

## Research Article

# A Study on Organic Tomatoes: Effect of a Biostimulator on Phytochemical and Antioxidant Activities

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The objective of the present study is to investigate nutritional and antioxidant activity of four types of organic tomato cultivars. The differences in tomato quality are also tested between groups with or without treatment using an organic biostimulator, Stimplex. Total phenolic compounds (TPC), lycopene,  $\beta$ -carotene, DPPH free radical scavenging activity, reducing power, and color parameters were investigated in the current study. The results showed that there was no significant difference in TPC among cultivars regardless of Stimplex treatment. Higher lycopene and  $\beta$ -carotene were obtained in Stimplex treated tomatoes. Lycopene and  $\beta$ -carotene contents were significantly different among cultivars (P < 0.05). DPPH scavenging activity in controlled group was significantly higher than that in the Stimplex treated tomatoes (P < 0.05). No significant difference in reducing power was detected among cultivars treatment groups. The study showed that the darker the tomato color, the higher the lycopene and  $\beta$ -carotene contents and the stronger the reducing power.

## 1. Introduction

Tomato (*Lycopersicum esculentum*) is a widely grown and versatile vegetable throughout the world for taste, color, high nutritive value, and diversified use. Tomato fruit mainly contains fiber, phytonutrients, vitamin A, C, B complex, and carotenoids. Carotenoids include  $\beta$ -carotene and lycopene, where  $\beta$ -carotene is a pro vitamin and lycopene is a bright red carotene which has antioxidant properties two times higher than  $\beta$ -carotene in destruction of free radicals [1].

Color is the most important quality indicator of tomato fruit. Pigmentation of red ripe tomato fruit is a result of synthesis of carotenoids, lycopene, and  $\beta$ -carotene [2]. Lycopene in tomato is responsible for the redness, and  $\beta$ carotene can cause orange coloration. Intense red color tomato indicates predominant amounts of lycopene and high levels of antioxidants, which prevents cancerous and cardiovascular diseases [3].

Due to the restrictions on organic foods, not many fertilizers or growth promoters are available for organic tomatoes. Products of natural origin, such as seaweed extract, are already used in plant production. Many species of seaweed (*Ecklonia maxima, Kappaphycus alvarezii, Ascophyllum*  nodosum, Laminaria digitata, Laminaria hyperborea, Fucus vesiculosus, Durvillaea potatorum, and Fucus serratus) are used in agricultural and horticultural crops as a stimulator of the growth and development of plants. Application of these stimulators may contribute to enhanced growth, yield, and resistance against agricultural and horticultural plant pathogens, as well as the positive impact of content and activity of certain bioactive compounds [4]. Zarzecka et al. [5] also concluded that application of biostimulators, for example, seaweed extract Kelpak SL and Asahi in the study, can increase the total phenolic level in potato tubers, but the degree of the increase varied depending on the potato cultivars.

Stimplex is a seaweed extract (*Ascophyllum nodosum*) containing 0.01% cytokinin as an active ingredient, which stimulates plant growth and development, promotes yields and earlier maturity, improves resistance to environmental stress, improves fruit quality, and increases fruit set. A study showed that Stimplex treated drought stressed trees performed better than the untreated trees [6]. There is little available literature on the effect of biostimulants on the phytochemical contents and antioxidant activities in tomatoes.

Hence, the objectives of this study were to compare the tomato qualities grown organically and to observe the effect of biostimulant (Stimplex) on fruit quality attributes including fruit color, phytochemical level, and antioxidant activities in four tomato cultivars, namely, Black Cherry, Brandywine, German Johnson, and Roma. Correlation between fruit color, phytochemical level, and antioxidant activity was studied.

#### 2. Materials and Methods

2.1. Materials. In the current study, organic tomato cultivar experiment was conducted during spring and summer in 2015 at Tennessee State University certified organic farm in Nashville (latitude 127.30 m, 36°10′ N, 86°49′ W). The experiments were run using randomized block design (RBD) with three replications. Four tomato cultivars, Black Cherry, Brandywine, German Johnson, and Roma, were investigated under organic farming system. Organic seeds were obtained from an organic seed company (Johnny's Selected Seeds and High Mowing Organic Seeds company, Winslow, Maine, United States). Stimplex was procured from Acadian AgriTech (Alberta, Canada). Each cultivar was treated with Stimplex on a weekly basis at 2.5 ml/gallon via foliar spray.

All the chemicals and reagents for analysis were purchased from Sigma-Aldrich (St. Louis, MO, United States).

#### 2.2. Methods

2.2.1. Sample Preparation. After physical analysis of tomato fruit, fruits from each cultivar were selected randomly in each replication. Freshly harvested tomatoes were rinsed and dried for color measurement and then chopped and blended in waring blender to produce a homogenous mass for further analysis [7]. The homogenized samples were tightly sealed in sample vials and stored in a  $-20^{\circ}$ C freezer. Before analysis, the samples were thawed to room temperature and homogenized using a polytron (PT 2100, Kinematica AG, Schweiz) for 3 min before determination of total phenolic content, total lycopene content, total carotenoid content and antioxidant activities.

2.2.2. Moisture Content Determination. The fresh tomato samples were cut into small pieces  $(1 \text{ cm}^3)$ , weighed  $(100 \pm$ 5 g), and kept at 103°C to reach the consistent weights. Moisture content was calculated as follows:

Moisture content (%) = 
$$\frac{\text{wet weight} - \text{dry weight}}{\text{wet weight}}$$
 (1)  
× 100%.

2.2.3. Total Phenolic Content (TPC). TPC was determined by Folin-Ciocalteu method with some modifications [8]. Briefly 1g of tomato sample was added to 5 ml of 50% methanol and kept in a tightly capped bottle in the dark for 24 hours at 60°C with constant stirring. After being cooled to room temperature, the samples were centrifuged at 10,000 g for 5 min. Supernatant was used for the determination of total phenolic content. The supernatant (0.5 ml) was thoroughly mixed with 0.5 ml of distilled water and  $125 \,\mu$ l of Folin-Ciocalteu reagent for 6 min before the addition of 1.25 ml of 7% sodium carbonate. Final volume of solution was 3 mL using distilled water and was left in the darkness for 90 min. Absorbance was measured at 760 nm. Each test was repeated 3 times in triplicate each time.

TPC was calibrated by using a standard curve of gallic acid. TPC for each cultivar extract was expressed as gallic acid equivalent (GAE) in  $\mu g$  of GAE/g fresh tomatoes.

2.2.4. Total Lycopene and Total  $\beta$ -Carotene Estimation. Measurement of total lycopene and total carotenoids has followed the method described by Barros et al. [9], which is based on the method by Nagata and Yamashita [10] with modifications. The extraction of freshly harvested tomatoes was conducted using a mixture of solvents hexane: acetone: ethanol (2:1:1 v/v/v) with 2.5% butylated hydroxytoluene (BHT) added to each cultivar bottle [11]. Aliquots of 0.25 g homogenized samples were added to 25 ml extraction solvent. The bottles were tightly closed and agitated for 10 minutes on a shaker at 300 rpm, followed by leaving the bottles at room temperature to stand for 15 min for phase separation to form a distinct aqueous polar layer and a nonpolar layer. Aliquots of 4 ml extracts from the top layer of the nonpolar phase were withdrawn and filtered through Whitman's filter paper. Absorbance was measured spectrophotometrically at 453, 505, 645, and 663 nm [12]. Contents were calculated according to the following equations:

 $\beta$ -carotene (mg/100 mL)

$$= 0.216 \times A663 - 1.220 \times A645 - 0.304 \times A505$$
  
+ 0.452 × A453;  
vcopene (mg/100 mL) (2)

Ly

$$= -0.0458 \times A663 + 0.204 \times A645 - 0.304 \times A505$$
$$+ 0.452 \times A453.$$

The concentration of total lycopene and total  $\beta$ -carotene was further expressed in  $g kg^{-1}$  of fresh weight (fw).

#### 2.2.5. Antioxidant Activity

DPPH Free Radical Scavenging Property. DPPH free radical scavenging property was evaluated using the method described by Shahzad and others [13] with modifications. About 300 g of freshly harvested tomato was cut into pieces and homogenized using a polytron at medium speed for 5 min. Methanol (10 ml, 100%) was mixed with sample (10 g), and the mixture was centrifuged at 8000 g for 10 min. Supernatant (2 ml) was filtered through a 0.45 um Nylon Syringe filter for absorbance measurement at 517 nm.

DPPH free scavenging effect was calculated using the following equation:

DPPH scavenging effect (%)  
= 
$$\left[\frac{A_0 - (A - A_1)}{A_0} \times 100\right],$$
 (3)

Where  $A_0$  is the absorbance of the DPPH solution without sample, A is the absorbance of the test sample mixed with DPPH solution, and  $A_1$  is the absorbance of sample without DPPH solution.

*Reducing Power*. The reducing power assay was determined according to Oyaizu [14] with slight modification. Fresh tomato samples were homogenized using the polytron at medium speed for 5 min. Methanol (10 ml, 100%) was added to sample (10 g), mixed well, and followed by centrifugation at 8000 g for 10 min. Tomato supernatant (0.5 ml) was mixed with 0.5 ml of 0.1 M sodium phosphate buffer (pH 7.0) and 0.5 ml of 1% (w/v) potassium ferricyanide. The mixtures were incubated at 50°C for 20 min, followed by adding 1 ml of 10% (w/v) trichloroacetic acid, 1 ml of deionized water, and 0.1 ml of 0.1% ferric chloride sequentially. The absorbance was measured at 700 nm against a blank. A higher absorbance indicates a higher reducing power. Ascorbic acid was used for comparison.

Color Measurement. Color measurement was performed following the method described by Khairi and others [15] using a Minolta CR 300 Chroma Portable Colorimeter purchased from Hunterlab, Reston, Virginia, United States, with C illuminant expressed as  $L^*$ ,  $a^*$ , and  $b^*$ . The instrument was calibrated using the black and white tiles provided. Three measurements were performed with three replicates in each measurement. In addition, hue angle and total color difference ( $\Delta E$ ) were calculated using the following equation:

$$\Delta E = \sqrt{\left(\Delta L^{*2} + \Delta a^{*2} + \Delta b^{*2}\right)},\tag{4}$$

where the control values are from a reference tomato, Bing Cherry tomato.

2.3. Statistical Analysis. Statistical analysis was performed using Microsoft Excel and SAS Software. For each cultivar sample, three extracts were obtained and all the attributes were carried out in triplicate. Results were expressed as means  $\pm$  standard error. Analysis of Variance (ANOVA) test (5% Confidence Interval) was used to determine significant differences between the results. Correlation analysis was examined by using the CORR and TTEST protocol for paired *t*-test. In *t*-test, P < 0.05 was considered to be statistically significant.

## 3. Results and Discussion

3.1. Effect of Stimplex on Moisture Content. Moisture contents are presented in Figure 1. Control plants of Black Cherry, German Johnson, and Roma tomatoes had higher levels of moisture content than that of Stimplex-treated tomatoes. However, the control plant of Brandywine tomato showed a lower level of moisture content than that of Stimplex treated tomatoes. These differences were not significant (P < 0.05) for the Stimplex treated tomatoes and control in each cultivar. These results agreed with the study by Pinela et al. [16], who reported that the moisture content of different tomato cultivars ranged from 90.63 to 93.70 g/100 g fresh weights.

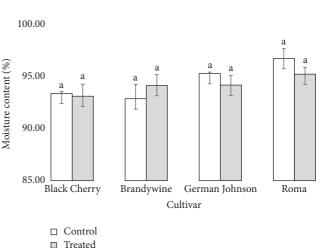


FIGURE 1: Moisture content of Stimplex treated and control tomato cultivars. *Note.* Values with different superscript letters were significantly different at P < 0.05.

Idah and others [17] treated Roma cultivar using chemicals and revealed that moisture was lower in control plants than that in chemically treated groups. Togun and Akanbi [18] used different types and doses of fertilizers on tomato plants and found that the dry matter accumulation increased in tomato fruits. However, the difference between control and treatment groups was not significant. They explained that the increased dry matter accumulation was due to the increased nutritional level for the plant growth. In the current study no significant increase of dry matter was detected although three out of the four Stimplex treated cultivars, Black Cherry, German Johnson, and Roma, showed increase in dry matter (Figure 1).

3.2. Total Phenolic Content (TPC). The results of TPC are presented in Table 1. Stimplex treated Black Cherry and Brandywine tomatoes have shown slightly higher TPC, and TPC in controlled cultivar German Johnson and Roma was higher than that in Stimplex treated tomatoes. No significance was detected among treatment groups and varieties (P > 0.05). The TPC obtained from the current study were comparable with the TPC values obtained from other research groups; for example, TPC was  $0.68 \,\mathrm{g \, kg^{-1}}$  fresh weight (FW) in Indian tomato varieties [19]. Figàs and others [20] analyzed TPC in 8 local tomato cultivars in Spain using a similar method and found that the TPC ranged from 0.35 to 1.37 g kg<sup>-1</sup>. Aldrich and others [21] reported a TPC range of 0.60–0.90 g kg<sup>-1</sup> FW in 10 tomato cultivars. A lower range in TPC was found from various research groups, for example, 0.25–0.31 g kg<sup>-1</sup> FW [14], 0.15–0.17 g kg<sup>-1</sup> FW [22], 0.07–0.22 g kg<sup>-1</sup> FW [7], and 0.14 g kg<sup>-1</sup> FW [23].

In the current study, TPC ranged from 0.82 to 0.95 g kg<sup>-1</sup> FW. TPC in Brandywine cultivar treated with Stimplex contained the highest TPC but the difference was not significant (P > 0.05). Therefore, the results indicated that Stimplex had no effect on TPC in tomato cultivars. Similarly, Riahi and Hdider [22] have used different organic fertilizers on

Treatment	Varieties	Total phenolic content (g kg <sup>-1</sup> FW)
	Black Cherry	$88.00 \pm 2.02E - 02^{a}$
Control	Brandywine	$87.71 \pm 4.66E - 02^{a}$
Control	German Johnson	$86.85 \pm 5.17E - 02^{a}$
	Roma	$86.06 \pm 3.04E - 02^{a}$
	Black Cherry	$90.50 \pm 3.06E - 02^{a}$
Stimplex treatment	Brandywine	$95.81 \pm 5.15E - 02^{a}$
Sumplex treatment	German Johnson	$82.48 \pm 5.03E - 02^{a}$
	Roma	$84.70 \pm 2.03E - 02^{a}$

TABLE 1: Total phenolic content in different tomato cultivars.

Note. Values with the same superscript letters were not significantly different at P < 0.05.

Treatment	Varieties	Lycopene (g kg <sup>-1</sup> FW)	$\beta$ -Carotene (g kg <sup>-1</sup> FW)	
	Black Cherry	$6.97 \pm 0.35E - 02^{b}$	$1.99 \pm 0.11E - 02^{a}$	
Control	Brandywine	$5.21 \pm 0.24E - 02^{c}$	$1.69 \pm 0.03E - 02^{\circ}$	
Control	German Johnson	$5.82 \pm 0.07E - 02^{\circ}$	$1.82 \pm 0.02 E - 02^{ m b}$	
	Roma	$6.78 \pm 0.04 E - 02^{b}$	$1.88 \pm 0.04 E - 02^{ m ab}$	
	Black Cherry	$7.32 \pm 0.22E - 02^{ab}$	$1.96 \pm 0.14E - 02^{a}$	
Stimplex treatment	Brandywine	$5.34 \pm 0.14E - 02^{\circ}$	$1.71 \pm 0.09E - 02^{ m bc}$	
Sumplex meannent	German Johnson	$6.83 \pm 0.16E - 02^{b}$	$1.90 \pm 0.09 E - 02^{a}$	
	Roma	$7.83 \pm 0.24E - 02^{a}$	$1.93 \pm 0.11E - 02^{a}$	

TABLE 2: Estimated lycopene and  $\beta$ -carotene concentrations in tomato cultivars.

*Note.* Values with different superscript letters were significantly different at P < 0.05.

two tomato cultivars and found that organic fertilizers had no significant effect on TPC in tomatoes. There was no significant difference among the tomato varieties, which agrees with Riahi and others [24]. However, Aldrich and others [21] reported that variety affects TPC in organic tomatoes.

Kocira and others [4] carried out a study investigating effect of a seaweed extract, Lelpak SL, on two common bean cultivars. Their results have shown that the biostimulator resulted in an increase in phenolic contents and the degree of increase varied based on the cultivar. Similarly, Fan and others [25] in their research have found that the commercial extracts of brown seaweed (*A. nodosum*) have enhanced the total phenolic and flavonoid content and antioxidant activity in spinach leaves. Also, Keitt mango trees treated with algae extracts reacted to the increased production of vitamin C in fruits [26].

3.3. Total Lycopene and  $\beta$ -Carotene Content. Absorbance of tomato extracts was obtained at different wavelengths for calculating total lycopene and total carotenoid concentrations. Results are presented in Table 2. Guinan and others [27] reported that biostimulants improve stress tolerance due to a greater production of antioxidants. The results from this current study concurred with Guinan and others [27] and showed that Stimplex treated tomatoes contained higher lycopene (antioxidant) content than that in the controlled group. Significant difference was detected in two cultivars, German Johnson and Roma, respectively (P < 0.05). Higher  $\beta$ -carotene contents were found in Stimplex treated tomatoes; however, only one cultivar, German Johnson, was detected with significant difference

(P < 0.05). The results from the current study showed that Stimplex application has affected lycopene and  $\beta$ -carotene contents. Among the cultivars treated with Stimplex, Roma cultivars exhibited the highest lycopene content (0.078 g  $kg^{-1}$ FW), followed by Black Cherry (0.073 g kg<sup>-1</sup> FW), German Johnson (0.068 g kg<sup>-1</sup> FW), and Brandywine (0.053 g kg<sup>-1</sup> FW), respectively. Moreover, Black Cherry contained higher carotenoids  $(0.020 \,\mathrm{g \, kg^{-1}} \,\mathrm{FW})$ , followed by Roma  $(0.019 \text{ g kg}^{-1} \text{ FW})$ , German Johnson  $(0.019 \text{ g kg}^{-1} \text{ FW})$ , and Brandywine (0.017  $g kg^{-1}$  FW), respectively (Table 2). Black Cherry recorded approximately the same amount of carotene for Stimplex treated and control fruits. The results showed agreement to the earlier studies conducted by Shahzad and others [13] that lycopene content in tomatoes ranged from 0.055 to 0.181 g kg<sup>-1</sup> FW. Pinela and others [16] analyzed tomato and  $\beta$ -carotene content using the same methods on four tomato cultivars and reported that the lycopene content ranged within 0.051–0.095 g kg<sup>-1</sup> FW and  $\beta$ -carotene content ranged within 0.003–0.004 g  $kg^{-1}$  FW. Figàs and others [20] studied eight tomato cultivars and found that the ranges for total lycopene and  $\beta$ -carotene were 0.011–0.098 g kg<sup>-1</sup> FW and  $0.004-0.022 \,\mathrm{g \, kg^{-1}}$  FW, respectively. The differences in lycopene and  $\beta$ -carotene were significant (P < 0.05) among some tomato varieties as shown in Table 2, which has agreed with the results from other groups [16, 20]. Pise and Sabale [28] studied the effect of three seaweed extracts (Ulva fasciata, Sargassum ilicifolium, and Gracilaria corticata) on the yield and quality of T. foenum-graecum L. and found significantly higher chlorophyll, carotenoid, and phenolic contents in plant sprayed with the tested extracts.

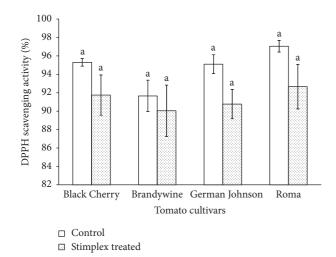


FIGURE 2: DPPH scavenging activity of different tomato cultivars. *Note.* Values with different superscript letters were significantly different at P < 0.05.

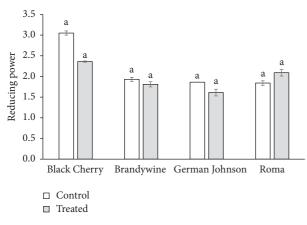


FIGURE 3: Reducing power of different tomato cultivars. *Note*. Values with different superscript letters were significantly different at P < 0.05.

3.4. Antioxidant Activities. Comparison of DPPH scavenging activities of tomatoes between Stimplex treated and control cultivars is presented in Figure 2. Stimplex treated tomato cultivars exhibited significantly lower DPPH scavenging activity than that of control (P < 0.05). The DPPH free radical scavenging ability of tomatoes also varied depending on the cultivars. Of the four tomato cultivars, Roma presented the highest DPPH free radical scavenging activity, followed by Black Cherry, German Johnson, and Brandywine. Liu and others [29] found a negative correlation between lycopene and DPPH scavenging activity. The results from the current study concurred with Liu and others' [29] statement by showing an increased lycopene level but a decreased DPPH scavenging activity in treated cultivars compared to the control.

The results of reducing power are presented in Figure 3. Three tomato cultivars in control group, namely, Black Cherry, Brandywine, and German Johnson, exhibited higher reducing power than that of the Stimplex treated tomatoes but the difference was not significant (P > 0.05), while

Stimplex treated Roma had a slightly higher reducing power than that of the control group but the difference was also not significant (P > 0.05). This result also indicated that there was no significant difference among tomato cultivars in reducing power (Figure 3).

Kocira and others [4] also investigated the effect of seaweed extract, Kelpak KL, on the antioxidant activities of two common bean cultivars. The methods they have used for antioxidant activity determination were antiradical activity (ABTS) and reducing power (RP). They have found that the antioxidant capacity of one cultivar was significantly improved by all the studied treatments, while, for another cultivar, the antioxidant activity varied based on the application treatments.

Many compounds contribute to the antioxidant activities; for example, Pinela and others [16] and Ochoa-Velasco and others [30] concluded that higher DPPH scavenging activities and reducing power are related to the higher antioxidant contents such as phenolic, flavones, anthocyanins, carotene, lycopene, vitamin C, and tocopherol. Kubola and Siriamornpun [31] reported that DPPH, FRAP, and hydroxyl radical scavenging ability had significant correlation with TPC. Shan and others [32] reported that, with the concentration increase of flavonoids, the antioxidant activity and free radical scavenging power were enhanced as well. However, Islam and others [33] reported that there was no significant difference in the antioxidant activities among species and preparing methods, even though the TPC was significantly influenced. Therefore TPC was not influencing the antioxidant activities. The result discrepancy between DPPH and RP methods may be due to the difference in principles of the methods. For DPPH method, the antioxidants transfer either electron or hydrogen atom to DPPH [31]. On the other hand, ferricvanide method was a ferric iron-based total antioxidant capacity assay. This assay was used to convert the potassium ferricyanide complex to potassium ferrocyanide complex, which would constitute ferrous complex by reducing ferric chloride [34]. The reducing potential was related to the ratio of compounds which donate hydrogen atoms to break the free radical chain [31]. Thus, it was the hydrogen on carboxyl groups of phenolic acid that helped reduce DPPH free radicals and potassium ferricyanide complex.

For the reducing power method, the hydrogen group ionized from carboxyl and hydroxy donated by the phytochemical compounds were oxidized by Fe<sup>3+</sup> processed by ferricyanide complex. Reducing power also varied according to different fractions based on molecular weight cutoff (MWCO). Fractions with smaller MWCO exhibited superior antioxidant activities compared to the other fractions [35].

In the current study, three phytochemical compounds were measured. The trend of antioxidant activities may involve other phytochemicals that are not investigated here.

3.5. Color. Color measurements of tomatoes were performed in triplicate with the  $L^*$ ,  $a^*$ , and  $b^*$  values received directly using a Minolta CR 300 Chroma Portable Colorimeter. These values were used to calculate  $\Delta E$  (color variation) and redness to yellowness ( $a^*/b^*$ ) values (Table 3).  $\Delta E$  represents actual difference in color and larger  $\Delta E$  values show higher redness

Treatment	Varieties	$\Delta E$	Redness to yellowness $(a^*/b^*)$	
	Black Cherry	$54.00 \pm 0.02^{d,x}$	$1.25 \pm 0.32$	
Control	Brandywine	$70.53 \pm 0.23^{a,x}$	$0.92 \pm 0.13$	
	German Johnson	$65.61 \pm 0.03^{b,x}$	$1.09 \pm 0.16$	
	Roma	$61.93 \pm 0.06^{c,y}$	$1.02 \pm 0.23$	
	Black Cherry	$52.74 \pm 0.03^{c,y}$	$0.86 \pm 0.14$	
Stimplex treatment	Brandywine	$67.82 \pm 0.30^{b,y}$	$0.93 \pm 0.21$	
stimplex treatment	German Johnson	$64.78 \pm 0.09^{b,y}$	$0.89 \pm 0.22$	
	Roma	$71.84 \pm 0.06^{a,x}$	$0.86 \pm 0.14$	

TABLE 3: Color change and redness to yellowness of tomatoes.

*Notes.* a, b, c, d as superscript letters show significant differences in the treatment (P < 0.05); x, y as superscript letters show significant differences between treatment groups (P < 0.05).

TABLE 4: Correlation coefficients of different nutritional parame	eters.
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Lyc	TPC	Caro	DPPH	RP	$L^*$	$a^*$	$b^*$	$\Delta E$	$a^{*}/b^{*}$
1.000									
$-0.405^{*}$	1.000								
$+0.845^{**}$	-0.399	1.000							
+0.154	-0.359	+0.257	1.000						
+0.340	+0.125	+0.518**	+0.276	1.000					
+0.379	-0.076	+0.305	+0.316	-0.039	1.000				
-0.239	-0.165	$-0.455^{*}$	+0.039	$-0.820^{**}$	+0.093	1.000			
-0.147	-0.156	$-0.413^{*}$	-0.176	-0.836**	-0.014	+0.953**	1.000		
-0.361	-0.106	$-0.530^{**}$	-0.200	$-0.662^{**}$	$-0.510^{**}$	$+0.800^{**}$	+0.853**	1.000	
-0.085	-0.017	+0.152	+0.633**	+0.576**	+0.149	-0.318	$-0.560^{**}$	$-0.428^{*}$	1.000

Symbol "+" indicates positive correlation. Symbol "-" indicates negative correlation. \*\* Significantly correlated at P < 0.01. \*Significantly correlated at P < 0.05; Lyc: lycopene concentration; TPC: total phenolic content; Caro:  $\beta$ -carotene content; DPPH: DPPH free radical scavenging ability; RP: reducing power;  $\Delta E$ : actual color change;  $L^*$ : lightness;  $a^*/b^*$ : reduces to yellowness.

values [36]. The pattern of color change ( $\Delta E$ ) was significantly different for Black Cherry compared with the other 3 cultivars for both treated and control groups (Table 3). Lower  $\Delta E$ for Black Cherry was obtained as a result of the typical blackish red (purple) color tomato. The Stimplex treatment had significant effect on two tomato cultivars in  $\Delta E$  (P <0.05), Black Cherry and Roma, respectively. The difference in  $\Delta E$  is also significant among cultivars (P < 0.05).

3.6. Correlation Analysis. Correlation coefficients among TPC, lycopene content,  $\beta$ -carotene, antioxidant activity, and color indexes are presented in Table 4. The study showed that lycopene content was positively correlated with DPPH and RP but the result is not significant. However, Liu and others [29] reported a negative correlation between lycopene and DPPH. No significant correlation was detected between  $\beta$ -carotene content and DPPH in the present study, which agrees with Liu and others [29] and Duan and others [37]. However,  $\beta$ -carotene content was significantly correlated with RP (P < 0.01). TPC and lycopene were not correlated with any of the color indices. Carotenoids were correlated with  $a^*$  (P < 0.05),  $b^*$  (P < 0.05), and  $\Delta E$  (P < 0.01). The index  $a^*/b^*$  was used to present linear relation in maturity stages of the tomatoes [38]; therefore no correlation with  $a^*/b^*$  was detected because all samples were collected when they were ripened. In the present study,  $\Delta E$  was not significantly correlated with lycopene and  $\beta$ -carotene content. The fruit lightness ( $L^*$ ), redness to yellowness ( $a^*/b^*$ ), and actual color change ( $\Delta E$ ) are not correlated to lycopene and total phenolic content in this study. This might be due to the complex nature of the tomato color which ranged from yellow and red to purple [39].

### 4. Conclusions

The results showed that there was no significant difference (P > 0.05) in TPC among cultivars regardless of Stimplex treatment. Higher lycopene and  $\beta$ -carotene were obtained in Stimplex treated tomatoes; however, only one cultivar, German Johnson, showed significant difference (P < 0.05). Lycopene and  $\beta$ -carotene contents were significantly different among cultivars (P < 0.05). DPPH scavenging activity in controlled group was significantly higher than that in the Stimplex treated tomatoes (P < 0.05). No significant difference in reducing power was detected among cultivars in control and treatment groups. The Black Cherry tomato, a purple colored cultivar, exhibited a much lower  $\Delta E$  value and a higher  $a^*/b^*$  value compared to the other red colored cultivars. Correlation study has revealed that  $\Delta E$  was significantly correlated to lycopene content,  $\beta$ -carotene, and reducing power (P < 0.01). Lycopene content was negatively correlated to DPPH (P < 0.05). The result indicated that the darker (from red to purple) the tomato color, the higher the lycopene and  $\beta$ -carotene contents and the stronger the reducing power.

## **Conflicts of Interest**

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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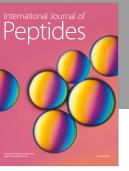
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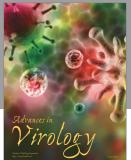
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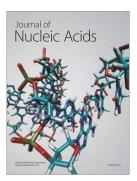


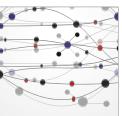
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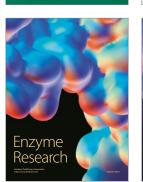
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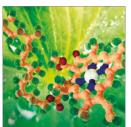
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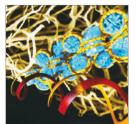


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