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The Effects of Planting Distances and Different Stages of Maturity on the Quality of Three Cultivars of Tomatoes (*Lycopersicon esculentum* Mill)

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Abstract

In this investigation the effects of cultivar, row spacing and different stages of maturity on qualitative characteristics of tomato (ascorbic acid, total soluble solids (TSS), β -carotene and lycopene) have been evaluated. Experiment was performed by factorial analysis with 3 replicates in completely randomized design (CRD). First treatment was three cultivars of tomato, second treatment was four planting distances and third treatment was different stages of maturity. The results showed that all treatments had significant influence on the levels of ascorbic acid, soluble solids, β -carotene and lycopene. Generally, wider spacing and deep red fruits had the highest quality. Therefore, choosing appropriate cultivars, special planting distances and suitable stage of maturity can increase fruit quality of tomato.

Keywords: ascorbic acid, carotenoid, lycopene, soluble solids, spacing

Introduction

Lycopen, a participant in the carotenoid family along with a pigment which attributes to the red color of tomatoes, is a leading factor to its health promoting ability (Tonucci et al., 1995; Fanasca et al., 2007). Many studies have shown that tomatoes can help decrease the chance of getting lung cancer, because lycopene acts as antioxidant (Young et al., 1993). Qualitative factors of tomato (flavor, color, soluble solids and nutrition value) were affected by cultivar, weather, condition storage, fruit maturity and cultivation methods (Gould, 1983). Changes in tomato fruit ripening occur within the plastids after the disappearance of chlorophyll. The two major groups of pigments found in tomato fruit are chlorophyll and carotenoids. The most noticeable change during ripening is the remarkable increase in the carotenoid content of the fruit (Laval-Martin et al., 1975). During ripening the chlorophyll concentration decreases while carotenoids, especially lycopene, accumulate in the fruit (Laval-Martin et al., 1975). According to Brandt et al. (2003) higher lycopene content was observed in tomato harvested in glasshouse-grown than in field-grown at different harvesting times. As suggested by Luh and Daoud (1968) the amount of lycopene in different cultivars reveals a considerable difference. The light intensity is influential in the biosynthesis of carotenoids and development of fruit color (Shiraghinge et al., 2010). Ascorbic acid, total soluble solids (TSS) and contents are commonly considered as fruit quality determining properties in tomato. Soluble solids include mainly the sugars such as glucose, fructose and sucrose. In tomato fruit, organic acids with sugars make a major contribution to the taste of the fruit. Most variation in flavor can be related to differences in the sugars and acids contents of the fruits. Bradly (1964) and Mauz (1966) showed that there is a fundamental difference in acidity between tomato cultivars. Although the cultivar has a dominant influence on the quality determinant properties, the environment in which it grows also has a significant impact on quality characters (Purseglove *et al.*, 1986). As shown by Malews-ki and Markakis (1971) ascorbic acid content decreases with ripening thus conflicting results could have been due to differences in ripeness at time of analysis.

Spacing is one of the important factors, which influence the quality of tomato fruit. Many experiments have been conducted on the effect of spacing on growth and quality of tomato. Tanaka and Komochi (1982) studied the relationship between plant density and topping on the growth and quality of tomato in greenhouse. They reported with increasing plant density, leaf size, stem diameter, weight of leaves and stem per plant decreased and flowering as well as ripening was delayed up to 7 days and yield per unit area increased. Srinivas and Hegde (1984) demonstrated that increased spacing increased dry matter production and quality attributes of tomato. A positive correlation was reported between plant density and yield and negative correlation between plant density and quality of fruits, so that the highest fruit quality was recorded in lowest plant density (Manchanda and Bhopal Singh, 1988). Gaye et al. (1992) reported that quality attributes of tomato declined with increasing plant density (Davies and Hobson, 1981; Baxter et al., 2005). Srinivasa et al. (1988) observed that

372

wider row spacing gave significantly more fruit quality than narrow spacing in tomato cultivars.

Optimum plant spacing may help in proper utilization of land and for obtaining good quality fruits. On the other hand, stem pruning also may influence on the production of yield. Therefore, the present study was undertaken to investigate the effect of optimum plant density and different stages of maturity for higher yield and better quality of tomato.

Material and methods

Plant materials and treatments

In this study three cultivars of tomato ('Super A', '1449', and 'Nemark 49') grown in greenhouses of (Ferdowsi University of Mashhad in Iran). Each tomato cultivar was planting in four distances (25 cm, 33 cm, 41 cm and 49 cm). These planting distances are common in this area The purpose of these intervals, was Investigate the effect of light and environmental conditions on fruit quality. Fruit sample from four maturity stages according Huerres Perez (1987) were gathered randomly by hand in three repetitions. Stages of maturity were classified as follows:

Mature green: fruits are mature and entirely light to dark green.

Pink: pink or red color ranges between 10 to 30%.

Red: red color is more than 60% but less than 90%.

Deep-red: red color exceeds 90% (Yamaguchi, 1983).

Traits measured were included soluble solid, ascorbic acid, β -carotene and lycopene at four different stages of fruit ripening.

Soluble solids analysis

Total soluble solid contents were determined by extracting and mixing one drop of juice from each fruit into a refractometer (ERMA, TOKYO).

Ascorbic acid analysis

Ascorbic acid was measured according to the method of AOAC (2005). In order to measure the ascorbic acid, 10 cm³ from tomato juice was mixed with 20cc distilled water and then 2 cc from (1%) one-percent soluble starch was added. Then ascorbic acid was determined by titration of 10 ml filtrated juice which contained potassium iodide (KI). In fact, it was based on mg ascorbic acid per100 g FW. Ascorbic acid content was calculated using the following formula (AOAC, 2005).

β - Carotene analysis

The β -carotene was analyzed by a modified AOAC method (AOAC, 1970) and was extracted with an acetone-hexane mixture. The MgO-Hyflo Supercel chromatographic column was washed with 0.5% acetone in hexane so that isomerization of carotenoids would be minimized (Wiseman *et al.*, 1952). β -carotene was eluted with 5% acetone in hexane. The concentration of β -carotene was based on the absorbance at 451 nm with an extinction coefficient of 2505 (Goodwin, 1955).

Lycopene analysis

The tomato samples (approximately 500 mg) were ground in a mortar in liquid nitrogen, followed by grinding in 4 ml of acetone with 50 mg CaCO₂. After centrifugation, pellets were re-extracted with 4 ml of acetone, 2 ml of hexane and 5 of ml acetone: hexane (4:1), successively. The supernatants were combined, measured and filtered through 0.2 µm nylon syringe filters into HPLC vials. All processes were performed as much as possible under subdued or safe light and a nitrogen atmosphere. For HPLC analysis a Spectra Physics SP8800 pump system was used equipped with a Spectra Physics Spectra 100 UV-Vis detector (Spectra Physics, Mountain View, CA, USA). For system processing and data acquisition a computer system with WINner on Windows software was used (Thermo Separation Products, Wirral, UK). Sample injections were carried out by means of a Waters 717 (Milford, MA, USA) autosampler equipped with a cooler. Separation of the pigments was carried out according to Gilmore and Yamamoto (1991) using a non-endcapped Allsphere ODS-1 HPLC column (4.6 mm × 250 mm, 5 µm particle size) preceded to a ODS-1guard column (Alltech Associates). Stainless steel column frit/insert material was replaced by a Peek Alloyed with Teflon (PAT) column frit/insert. The column temperature was 30°C. The mobile phase consisted of acetonitrile, methanol and tris buffer 0.1 M (pH 8.0). The mobile phase contained 0.1% (w/v) butylated hydroxytoluene. The flow rate was 1 ml min⁻¹, sample injection volume was 20 µl and spectrophotometric detection was performed at 445 nm. The concentrations of pigment standard stock solutions were determined spectrophotometrically using published absorbance coefficients (Konings and Roomans, 1997).

Statistical analysis

Effect of treatments were verified based on ANOVA using Excel program and the means were compared using LSD test at 5% level.

Results and discussion

Ascorbic acid

The experiment results demonstrated that there was a vital difference between three cultivars, planting distances and different stages of fruit ripening in ascorbic acid content. So the highest content of ascorbic acid was related to deep red fruits in 49 cm of planting distance (Tab. 2). As shown by Sing and Dillon (1986) the ascorbic acid and other acidity content of tomato juice was increased with maturity stages and reached the peak and thereafter started to decrease Singh and Dillon (1986). Also our results are similar to Mauz (1966) who reported that increasing light in to the plant leads to raised ascorbic acid. It can

be concluded that in 49 cm of row spacing, more light to the crown of the plant was penetrated, which results in increased fruit ascorbic acid. Mahendran and Bandara (2000) reported that moisture stress reduced the ascorbic acid content of tomato fruits when the stress was imposed during the fruit ripening stage. It can be concluded that close spacing retains soil moisture. Also they stated a reduction in the D-glucose synthesis would have occurred during the period of stress, which in turn may have reduced the synthesis of ascorbic acid. Moisture stress may have reduced the substrate concentration for ascorbic acid synthesis. Reduction in the substrate may possibly be due to reduced photosynthetic rate (Mahendran and Bandara 2000).

Total soluble solids (TSS)

The study results revealed that there was a significant difference between three cultivars, row spacing and different stages of fruit ripening in soluble solids. The most soluble solids were obtained from red fruits of 'Super A' cultivar in the 49 cm of plant-spacing. Fernando and Alisdair (2006) suggested more space of planting distances increased photosynthesis which results in increased soluble solids. It should be noted that there are a lot of light penetrated in wider spacing in comparison with close spacing. Purseglove *et al.* (1986) reported that fruit ripening had a major impact on quality attribute and raised the dry matter of fruit and soluble solids. These results are in close agreements with our findings (Tab. 1).

β – Carotene

The results of this experiment showed that cultivar, row spacing and different stages of fruit ripening had central effects on carotenoied level, so that the highest β -carotene content belonged to 'Nemark 49' cultivar in 49 cm of planting distance of pink fruits (Tab. 3).

Young *et al.* (1993) mentioned that there is a prominent difference between cultivars. They also expressed that presence amount of carotenoid in some cultivars and their absence in the other varieties is due to differences in their genes controlling biosynthesis. In addition, they reported that light density is a major factor to improve carotenoid Chlorophyll content. Bradly (1964) stated that fruits receiving more light have higher levels of carotenoid. So, it can be concluded that increasing row spacing, revealed a better penetration of light to the crown of the plant that leads to β - carotene content increasing.

Lycopene

The results demonstrated that cultivar, row spacing and different stages of ripening fruit had an essential difference on lycopene level. The results of our investigation established that the content of lycopene in all the investigated tomatoes during fruit ripening significantly increased. The lowest concentration of lycopene (32 mg 100 g⁻¹) was recorded in the green fruits of the cultivar '1449', in 25 cm of row spacing and the highest concentration of lycopene (434 mg 100 g⁻¹) was measured in completely ripened tomatoes of the cultivar 'Nemark 49' in 41 cm of row spacing (Tab. 4). Muhammad and Singh (2007) reported that

Tab. 1. Comparison of interaction between variety, row spacing and different stages of maturity for TSS

Different stages			gre	een			pi	nk			re	ed		deep red				
Planting distances		25	33	41	49	25	33	41	49	25	33	41	49	25	33	41	49	
Cultivar	'Super A'	2.2 ¹	2.3 ^k	2.7 ^j	2.8 ⁱ	3.0 ^h	3.1 ^g	3.1 ^g	$3.2^{\rm f}$	3.45°	3.46°	3.68 ^d	3.7 ^d	4^{b}	3.9°	4.38ª	4. 9ª	
	ʻ1449'	2.2 ^k	2.65 ^j	2.65 ^j	2.7 ^j	3.1 ⁱ	$3.35^{\rm h}$	3.5 ^g	3.46 ^g	3.68^{f}	3.7 ^f	3.98°	4 ^c	4.3 ^d	4.5°	4.7 ^b	4.4ª	
	'Nemark 49'	2.6 ⁱ	2.3 ^k	2.4 ^j	2.6 ⁱ	2.76 ^h	2.8 ^h	2.98 ^g	3.0 ^g	$3.13^{\rm f}$	$3.2^{\rm f}$	3.36°	3.4°	3.6 ^d	3.7°	3.8 ^b	4.4ª	

Means with similar letters in each table are not significantly different by LSD multiple range test (p < 0.05)

Tab. 2. Comparison of interaction between variety, row spacing and different stages of maturity for ascorbic acid	Tab. 2. Comparison	of interaction betwee	n variety, row spa	acing and different s	stages of maturity	for ascorbic acid
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Different stages			gre	een			pii	nk			re	ed		deep red				
Planting distances		25	33	41	49	25	33	41	49	25	33	41	49	25	33	41	49	
Cultivar	'Super A'	11.86 ⁱ	12.70^{h}	14.46 ^g	14.20 ^g	17.03^{f}	17.26 ^f	20.9 ^{ds}	20.3^{de}	23.8 ^b	23.2ª	27.46ª	27.10ª	20.10 ^c	20.86 ^d	22.33°	22.40°	
	ʻ1449'	11.73 ^j	12.40 ⁱ	14.66 ^h	14.06 ^h	16.30g	17.63 ^f	20.3 ^{de}	19.9 ^d	23.1 ^b	23.5ª	27.36ª	27.16ª	20.30^{de}	20.60^{d}	22.03 ^c	21.76°	
	'Nemark 49'	11.56 ⁱ	12.13 ⁱ	14.63 ^h	14.30^{h}	16.53 ^g	17.23 ^f	20.6 ^d	21.0°	23.9 ^b	23.5ª	26.96ª	26.53ª	19.96°	20.43 ^{de}	21.66 ^c	21.06 ^{cd}	
	I Willark 47	11.50	12.15	14.05	11.50	10.55	17.25	20.0	21.0	23.7		20.70	20.95	17.70	20.15	21.00	21.0	

Means with similar letters in each column are not significantly different by LSD multiple range test (p< 0.05)

Tab. 3. Comparison of interaction between variety, row spacing and different stages of maturity for B-carotene

Differ		gre	een			р	ink			re	ed		deep red				
Planting distances		25	33	41	49	25	33	41	49	25	33	41	49	25	33	41	49
	'Super A'	$51.0^{\rm f}$	51.4^{f}	57.4°	49.4^{f}	258 ^d	263°	288ª	271.4^{b}	24^{g}	21.4^{h}	7.4 ^j	11^{i}	0.50 ^k	0.76 ^k	0.50 ^k	0.16 ^k
Cultivar	'1449'	63.7 ^g	$66.7^{\rm f}$	69.4°	64.4 ^g	228 ^d	238.7°	251.7 ^b	264.7ª	35.4 ^g	27^{h}	19.4 ⁱ	17^{j}	8.26 ^k	9.7 ^k	5.3 ¹	7.4^{k}
	'Nemark 49'	36.4^{f}	36 ^f	35.7 ^f	37.4^{f}	313 ^d	324°	342.4 ^b	351ª	58°	39°	$31.7^{\rm h}$	23.7 ⁱ	11.4 ^j	12.5 ^j	7.6 ^k	6.8 ^k

Means with similar letters in each column are not significantly different by LSD multiple range test (p < 0.05)

	1						1	0		0			1	1			
Different stages		green					pi	nk			re	ed		develop red			
Planting distances		25	33	41	49	25	33	41	49	25	33	41	49	25	33	41	49
	'Super A'	50 ^k	55 ⁱ	60 ⁱ	68 ^h	217 ^g	$223^{\rm f}$	226°	235 ^d	37°	371.7°	374.7 ^b	377 ^b	411.7ª	411ª	411.4ª	409.7ª
Cultivar	ʻ1449'	32°	37.4 ⁿ	42 ^m	51.4 ¹	165 ^k	172 ^g	183 ⁱ	196.7 ^h	301.7 ^g	307.4^{f}	311.4°	316.4 ^d	361 ^b	368.7ª	357.4°	370.4ª
	'Nemark 49'	61.4 ^m	73.4^{l}	83 ^k	98 ^j	23 ⁱ	246.4 ^h	260.4^{g}	277 ^f	381°	389 ^d	386.4 ^d	386.4°	420.4 ^b	431.4ª	434.4ª	418 ^b

374 Tab. 4. Comparison of interaction between variety, row spacing and different stages of maturity for lycopene

Means with similar letters in each column are not significantly different by LSD multiple range test (p < 0.05)

the amount of lycopene is influenced by the cultivar. Anderson (1969), Uddin *et al.* (1997) and Muhammad and Singh (2007) reported that plant spacing had a large affect on the PAR (photo synthetically active radiation) so the ratios of blue, far-red, red far and blue red as a result of decreased plant density were increased. This issue has an impact major on lycopen level. Also, Law and Eghrevba (2009) showed that tomato fruit exposed to sunlight during ripening contained more lycopene than those ripened in shade.

Conclusions

Several factors including: cultivar, row spacing and different stages of maturity can influence ascorbic acid, soluble solids, β -carotene and lycopene in tomato fruits. According to this study, sunlight is a crucial factor on fruit quality. In general, increasing row spacing causes more light enter to the plant, so it increases ascorbic acid, lycopen and carotenoids content. In high row spacing, more photosynthesis take place because there is more light penetration in to the canopy and thus increases the amount of total soluble solids.

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