02a	4.05±0.06b	4.89b
Dynamo	0.20±0.01b	3.33±0.04c	3.53c
Sympathy	0.75±0.02a	5.20±0.05a	5.95a
After 3week storage at 7 °C + 3 days at 20 °C			
Selika	2.31±0.04a	3.76±0.05b	6.07a
Dynamo	0.20±0.02b	3.14±0.0b	3.34b
Sympathy	0.78±0.02b	5.05±0.07a	5.83a

DISCUSSION

Many vegetables become shriveled after losing only a small percentage of their original weight due to water loss. Fruit weight loss is a function of fruit surface area [5], pericarp thickness and content of epicuticular wax [3]. The thicker pericarp tissues (7.4 mm) and high skin wax (0.9 ng cm^{-2}) in 'Selika' fruit could serve as a good water reservoir and contribute to fruit firmness (2.2mm). Similar results on cultivar differences in weight loss based on their morphological characteristics also reported by [13; 17]. These results with maximum permissible weight loss in bell pepper agree with the 4-6% weight loss data described by Bustan and Lahav [18] or the 7% reported by Ben-Yehoshua et al. [15]. Genetic differences appeared to be responsible for some of the quality differences between the three cultivars. These results are similar to the work of Maalekuu et al. [13] who found that yellow and orange cultivars are in general more susceptible than red cultivars to physiological degradation after storage. Similarly, Ilić et al. [14] found the higher weight loss in 'Sweetbite orange' fruits resulted in very soft fruits, high decay incidence and the lowest value in external appearance (below marketable limit) among the three mini pepper cultivars after storage and shelf life period.

Because the bell pepper fruit is hollow, with a thin wall of approximately 5-8 mm thickness, it has a reduced capacity to store large volumes of water for long periods. The morphological advantages of 'Selika' over 'Dynamo' and 'Sympathy' in terms of pericarp thickness and epicuticular wax content played a significant role which affected fruit stability in several quality traits tested here. Thicker pericarp tissues and high skin wax in 'Selika' fruit could serve as a good water reservoir and would most probably contribute to fruit quality and marketing stability after prolonged storage and shelf life.

The low skin wax content and thin pericarp tissue in 'Sympathy' appeared to have contributed greatly to the high weight loss, resulting in very soft fruits, high decay incidence and lowest value in general appearance among to the three cultivars. Similarly, studies showed strong correlation between weight loss and fruit firmness and general fruit appearance [13]. In addition, relatively low decay incidence found in 'Selika' suggests that high skin wax content could impede decay as the first barrier against pathogen infection. The stem and calyx are more sensitive fruit parts in comparison to the fruit mesocarp. These morphological lesions may lead to *Alternaria*-induced rot on pods and calyxes, seed darkening and fruit shrinkage because of moisture loss. HWRB was reported to reduce decay incidence and chilling injury in fresh produce, although the fresh produce is exposed to the physical treatment for several seconds [12].

General appearance (GA) of fruits is of paramount importance in making purchasing decisions. It presents a summary of weight loss, firmness and decay, all of which are important quality parameters in fruits. Cultivars type significantly influenced general fruit appearance in bell pepper after storage and shelf life. All three cultivars immediately after harvest were very high in GA, with no decay. On the results, all three cultivars have been significant differences.

The ascorbic acid content has changed in response to the characteristic of varieties. Storage at 7 ^{o}C during 21 d decreased vitamin C content in red and orange pepper fruits, while content of vitamin C in yellow pepper did not change. Large differences in ascorbic acid content were observed among sweet pepper cultivars. Old cultivars and pepper populations from Serbia are rich in vitamin C (95.3 to 140.8 mg·100 g^{-1}) [19] but some new pepper selections from Turkey has content of vitamin C from 15.2 to 64.9 mg·100 g^{-1} in fresh sample [20]. These differences can be reasoned by genetic diversity. The ascorbic acid content was altered in response to the cultivars type, rippeness stage and storage conditions. Thus, the vitamin C content for green mature and breaker peppers stored at room temperature (20 ^{o}C) increased up to 10 days of storage, reaching similar values as those obtained for red peppers direct from the plant. Refrigeration at 4 ^{o}C for up to 20 days did not change the ascorbic acid content, except for red peppers, which showed losses around 15 % [21]. Temperature stresses before storage (hot water 55 ^{o}C for 12 sec) and low temperature during storage conditions (3 weeks on 7 ^{o}C) affect the pathways involved in the biosynthesis secondary metabolites, lead to higher phenolic metabolism and antioxidant capacity on pepper fruit. The relative capacity of antioxidants for scavenging ABTS$^{·+}$ radical was compared to the antioxidant potency of Trolox (water-soluble vitamin E) as standard.

Red pepper 'Selika' had the highest content of antioxidant activity among the 3 cultivars. Increasing TAA during storage could be related to ripening processes and metabolism of phenolic compounds [22]. Previous studies have shown that carotenoid content increases from 0.10 mg/g to 0.29 mg/g f.w. after 21 days of storage and shelf-life [1]. This has been suggested also by Jimenez et al. [23] that related to TAA, green fruits increased their antioxidant content during storage more than the red peppers, but red peppers had almost double the content than that of green fruits. Additionally, several authors point out that it is advisable to consume peppers at the full stage to achieve the maximum antioxidant capacity and potential health-beneficial effects [24]. Antioxidant activity was strongly correlated with the content of bioactive compounds from peppers [2].

CONCLUSIONS

Genetic specificity appeared to be responsible for some of the quality differences between the three bell pepper cultivars. Yellow and orange fruit are in general more susceptible than red cultivars to physiological and pathological deterioration after prolonged storage. Red 'Selika' with a relatively high amount of cuticular wax, remained firmer than 'Sympathy' and 'Dynamo', and had relatively low decay incidence. After 3 weeks storage at 7 ^{o}C and 3days at 20 ^{o}C (shelf life) 'Selika' also obtained the best internal quality traits with TAA of 6.07 µmol TE/g fresh weigh.

COMPETING INTERESTS

The author(s) declare that they have no competing interests.

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