

Effect of compost on antioxidant components and fruit quality of sweet pepper (*Capsicum annuum* L.)

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Abstract

In order to determine the effect of compost (CO) on antioxidant compounds and fruit quality of sweet pepper (*Capsicum annuum* L.), an experiment was conducted in open field. Treatments consisted of four levels of compost (0, 5, 10 and 15 ton ha⁻¹). The experiment was designed in randomized block design with three replications. Compost treatments positively affected fruit antioxidant compounds of pepper (antioxidant activity, total phenolic and carbohydrate content). But, no significant difference was found in total flavonoid content between compost and control treatments. The highest antioxidant activity and carbohydrate content were obtained in plants treated with 10 ton ha⁻¹ of compost. Fruit quality factors (pH, total soluble solids, titratable acidity, ascorbic acid and fruit firmness) were influenced by compost treatments. Total soluble solids, and fruit firmness significantly increased in response to compost treatments and the highest values were obtained from the most level of compost treatment (15 t ha⁻¹). Thus, these results showed that compost has strong impact on fruit quality and antioxidant compounds of pepper plants under field conditions.

Keywords: antioxidant activities, compost, fruit quality, pepper.

Introduction

The use of composts has been used to increase crop productivity and yields, and their use is usually associated with improved soil structure and enhanced soil fertility, increased soil microbial populations and activity and an improved moisture-holding capacity of the soil (Arancon, et al., 2004). Fruits and vegetables are a good source of natural antioxidants, containing many different antioxidant components that provide protection against free radicals and are associated with health-promoting properties (Velioglu, et al., 1998). Pepper is considered an excellent source of bioactive nutrients. Ascorbic acid (vitamin C), carotenoids and phenolic compounds are its main antioxidant constituents (Marin, et al., 2004). The levels of vitamin C, carotenoids and phenolic compounds in peppers and other vegetables depend on several factors, including cultivar, agricultural practice (organic or conventional), maturity and storage conditions (Lee and Kader, 2000). While plant growth media

and fertilizer regime may influence antioxidant concentrations (Aini, et al., 2009). Asami, et al. (2003) reported significantly higher total phenolics in arionberries grown with organic agricultural methods as compared with conventional method. Olsson, et al. (2006) demonstrated that strawberries grown organically had higher levels of all antioxidants including total phenolic, ellagic acid, and flavonols, than the conventionally grown strawberries. It was reported that, cabbages from organic management presents higher phenolics content than cabbages from the conventional management (Sousa, et al., 2005). Karakurt, et al. (2009) reported that humic acid application significantly influenced total carbohydrate content and total yield of pepper. Also, increasing nitrogen fertilisation has been found to decrease ascorbic acid concentration in several fruits and vegetables (Lee and Kader, 2000). Although positive influences of compost on plant growth and development have been well established for plants, their effects on fruit antioxidant activity and quality have not received much attention. Therefore, in this study we determine the influence of compost application on fruit antioxidant activities and quality of pepper under field conditions.

Martial and methods

Plant preparation: The investigation was conducted during the 2010 growing season at the experimental field of the Agricultural Faculty, Ferdowsi University of Mashhad, Iran (latitude 36° 17' N, longitude 59° 35' E and 985 m elevation). Soil sample (0-30 cm depth) was taken with auger after the site had been prepared for cultivation. The sample was analyzed for physical and chemical properties using standard laboratory procedures described by Mylavarapu and Kennelley (2002) and data shown in table 1. The experimental field was cleared, ploughed, harrowed and divided into plots. Sweet pepper seeds (*Capsicum annum* L. var. California Wonder) were established in a greenhouse in large trays with a 1:1 mixture of sand and peat (1:1 v/v). Irrigation was done after sowing when necessary. Seven-week-old pepper plants were hand-transplanted into well-prepared beds in the field. The plants spaced at 50 and 35 cm between rows and plants on row, respectively. All necessary cultural practices and plant protection measures were followed uniformly for all the plots during the entire period of experimentation.

Table1: Soil characteristics of experimental field

N (%)	P (ppm)	K (ppm)	Fe (ppm)	Cu (ppm)	Mn (ppm)	Zn (ppm)	pH
0.101	15.7	184	4.42	1.06	17.0	1.02	7.68

Experimental design and treatments

The experiment was arranged in a completely randomized block design (CRBD) with four treatments and three replications. Treatments consisted of four levels of compost (0, 5, 10 and 15 ton ha⁻¹). Composts were applied before transplanting the peppers and supplemented with amounts of inorganic fertilizer correspond with the recommended full rate of 120-150-100 kg NPK ha⁻¹ that was applied to the inorganic fertilizer-treated plots. Compost (as per treatment) and the inorganic fertilizers were applied and incorporated to the top 15 cm layer of soil in experimental beds.

Pepper fruits were harvested at the green mature stage. There were three plots per treatment, and three replicates per plot were collected. Each replicate comprised twenty peppers harvested from ten different randomly selected plants. Fruits were weighed and washed with distilled water. Part of the samples were immediately used for some analyses (titratable acidity, pH, total soluble solids, ascorbic acid and fruit firmness) and the other part was freeze-dried and ground for antioxidant analysis determination and stored at $-18\text{ }^{\circ}\text{C}$ before chemical analysis commenced.

pH, total soluble solids, titratable acidity and ascorbic acid: Pepper fruits from each treatment were cut into small slices and pooled. Samples were homogenized in a blender and portions of the homogenate were taken to determine the fruit quality. The pH value of fruit was measured with a pH meter at $20\text{ }^{\circ}\text{C}$. Titratable acidity (TA) was determined by titration with 0.1 N NaOH until pH 8.1 was reached and reported as g L^{-1} of citric acid fresh weight using citric acid as a control (Horwitz 1975). Total soluble solids content (TSS) was determined at $20\text{ }^{\circ}\text{C}$ with a refractometer and reported as $^{\circ}\text{Brix}$. Ascorbic acid contents (vitamin C) was measured by classical titration method using 2, 6-dichlorophenol indophenol solution and expressed as $\text{mg } 100\text{ g}^{-1}$ of fresh weight (Miller 1998).

Fruit firmness: Fruit firmness was determined for 5 fruits from each sample using a Chattillon hand penetrometer (Model DPP 1000) with a 0.6 mm probe. Measurements were performed at the center of each fruit. The maximum force (N) required to reach the bio yield point was recorded.

Antioxidant activities and total phenolic (extraction and analysis): Methanol extracts of freeze-dried fruits were prepared for the determination of antioxidant activity and total phenolic content. Weighed pepper fruit samples (5 g) were placed in a glass beaker and homogenised with 50 mL of methanol at $24\text{ }^{\circ}\text{C}$ overnight. The homogenate was filtered and then centrifuged at 6000 rpm for 15 min . Free radical scavenging activity of the samples was determined using the 2,2,-diphenyl-2-picrylhydrazyl (DPPH) method (Turkmen, et al., 2005). An aliquot of 2 ml of 0.15 mM DPPH radical in methanol was added to a test tube with 1 ml of the sample extract. The reaction mixture was vortex mixed for 30 s and left to stand at room temperature in the dark for 20 min . The absorbance was measured at 517 nm , using a spectrophotometer (Bio Quest, CE 2502, UK). The antioxidant activity was calculated using the following equation: $\text{Antioxidant activity (\%)} = 1 - A_{\text{Sample } (517\text{ nm})} / A_{\text{Control } (517\text{ nm})} \times 100$. The total phenolic content in methanol extracts was determined using Folin–Ciocalteu's reagent (Singleton and Rossi, 1965). Each methanol extract solution (0.5 mL) was mixed with 6 mL of distilled water and 0.5 mL of Folin–Ciocalteu's phenol reagent. After 5 min , 2 mL of 20 g L^{-1} sodium carbonate solution was added and the mixture was vortexed vigorously. The same procedure was also applied to standard solutions of gallic acid. After incubation at room temperature for 2 h , the absorbance of each mixture at 750 nm was measured using spectrophotometer. Results were expressed as mg of gallic acid equivalents (GAE) 100 g^{-1} on dry weight.

Total flavonoid content: The flavonoids content was determined spectrophotometrically using a method based on the formation of a flavonoid–aluminium complex (Yoo, et al., 2008). Each sample (2 g) was extracted with 10 mL methanol for 24 h . One millilitre of the extracts was added to a 10 ml volumetric flask. Distilled water was added to make a volume of 5 ml . At zero time, 0.3 ml of 5% (w/v) sodium

minute was added to the flask. After 5 min, 0.6 ml of 10% (w/v) $AlCl_3$ was added and then at 6 min 2 ml of 1 M NaOH were also added to the mixture, followed by the addition of 2.1 ml distilled water. Absorbance at 510nm was read immediately. Quercetin was chosen as a standard and the levels of total flavonoid content were determined in triplicate and expressed as quercetin equivalents in $mg\ 100\ g^{-1}$ on dry weight.

Carbohydrate content: Carbohydrate content was measured according to the method of Yemm and Willis (1954) using anthrone reagent. Sugars were extracted with 80% ethanol at 45 C, followed by centrifugation at 5000 rpm for 10 min. The reaction mixture consisted of 0.5 ml of extract and 5 ml of anthrone reagent which was boiled at 100 °C for 30 minutes. Absorbance was determined at 620nm. The carbohydrate content is expressed as $mg\ g^{-1}$ on dry weight.

Statistical analysis

Data were analyzed using SAS (SAS Institute, 2000) and means were compared by Duncan's multiple range test (DMRT) at 5% level of confidence.

Results and discussion

Effect of compost on fruit antioxidant compounds of pepper

Compost applications significantly affected fruit antioxidant activity (Table 2). Fruit antioxidant activity increased with increasing compost levels, and the highest fruit antioxidant activity was obtained from 10 t ha^{-1} of compost treatment (81.7%), while the control (without compost application) had the lowest fruit antioxidant activity (73.4%). Wang and Lin (2002) speculated that compost applications contributed to increased total antioxidant capacity in strawberries. A number of factors (light intensity, temperature and cultivar) can influence total antioxidant capacity of plant tissues and the type of soil and the content of humic compounds in soil can have a decisive effect: the higher the content of humic compounds in soil, the stronger antioxidant activity (Rimmer 2006); Therefore, plant growth media and organic fertilizer influence antioxidant concentrations (Ahn, et al., 2005). Another hypothesis explaining increases of antioxidant compounds in organic foods is that, since insecticide, fungicide, and herbicide use is limited in organic agriculture(similarly, in our experiment), plants devote greater resources to fight pathogen attacks, which, includes generation of antioxidant compounds (Winter and Davis, 2006). The result indicated that total phenolic content was affected by compost treatments (Table 2). The most total phenolic content was observed in the highest level of compost (15 t ha^{-1}) with 95.2 $mg\ 100g^{-1}$, while the lowest total phenolic content was recorded in control with 71.6 $mg\ 100g^{-1}$. These results are in agreement with those observed by Asami, et al. (2003) and Estiarte, et al. 1994. It has been reported that plants cannot simultaneously allocate resources to growth and defence and that there is competition between proteins and phenolics in plants for the common precursors involved in their biosynthesis (Riipi, et al., 2002). These results led us to presume that pepper plants may utilise benefits from compost fertilizer for their protein synthesis and growth development. On the other hand, organic acids and organic fertilizers (compost) act as precursors or activators of phytohormones and growth substances and secondary compounds in plants (Vernieri, et al., 2006). Moreover, But because there was sufficient light for normal photosynthetic rates in our experiment, the extra C could have been allocated for synthesis of C-based secondary compounds like phenolics in plants treated with the organic fertilisers

(Toor, et al., 2006). Compost treatments did not affect the total flavonoid content, however, treatment with 10 t ha⁻¹ of compost resulted in higher concentration of total flavonoid (125.4 mg 100 g⁻¹) than the other treatments (Table 2). Our results disagree with Mitchell, et al. (2007) found that organic crop management practices increased the content of flavonoids in tomatoes. Our results showed that compost affected on carbohydrate content (Table 2). The highest carbohydrate content resulted from 10 t ha⁻¹ of compost treatment application with 194 mg g⁻¹, while the lowest carbohydrate content was recorded in control with 170.4 mg g⁻¹. Our results agree with those of Copetta, et al. (2011), who determined that compost application at different concentrations improved carbohydrate content. Wang and Lin (2002) have reported that carbohydrate content and total soluble solids in strawberry fruits were positively correlated and sugar and organic acids are important for the sensory quality of fruits, i.e., fruits with low sugar and acid content taste flat.

Table 2: Effect of compost on fruit antioxidant compounds of pepper

Treatments (compost)	Fruit antioxidant activity (%)	Total phenolic content (mg 100 g ⁻¹)	Total flavonoid content (mg 100 g ⁻¹)	carbohydrate content (mg g ⁻¹)
CO1 (5 t ha ⁻¹)	74.9c	83.0b	91.3a	170.7b
CO2(10 t ha ⁻¹)	81.7a	80.4b	125.4a	194.0a
CO3 (15 t ha ⁻¹)	78.2b	95.2a	84.3a	170.9b
Control (0)	73.4c	71.6c	100.3a	173.5b

Within each column, same letter indicates no significant difference between treatments at 5% levels.

Effect of compost on fruit quality of pepper

Results presented in table 3 indicate that compost significantly affected titratable acidity (TA) of fruit. The maximum titratable acidity was produced at 10 t ha⁻¹ treatment with 10.5 g L⁻¹, while the minimum TA was in control with 7g L⁻¹. This result is the same trend with the findings of Santiago, et al. (2009) and Kashem and Warman (2009). It is likely that, in order to maintain the C:N ratio in the plants supplied with organic fertilizer, the extra C may have been used for the production of organic acids like citric acid and malic acid, which are responsible for the acidity of fruit [28]. Thus, our data confirmed previous workers that organic fertilizers increased levels of organic acids in fruits of pepper. The pH of fruit was significantly affected by compost treatments applied as shown in table 3. The highest fruit pH was in the lowest level of compost (5 t ha⁻¹) with 5.83, while the lowest fruit pH was in control with 5.18; however, no significant difference was found between treatments:10 and 15 t ha⁻¹ of compost. This result is the same trend with the findings of Giovanni, et al. (2011) and Toor, et al. (2006). The pH of fruit is correlated with acidity and acid content and citric acid is the primary organic acid found in most fruits (Wang and Lin, 2002). Fruits with low pH value (grown in organic fertilizers) indicate more citric acid, which is beneficial for human consumption (Wang and Lin, 2002). Additionally, fruit with low pH is more suitable for ripening while it also improves shelf life (Hernandez-Perez, et al., 2005). Compost application significantly increased total soluble solid (Table 3). The level of 15 t ha⁻¹ treatment produced the most total soluble solid (4.70 °Brix) and the least value related to the control (3.75 °Brix). Similar results were also

reported by Poor, et al. (2006) and Santiago, et al. (2009) observed that fruits harvested from plants that received compost had significantly greater total soluble solid (TSS) than those harvested from the mineral fertilizer plot. The improvement of fruit quality may be attributed to better growth of plant at different rate of organic fertilizer, which might have favoured the production of better quality fruit (Rajbir, et al., 2008). Table 3 shows the effect of compost treatments on vitamin C of fruit. Among compost treatments, the greatest vitamin C content was produced in the lowest level of compost (5 t ha⁻¹) with 145 mg 100 g⁻¹, while the least vitamin C was recorded in control with 96.8 mg 100 g⁻¹, but the difference between 5, 10 and 15 ton ha⁻¹ treatments was not statistically significant. Vitamin C levels in vegetables depend on several factors, including cultivar, plant nutrition, production practice and maturity (Antonio, et al., 2007). Our results agree with those of Taiwo, et al. (2007) who determined that compost application at different concentrations improved vitamin C of fruit. Organic fertilisation has been reported to give a low yield of tomatoes with a high ascorbic acid content, whereas mineral or mineral+ organic fertiliser gave a high yield of fruit with a lower ascorbic acid content (Dumas, et al., 2003). Therefore, our study confirmed previous results that the level of vitamin C in organically grown peppers was consistently higher than that in conventionally grown peppers. Fruit firmness was significantly greater in compost treatments than control (Table 3). The highest value of fruit firmness was obtained at 15 t ha⁻¹ treatment with 8.5 N, while the least fruit firmness was recorded in control with 5.8 N. This is in agreement with Riahi, et al. (2009) and Mccollum, et al. (2005) who reported that organic tomato fruit were generally firm. Firm fruits do not lose too much juice when sliced and are less susceptible to physical damage in shipping.

Table 3: Effect of compost on fruit quality characteristics of pepper

Treatments (compost)	pH	Titrateable acidity (g L ⁻¹)	Total soluble solid (°Brix)	vitamin C (mg 100 g ⁻¹)	Fruit firmness (N)
CO1 (5 t ha ⁻¹)	5.83a	7.0b	4.35ab	145.2a	8.5ab
CO2 (10t ha ⁻¹)	5.57ab	10.5a	4.25b	116ab	7.3bc
CO3 (15 t ha ⁻¹)	5.33bc	9.1ab	4.70a	121.ab	9.30a
Control (0)	5.18c	7.0b	3.75c	96.8b	5.8c

Within each column, same letter indicates no significant difference between treatments at 5% levels.

Conclusions

In conclusion, fruit antioxidant activities and quality of pepper affected by different compost levels. Although there were no significant differences between compost levels on almost all cases of variables, however, these were improved more with intermediate compost levels. Therefore, the usage compost could be suggested as easily bio-treatment for improvement in the fruit antioxidant activities and quality of pepper.

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