Journal of Food and Agriculture Research
Vol. 1, No. 1, 2021, pp. 15-29

URL: www.arfjournals.com

Variation of Bioactive Compounds and Antioxidant Activity during Ripening of Tomato Cultivars

RADHA KUSHWAHA[1], MONIKA SINGH[1], VINTI SINGH[1], VINITA PURANIK[1] AND DEVINDER KAUR[1*]

[1]*Centre of Food Technology, University of Allahabad, Prayagraj, India*
[*]*Corresponding author E-mail: devi_sonu@yahoo.com*

Abstract: The lipophilic and hydrophilic (LAA and HAA) antioxidant activities, lycopene, β-carotene and color values of four tomato cultivars (Rupali, Himsona, NDT, 1897) at different ripening stages (green, breaker, turner and ripe) were determined and their correlation with ascorbic acid, total phenolic, flavonoid, lycopene and β-carotene content were studied. The stages of ripening significantly influenced the β-carotene, lycopene and antioxidant activity. Beta carotene content increased until breaker and turner stages, decreasing afterwards. In contrast amount of lycopene accumulated through the ripening stages. The LAA followed the same pattern as lycopene and has good correlation with FRAP assay. The HAA of studied tomato cultivars were also significantly influenced by ripening stages, at the red ripe stage cultivar Rupali showed the highest amount of lycopene whereas cultivar Himsona showed highest total phenol, flavonoid, reducing power and ascorbic acid content.

Keywords: Lycopene; β-carotene; Antioxidant; Tomato.

Received : 5 January 2021
Revised : 9 January 2021
Accepted : 2 February 2021
Published : 30 June 2021

TO CITE THIS ARTICLE:

Radha Kushwaha, Monika Singh, Vinti Singh, Vinita Puranik & Devinder Kaur. 2021. Variation of Bioactive Compounds and Antioxidant Activity during Ripening of Tomato Cultivars *Journal of Food and Agriculture Research,* 1: 1, pp. 15-29

1. Introduction

Tomato (*Lycopersicon esculentum*) is one of the most important worldwide agricultural crops, rich in plethora of natural antioxidant compounds. The ripening of tomato fruits involves various morphological, physiological, biochemical and molecular changes including chlorophyll degradation, synthesis and storage of carotenoids, particularly lycopene. Lycopene is a powerful natural antioxidant. It exhibits the highest physical quenching rate constant with singlet oxygen among dietary carotenoids. The amount of important antioxidants, such as ascorbic acid and phenolics, is also variable

during tomato ripening, thus varying the nutritional value and the health properties of the fruit (Kaur *et al*. 2006).

The qualitative-quantitative analysis of different antioxidants, as well as their variation during ripening, is of great relevance both to human health and to commercial purposes. Along with carotenoids, other antioxidant compounds of tomato fruits, such as ascorbic acid and phenolics, play a determinant role in disease prevention (Robards *et al*. 1999; Karakaya *et al*. 2001). It has been reported that the levels of the health promoting bioactive compounds and of the antioxidant activity of tomato extracts are strongly influenced by several cultural practices and agronomic aspects, particularly the varietal genotype and ripening stage of the fruit (Cano *et al*. 2003; Dumas *et al*. 2003; George *et al*. 2004; Garcia and Barrett, 2006). Moreover, it is widely recognized that the protective role of tomato consumption is due to the synergistic effect among the different classes of antioxidants (Pastori *et al*. 1998; Amir *et al*. 1999). Recently, the attention has been given to antioxidant compounds and antioxidant activities in fruits and vegetables. In fact, their estimation is becoming an important evaluation parameter for the nutritional quality of food and its quantification allows a real evaluation of this nutritional value rather than the analysis of each single antioxidant compound (Lenucci *et al*. 2006; Pellegrini *et al*. 2007).

In this study changes in major antioxidant compounds and antioxidant activities of four tomato cultivars (cvs) harvested at four different stages of ripening were investigated. The hydrophilic and lipophilic antioxidant activities (HAA and LAA, respectively) and their correlation to different classes of antioxidants were also examined.

2. Materials and Methods

Four tomato cultivars Himsona, Rupali, NDT and 1897 were selected for the current study and it was procured from Krishi Vigyan Kendra, Karachchna Naini, Allahabad. All cultivars under investigation were subjected to identical cultural and agronomic practices as described by Eldridge *et al*. (2016) and of course, environmental conditions in order to minimize the influence of pre- and post-harvest factors.

2.1. Lycopene and β-carotene Determination

Carotenoids was extracted after homogenisation with pestle and mortar from tomato (0.3 g). The homogenate was mixed and extracted with hexane:methanol:acetone (2:1:1) containing 2.5% butylated hydroxyl toluene (BHT), the solution was allowed to separate the polar and non-polar layers.

Lycopene and β-carotene was analyzed using HPLC system (Agilent 1200 Infinity series). The separation was done by C-18 column (ZOBOX eclipse plus, particle size 4.6 μm, 150 mm, 2.0 mm). The mobile phase comprised of isocratic mixture of 80% methanol in water at flow rate of 1 mL/min, 35°C. The concentration of standard solution was calculated using the molar extinction coefficient of 17.2 X 10^4 for lycopene and 13.9 X 10^4 for β-carotene. Peaks were analyzed at 472 nm and 453 nm for lycopene and β-carotene, respectively. The results were reported as mg/kg, wet weight (wb) (Kotikova *et al*. 2011).

2.2. Flavonoid Determination

The flavonoid content was determined as described by Zhishen *et al*. (1999) on triplicate aliquots of the homogenous suspension (0.3 g). The absorbance was taken at 510 nm using a UV-Vis spectrophotometer. The linear reading of the standard curve was from 0 to 250 mg quercetin per mL and flavonoid content was expressed as mg of quercetin equivalent per kg of wb (mg QE/kg wb).

2.3. Phenolic Content Determination

In phenolic estimation, total phenols were extracted from tomato fractions and measured quantitatively. Five hundred mg of tomato samples were extracted using 5 mL of methanol and diluted 1:5 (v/v) with distilled water. The extract was diluted to 125 μL and mixed with 0.5 mL of distilled water, followed by the addition of 125 μL of Folin-Ciocalteu reagent and allowed to stand for 6 min. Then 1.25 mL of 7.5% sodium carbonate solution was added and the final volume was made up to 3 mL with distilled water. The samples were then allowed to stand for 90 min at room temperature and the absorbance was measured at 765 nm. The linear reading of the standard curve was from 0 to 500 mg of gallic acid/ mL (Singh *et al*. 2016).

2.4. Antioxidant Activity Determination

The measurement of HAA and LAA was performed using the ferric reducing antioxidant power (FRAP assay). Different solvents for the preparation of the hydrophilic and lipophilic fractions, (methanol and hexane) determinations according to Benzie and Strain, (1996) to FRAP assay.

2.4.1. Ferric Reducing Antioxidant Power (FRAP) Assay

Hydrophilic and lipophilic antioxidants were extracted from 0.3 g of homogenate (three replicates) with absolute ethanol or hexane at 4°C under constant shaking (300 rpm) overnight. Samples were centrifuged at 10,000

RPM. The supernatants were used for antioxidant activity measurement. 50 mL of hydrophilic and lipophilic tomato extract was added to 1.5 mL of FRAP reagent 1 mM 2,4,6-tripyridyl-2-triazine (TPTZ) and 20mM ferric chloride in 0.25M sodium acetate buffer, pH 3.6 and mixed thoroughly. After 4 min at 4°C, absorbance at 593 nm was read against a blank of water. A calibration curve was prepared using freshly prepared ammonium ferrous sulphate. The linear reading of the standard curve was from 0 to 1200 μM FRAP. Values were obtained from three replicates as mM FRAP/g of tomato wb (mM FRAP/g wb).

2.5. Color Value

Color space and values L* (degree of lightness), a* (degree of green a*<0, red a*>0), b* (degree of blue b*<0, yellow a*>0) was determined using Hunter Color lab (Hunter Associates Laboratory, Reston, VA, USA, 45°/0° geometry, 10° observer). For each sample at least three measurements were performed at different positions. Hue angle was calculated from arctan (b*/a*) and chroma by using formula $(a^{*2} + b^{*2})_{1/2}$.

3. Statistical Analysis

The experimental design was a randomised complete block with four factors (cvs) and results were estimated in triplicates. The data were subjected to one-way analysis of variance using SPSS statistical software version 20.0. Duncan test was applied to establish significant differences between means with a confidence level of 95%. Correlations were estimated using Pearson's correlation coefficient (R).

4. Results and Discussion

4.1. Lycopene and β-carotene

The amount of lycopene and β-carotene in tomato cultivars, harvested at four successive stages of fruit development and ripening corresponding to the unripe green, breaker, turner and red ripe colour of the fruit are reported in table 1. For all the investigated tomato cultivars, the amount of lycopene and β-carotene, expressed on wet weight (wb) basis, markedly increased during fruit ripening. The highest rate of synthesis and accumulation of lycopene in chromoplasts was detected in the transitional phase between the breaker and ripe stage. This may possibly be due to a progressive activation of the molecular mechanisms involved in carotenogenesis regulation during the transition between the breaker and ripe ripening stages, followed by a feedback

Table 1: Lycopene, β-carotene, total phenols, flavonoids, and ascorbic acid contents in tomato cultivars harvested at four different ripening stages

Cultivars	*Lycopene (mg/kg wb)*	*β-carotene (mg/kg wb)*	*Phenolic content (mg GAE /kg wb)*	*Flavonoids (mg QE/kg wb)*	*Ascorbic acid (mg/kg wb)*
Himsona					
Green	ND	1.22±0.09^{ab}	292.63±12.32^{def}	158.63±10.98^{gh}	189.58±14.27^{cd}
Breaker	6.44±1.18^{a}	2.01±0.19^{c}	342.23±12.73^{g}	171.11±14.92^{h}	302.62±14.20^{i}
Turner	27.28±2.50^{c}	2.76±0.29^{d}	314.82±16.45^{e}	125.53±10.66^{bcde}	252.68±10.26^{ghi}
Ripe	34.40±3.08^{d}	3.02±0.24^{de}	281.32±10.12^{cde}	147.85±8.34^{efg}	231.21±15.31^{efg}
1897					
Green	ND	0.92±0.10^{a}	232.39±12.77^{a}	132.87±11.76^{cde}	145.94±16.63^{a}
Breaker	6.09±1.22^{a}	2.04±0.30^{c}	294.07±11.13^{ef}	154.55±16.56^{fgh}	247.28±18.28^{efgh}
Turner	22.70±2.47^{b}	2.85±0.25^{d}	270.80±11.28^{bcde}	118.38±14.54^{abc}	230.29±20.65^{efg}
Ripe	38.99±3.81^{e}	3.28±0.23^{e}	229.38±14.58^{a}	109.61±8.76^{ab}	211.72±12.99^{de}
Rupali					
Green	ND	1.52±0.13^{b}	269.11±17.14^{bcd}	136.19±11.58^{cde}	164.49±9.88^{ab}
Breaker	8.23±1.20^{a}	2.21±0.28^{c}	314.54±12.07^{e}	161.43±16.74^{gh}	271.42±8.82^{i}
Turner	32.45±1.81^{d}	3.02±0.22^{de}	291.74±15.07^{def}	124.84±7.88^{bcd}	241.21±13.39^{efg}
Ripe	58.62±2.02^{g}	3.77±0.23^{f}	248.58±9.89^{ab}	113.88±11.96^{abc}	222.53±11.65^{ef}
NDT					
Green	ND	1.12±0.17^{a}	249.16±8.67^{ab}	124.48±10.26^{bcd}	172.17±10.58^{bc}
Breaker	6.52±1.16^{a}	2.22±0.22^{c}	287.87±12.53^{cde}	146.47±7.44^{defg}	259.52±10.31^{hi}
Turner	32.20±1.56^{d}	2.76±0.15^{d}	264.68±8.65^{bc}	109.79±9.54^{ab}	241.69±9.58^{efg}
Ripe	42.32±3.24^{f}	3.11±0.18^{de}	237.24±12.74^{a}	98.73±15.91^{a}	210.93±15.79^{de}

inhibition pathway by end products towards the end of fruit ripening (Egea *et al*. 2011). These mechanisms may include regulation at the transcriptional and/or post transcriptional level, metabolite flux into the carotenoid pathway and carotenoid sequestration has been found in tomatoes (Bramley, 2002). Difference among the cultivars were also evidenced at the ripe stage, where the highest amount of lycopene was detected in cv Rupali (58.62±2.02 mg/kg wb) followed by cv NDT (42.32±3.24 mg/kg wb), 1897 (38.99±3.81 mg/kg wb) and Himsona (34.40±3.08 mg/kg wb). The lowest amount of β-carotene was found at the green stage in cv 1897 (0.92±0.10 mg/kg wb). The highest amount of β-carotene content was analysed in cv Rupali (3.77±0.23 mg/kg wb) at ripe stage followed by cv 1897 (3.28±0.23 mg/kg wb), cv NDT (3.11±0.18 mg/kg wb) and cv Himsona (3.02±0.24 mg/kg wb). The result clearly showed that the lycopene and β-carotene content followed the similar pattern of increases from green to fully ripe stage of fruit ripening. The percentage increase in cv Rupali was nearly 612.27% and 148.02% in case of lycopene and β carotene respectively. The carotenoid content increased linearly at increasing maturation

stages. Cultivar Rupali showed highest content of lycopene at each ripening stage followed by NDT (Table 1). A study was done by Kotikova *et al*. (2011) on different varieties of tomato at different maturity supports the current findings of lycopene and carotene increment during the ripening of fruit. Gonçalves *et al*. (2019) reported an increase in lycopene content from 17.00-69.98 mg/100g in the reddish tomato at complete maturation. Similar results were reported by Gorecka *et al*. (2020) in tomatoes at different harvesting periods and the lycopene content ranges from 2.54-59.97 mg/kg and there was significant difference was found between the harvesting periods of cultivations.

4.2. Total Phenolics

Phenolic compounds, as secondary metabolites, are a large group of molecules widely distributed in fruits and vegetables. They are considered the main factors for the antioxidant capacity of plants and have also many benefits on human health, as free radical scavenger (Minatel *et al*. 2017). Recently, universal importance is given to identify the maturity stages with the best levels of polyphenols targeting increased functional properties of fruits and vegetables. But it is hard to estimate the real contents of phenolic compounds, due to the fact that the phenolic content of fruits and vegetables are largely influenced by many factors such as biotic and abiotic stress, senescence, and cultivar, tissue, harvesting time, post harvest treatment and also extraction techniques (Periago *et al*. 2009). Moreover, the content of phenolic compounds has varied significantly during fruit maturation, which collaborates with other researchers who suggested that phenolic compounds vary significantly between stages of maturity (Sukrasno and Yeoman 1993; Kondo *et al*. 2002). The accumulation of phenolic compounds was noticed in the intermediate stage of ripening. The highest phenolic content was found in cv Himsona (342.23±12.73 mg GAE/kg wb) at the breaker stage followed by cv Rupali (314.54±12.07 mg GAE/kg wb), cv 1897 (294.07±11.13 mg GAE/kg wb) and cv NDT (287.87±12.53 mg GAE/kg wb). The present results were higher than the earlier reported by (Periago *et al*. 2009) and i.e. phenolic compounds ranged between 66.66-235.00 mg GAE / kg and lower than results reported by Valverde *et al*. (2013) i.e total phenolic content ranged between 186.92-558.63 mg GAE/kg WB, corresponding to cv Ronaldo at green stage and cv Cherry Pera at red stage of ripening, respectively. Giudice *et al*. (2015) worked on three stages of tomato i.e. green, breaker and mature and reported the phenolic content between 212.30 mg/kg wb, 268.90 mg/kg wb, 169.70 mg/kg wb, respectively, which was similar to the current finding. As the results showed that maximum phenols was found at breaker

stage of ripening. Between the cultivars similar pattern of change in phenolic content were observed i.e. a peak in the amount of phenols was reached at the orange-red ripening stage (breaker) followed by turner. While Ilahy *et al*. (2011) reported variation among the cultivars some cultivar shows maximum phenols at orange-red ripening stage and some of them at green and green-orange stages of ripening.

4.3. Flavonoids

It is well known that flavonoids endow wide range of pharmacological and biochemical properties, such as antimicrobial activities, anti-inflammatory and inhibition of platelet aggregation (Kang *et al*. 2010). There was a significant variation in the accumulation of the total flavonoids. The maximum of flavonoid content was detected in the breaker stage in cv Himsona (171.1±14.92 mg QE/kg wb), cv Rupali (161.43±16.74 mg QE/kg wb), cv 1897 (154.55±16.56 mg QE/kg wb) and cv NDT (146.47±7.44 mg QE/kg wb) while low level was detected in the mature stages (98.73±15.91, 109.61±8.76, 113.88±11.96 and 147.85±8.34 mg QE/kg wb), in NDT, 1897, Rupali and Himsona, respectively). Flavonoid content follows the similar pattern of total phenolic compounds (Table 1). This result was in agreement with other authors who suggested that flavonoid content decrease with advanced maturity (Sukrasno and Yeoman 1993; Menichini *et al*. 2009). The flavonoid content decrease during ripening may be due to metabolic production of other phenolic compounds or degradation via enzyme action (Conforti *et al*. 2007). While, according to Ilahy *et al*. (2011) cv Rio Grande follow the similar pattern as decreasing the flavonoids at the ripening but other cvs showed contradictory results i.e. flavonoids increased at stages of ripening. Periago *et al*. (2009) reported flavonoid content of three cultivars and maturity stages of tomato ranged from 15.04-45.09 mg/kg which is lower than the current findings.

4.4. Ascorbic Acid

The ascorbic acid content showed the similar pattern as in phenolic and flavonoid content. Ascorbic acid contents were significantly different between the studied ripening stages ($p<0.05$). Such variability in ascorbic acid content during ripening was genotype. The highest amount of ascorbic acid was recorded at the breaker stage in cv Himsona (302.62±14.62 mg/kg wb), cv Rupali (271.42 ±8.82 mg/kg wb), cv NDT (259.52±10.31 mg/kg wb) and cv 1897 (247.28±18.28 mg/kg wb). The results account for the decrease of approximately 15-23 % from breaker to ripe stage of ripening in all selected cultivars. Results showed that, the ascorbic acid content were comparable to that reported by Kotikova *et al*.

(2011) and Ilahy *et al*. (2011) i.e. 217.00-258.00 mg/kg wb and 131.00-333.00 mg/kg wb, respectively. Table 1 shows that breaker stage of ripening has highest ascorbic acid content and all the cultivar follow same pattern. According to Nour *et al*. (2014) during the maturity of fruit ascorbic acid content increased continuously while at ripening ascorbic acid content continues to increase as maturity progressed from mature green to pink or light red stage and decreased afterward. The decrease in ascorbic acid could be attributed to its susceptibility to oxidative destruction as impacted by the ripening environments. While, according to Ilahy *et al*. (2011) only one cultivar Lyco 2 follow this pattern, while rest of the other cultivar showed that orange red or turner has highest amount of ascorbic acid content. Similar results were reported by Nour *et al*. (2015) that the ascorbic acid content increases with maturity and maximum value showed at deep red tomatoes. According to Dumas *et al*. (2003) changes might be seen due to environmental and agronomical factors. While Davey *et al*. (2000) explained that the ascorbic acid content is known to depend on factors such as cultivar, ripeness, and size, position on plant, light, soil type and indoor or outdoor cultivation. Periago *et al*. (2009) reported the lower values of ascorbic acid content ranges from 50.50-154.10 mg/kg wb at different ripening stages and ripe tomatoes have high ascorbic acid content as compared to others which is conflicting to the current results. Comparison with the values reported in the literature showed that the ascorbic acid content obtained here were somewhat in the range described by Nour *et al*. (2013) i.e. 91.90-329.70 mg/kg.

4.5. Antioxidant Activity of Tomato Extracts

The HAA and LAA determined by FRAP assay in tomato cvs at the four different ripening stages are shown in Table 2. Disregarding the analytical method used, for all tomato cvs, the higher HAA value was estimated in tomato fruits at the green stage of ripening and the lowest value was recorded at the red-ripe stage. Thus, the HAA exhibited a significant decrease during fruit ripening. Although Raffo *et al*. (2002) reported a similar trend for the HAA during fruit maturation of the cherry tomato cv Naomi, grown under green house conditions, different results were reported by Cano *et al*. (2003) who found that the HAA remained practically unchanged during ripening (ranging from 195-218 mM trolox100g wb) in the greenhouse grown tomato cv Marmande-Cuarenteno. Simultaneously to the reported decrease of HAA, an increase of LAA was evident in all tomato cvs as ripening process proceeds. This increase was in average slightly higher than 50% in red-ripe

tomato fruits compared to the green stage when LAA were measured with the FRAP assay. All the studied tomato cvs similarly exhibited a decline in HAA and an increase in LAA during a ripening process, it should be noted that at the red ripe stage of ripening both HAA and LAA values of cv 1897, Himsona, Rupali were respectively 5.2%, 12.0%, 80.0% and 2.2 %, 28.0%, 56.0% higher than NDT cv. This aspect could be considered as an indication of the superior antioxidant composition of such tomato cvs which suit the consumer requirement for nutritive and health foods. Many authors have studied correlations between bioactive compounds and antioxidant activities in numerous fruits and vegetables particularly tomatoes (Giovanelli *et al*. 1999; Raffo *et al*. 2002; Lenucci *et al*. 2006). In the present study, after considering data from all tomato cvs (Table 3), no significant correlation between HAA values measured by FRAP and total vitamin C ($R= -0.137$; $p<0.01$), phenolics ($R= 0.536$; $p<0.01$) were obtained, whereas they well correlated with flavonoids ($R= 0.469$; $p<0.01$). Although, it is likely that HAA depend upon synergistic effects among all hydrophilic antioxidants and there interaction with other constituents of the fraction (Diplock *et al* 1998; Lenucci *et al* 2006). Moreover, the test reaction used for antioxidant activity measurement might be differentially influenced by other compounds involved in the complex antioxidant system of tomato fruits, such as glutathione and enzymatic components (Jimenez *et al*. 2002). The

Table 2: Hydrophilic and lipophilic antioxidant activity of tomato cultivars harvested at four different ripening stages

Cultivars	*Ripening stages*	*FRAP assay (mm FRAP/g wb)*	
		Hydrophilic	*Lipophilic*
Himsona	Green	2.64±0.02^{d}	1.31±0.02^{a}
	Breaker	2.52±0.04^{c}	1.43±0.02^{a}
	Turner	2.29±0.03^{b}	1.69±0.01^{b}
	Ripe	2.07±0.03^{a}	2.13±0.03^{c}
1897	Green	2.29±0.03^{d}	0.62±0.02^{a}
	Breaker	1.74±0.05^{c}	0.84±0.01^{a}
	Turner	1.42±0.01^{b}	1.06±0.01^{b}
	Ripe	1.21±0.01^{a}	1.39±0.02^{c}
Rupali	Green	2.37±0.04^{d}	1.03±0.02^{a}
	Breaker	1.98±0.04^{c}	1.30±0.01^{b}
	Turner	1.62±0.02^{b}	1.49±0.03^{b}
	Ripe	1.29±0.03^{a}	1.74±0.02^{c}
NDT	Green	2.21±0.01^{d}	0.52±0.02^{a}
	Breaker	1.72±0.02^{c}	0.91±0.02^{b}
	Turner	1.48±0.03^{b}	1.03±0.01^{b}
	Ripe	1.15±0.02^{a}	1.36±0.01^{c}

LAA of tomato fruits has been mainly attributed to the presence of carotenoids particularly lycopene (Raffo *et al*. 2002; Cano *et al*. 2003; Toor and Savage 2005). After considering data from all tomato cvs, the significant correlations between FRAP values and both β-carotene(R=0.469; p<0.01) and lycopene (R= 0.442; p<0.01) contents were obtained during ripening (Table 3). It has been reported that antioxidants detected by FRAP are limited to water-soluble ones and that carotenoids, having no ferric reducing ability, should not react with this method (Apak *et al*. 2007). It should be underlined that no correspondence was found between the approximate increase in carotenoid contents in watermelon fruits (averaged across cultivars), when red-ripe compared to the white stage of ripeness, and the increase in LAA. This may probably be due to the presence of other lipophilic antioxidant compounds in the watermelon fruits which could account for most of the LAA at the white stage.

Table 3: Correlation coefficient and related significance between antioxidant content and antioxidant activity

Compounds	*FRAP assay*	
	R	*P*
Hydrophilic fraction		
Vitamin C	-0.137	<0.01
Phenolics	0.259	<0.01
Flavonoids	0.503	<0.01
Lipophilic fraction		
β-carotene	0.469	<0.01
Lycopene	0.442	<0.01

3.6. Color Values

The values color parameters (L*, a*, b*) and hue angle and chroma indexes of tomato fruits at different ripening stages given in table 4. During fruit ripening color parameter a* had a sharp increase, changing from negative to positive or green to red color, the highest increase being recorded between mature green and turning stages of ripening, evolution which is related to the beginning of carotenoids synthesis Nour *et al*. (2015). In the last three stages of ripening the color lightness (L*) decreased, indicating darkening of the tomato red color. The lowest ratio of the red and green color with a negative value (–6.01) was recorded in the green fruits of the cv Rupali while the highest value of color parameter a* (32.47) was recorded in completely ripe tomatoes of the cv NDT. Similar pattern was found with b* value and lowest ratio of blue and yellow color was recorded in the green fruit of the cv NDT (17.57) while highest value

of b* was reported in cv Rupali (36.57). Chroma is a color index representing the quantitative attributes of colorfulness, so that as chroma increases, color becomes more intense. Chroma index increases at during fruit ripeness stages of the cultivars. The lowest chroma value was detected in the green fruits of the cv NDT (18.21) and highest was recorded in same cv i.e. (48.13). Hue angle of all investigated cultivars significantly decreased during ripening. Similar results were reported by Kaur *et al*. (2006) and Nour *et al*. (2015) in tomato cvs at different maturity stages.

Table 4: Color values L*, a*, b* and hue angle and chroma indexes of tomato fruits at different ripeness stages

Cultivars	*L**	*a**	*b**	*Hue angle*	*Chroma*
Himsona					
Green	57.71±1.07^{fg}	-5.15±0.55^{a}	20.46±1.36^{abc}	-1.32±0.02^{a}	21.10±1.34^{b}
Breaker	55.73±1.10^{ef}	5.47±0.93^{b}	21.89±1.89^{bcd}	1.32±0.05^{f}	22.58±1.63^{bc}
Turner	51.36±2.96^{c}	15.32±1.55^{e}	24.02±1.94^{de}	1.00±0.06^{cd}	28.52±1.51^{e}
Ripe	45.35±1.80^{b}	27.36±1.40^{f}	26.96±1.85^{ef}	0.78±0.02^{b}	38.41±2.21^{gh}
1897					
Green	59.57±1.15^{g}	-4.54±0.59^{a}	20.20±1.79^{ab}	-1.35±0.02^{a}	20.71±1.90^{ab}
Breaker	54.77±1.2^{de}	6.62±1.07^{bc}	23.15±1.91^{bcd}	1.29±0.04^{f}	24.08±1.13^{cd}
Turner	50.47±1.03^{c}	15.44±1.00^{e}	28.78±2.08^{fg}	1.08±0.04^{d}	32.68±1.99^{f}
Ripe	45.88±1.86^{b}	27.00±2.24^{f}	30.32±2.39^{g}	0.84±0.06^{b}	40.65±2.19^{h}
Rupali					
Green	55.97±1.27^{ef}	-6.01±0.11^{a}	21.34±1.06^{bcd}	-1.29±0.01^{a}	22.16±1.05^{bc}
Breaker	50.48±1.73^{c}	8.63±0.76^{cd}	30.65±3.02^{g}	1.30±0.01^{f}	31.84±3.09^{f}
Turner	45.17±2.04^{b}	13.64±1.49^{e}	34.26±1.99^{h}	1.19±0.05^{e}	36.91±1.68^{g}
Ripe	41.70±1.55^{a}	26.12±3.25^{f}	36.57±3.00^{h}	0.95±0.09^{c}	45.06±1.84^{i}
NDT					
Green	58.98±1.81^{g}	-4.76±0.60^{a}	17.57±1.83^{a}	-1.30±0.04^{a}	18.21±1.74^{a}
Breaker	52.61±1.79^{cd}	9.46±1.98^{d}	23.75±2.12^{cd}	1.19±0.10^{e}	25.65±1.24^{d}
Turner	46.93±1.49^{b}	28.41±1.93^{f}	28.24±1.56^{fg}	0.78±0.03^{b}	40.07±1.40^{h}
Ripe	41.11±1.52^{a}	32.47±1.9^{g}	35.32±1.63^{h}	0.83±0.01^{b}	48.13±1.78^{j}

5. Conclusions

Data of this study highlight the important role played by genetic background and ripening stage in determining the antioxidant potential of tomato. The stages of ripening strongly influenced the bioactive compounds and antioxidant activity in selected tomato cultivars. The amount of lycopene and β-carotene, as well as the LAA, markedly increased in all the studied tomato cultivars with ripening, whereas the contents of total phenols, flavonoids, total ascorbic acid and HAA were affected by fruit ripening in a cultivar-dependent way.

The bioactive compounds namely lycopene, β-carotene in tomato cv Rupali was found to be higher in comparison to other cultivars. These compounds show the significant increase during ripening stages. Whereas the flavonoid, phenolics and ascorbic acid contents was found to be higher in cv Himsona during breaker stage followed by green, ripe and turner stages of ripening. The highest HAA value was estimated in tomato cv Himsona at the green stage of ripening and the lowest value was recorded at the red-ripe stage. Thus, the HAA exhibited a significant decrease during fruit ripening whereas the LAA exhibit a significant increase during ripening stages. Highest positive correlation is found between FRAP and flavonoids ($R=0.503$; $p<0.01$). Whereas β-carotene (0.469; $p<0.01$) show positive correlation with FRAP values but lesser than HAA. Hence, the increase of antioxidants during tomato fruit ripening must not be generalized; rather, their contents reach levels leading to the maximum nutritional quality depending on their genotypic differences. The study of changes in bioactive compounds and antioxidant activity during ripening is of great relevance both to human health and commercial purposes. In fact, it provides valuable information to understand the synthesis of these compounds and for evaluating the best harvesting period to reach the highest antioxidant potential. These data are useful to plant breeders and food scientists working to maximize the nutritional value and antioxidant contents of fresh tomato cultivars. Furthermore, they confirm that this fruit represents a nutritionally balanced source of dietary antioxidants for the population because of its availability and high consumption.

Disclosure Statement

The authors have declared no conflict of interest.

Acknowledgements

The authors are highly thankful to centre of Food Technology University of Allahabad, for providing facilities to carry out the investigation.

References

Amir H, Karas M, Giat J, Danilenko M, Levy R, Yermiahu T & Sharoni Y. 1999. Lycopene and 1, 25-dihydroxyvitamin D3 cooperate in the inhibition of cell cycle progression and induction of differentiation in HL-60 leukemic cells. Nutrition Cancer, 33, 105-112.

Apak R, Guclu K, Demirata B, Ozyürek M, Celik SE, Bektasoglu B & Ozyurt D. 2007. Comparative evaluation of various total antioxidant capacity assays applied to phenolic compounds with the CUPRAC assay. Molecules, 12, 1496-1547.

Benzie IF & Strain JJ. 1996. The ferric reducing ability of plasma (FRAP) as a measure of "antioxidant power": the FRAP assay. Analytical Biochemistry, 239, 70-76.

Bramley PM. 2002. Regulation of carotenoid formation during tomato fruit ripening and development. Journal of Experimental Botany, 53, 2107-2113.

Conforti F, Statti GA & Menichini F. 2007. Chemical and biological variability of hot pepper fruits (*Capsicum annuum* var. *acuminatum* L.) in relation to maturity stage. Food Chemistry, 102, 1096-1104.

Cano A, Acosta M & Arnao MB. 2003. Hydrophilic and lipophilic antioxidant activity changes during on-vine ripening of tomatoes (*Lycopersicon esculentum* Mill.). Postharvest Biology Technology, 28, 59-65.

Davey MW, Montagu MV, Inze D, Sanmartin M, Kanellis A, Smirnoff N & Fletcher J. 2000. Plant L-ascorbic acid: chemistry, function, metabolism, bioavailability and effects of processing. Journal of the Science and Food Agriculture, 80, 825-860.

Del Giudice R, Raiola A, Tenore GC, Frusciante L, Barone A, Monti DM & Rigano MM. 2015. Antioxidant bioactive compounds in tomato fruits at different ripening stages and their effects on normal and cancer cells. Journal of Functional Foods, 18, 83-94.

Diplock AT, Charuleux JL, Crozier-Willi G, Kok FJ, Rice-Evans C, Roberfroid M & Vina-Ribes J. 1998. Functional food science and defence against reactive oxidative species. British Journal of Nutrition, 80, S77-S112.

Dumas Y, Dadomo M, Di Lucca G and Grolier P. 2003. Effects of environmental factors and agricultural techniques on antioxidant content of tomatoes. Journal of Science and Food Agriculture, 83, 369-382.

Egea I, Bian W, Barsan C, Jauneau A, Pech JC, Latche A & Chervin C. 2011. Chloroplast to chromoplast transition in tomato fruit: spectral confocal microscopy analyses of carotenoids and chlorophylls in isolated plastids and time-lapse recording on intact live tissue. Annals of Botany, 108, 291-297.

Eldridge T, Łangowski Ł, Stacey N, Jantzen F, Moubayidin L, Sicard A & Ostergaard L. 2016. Fruit shape diversity in the Brassicaceae is generated by varying patterns of anisotropy. Development, 143, 3394-3406.

Garcia E & Barrett DM. 2006. Evaluation of processing tomatoes from two consecutive growing seasons: quality attributes, peelability and yield. Journal of Food Process and Preservation, 30, 20-36.

George B, Kaur C, Khurdiya DS & Kapoor HC. 2004. Antioxidants in tomato (*Lycopersium esculentum*) as a function of genotype. Food Chemistry, 84, 45-51.

Giovanelli G, Zanoni B, Lavelli V & Nani R. 2002. Water sorption, drying and antioxidant properties of dried tomato products. Journal of Food Engineering, 52, 135-141.

Goncalves DC, Morgado CMA, Silva EP, Correa DCG, Nascimento DRA, Junior LCC & Aguiar DOFC. 2020. Postharvest behavior and lycopene content of tomatoes at different harvest times. Acta Scientiarum Polonorum Technologia, 42, 48403-48403.

Gorecka D, Wawrzyniak A, Golinska JA, Dziedzic K, Hamułka J, Kowalczewski PŁ & Walkowiak J. 2020. Lycopene in tomatoes and tomato products. Open Chemistry, 18, 752-756.

Ilahy R, Hdider C, Lenucci MS, Tlili I & Dalessandro G. 2011. Antioxidant activity and bioactive compound changes during fruit ripening of high-lycopene tomato cultivars. Journal of Food Composition and Analyses, 24, 588-595.

Jimenez A, Creissen G, Kular B, Firmin J, Robinson S, Verhoeyen M & Mullineaux P. 2002. Changes in oxidative processes and components of the antioxidant system during tomato fruit ripening. Planta, 214, 751-758.

Kang J, Li Z, Wu T, Jensen GS, Schauss AG & Wu X. 2010. Anti-oxidant capacities of flavonoid compounds isolated from acai pulp (*Euterpe oleracea* Mart.). Food Chemistry, 122, 610-617.

Karakaya SNEl & Tas AAS. 2001. Antioxidant activity of some foods containing phenolic compounds. International Journal of Food Science and Nutrition, 52, 501-508.

Kaur D, Sharma R, Wani AA, Gill SB & Sogi DS. 2006. Physicochemical changes in seven tomato (*Lycopersicon esculentum*) cultivars during ripening. International Journal of Food Properties, 9, 747-757.

Kondo S, Tsuda K, Muto N & Ueda JE. 2002. Antioxidative activity of apple skin or flesh extracts associated with fruit development on selected apple cultivars. Scientia Horticulturae, 96, 177-185.

Kotikova Z, Lachman J, Hejtmankova A & Hejtmankova K. 2011. Determination of antioxidant activity and antioxidant content in tomato varieties and evaluation of mutual interactions between antioxidants. Food Science and Technology, 44, 1703-1710.

Lenucci MS, Cadinu D, Taurino M, Piro G & Dalessandro G. 2006. Antioxidant composition in cherry and high-pigment tomato cultivars. Journal of Agriculture Food Chemistry, 54, 2606-2613.

Menichini F, Tundis R, Bonesi M, Loizzo MR, Conforti F, Statti G & Menichini F. 2009. The influence of fruit ripening on the phytochemical content and biological activity of *Capsicum chinense* Jacq. cv Habanero. Food Chemistry, 114, 553-560.

Minatel IO, Borges CV, Ferreira MI, Gomez HAG, Chen CYO & Lima GPP. 2017. Phenolic compounds: Functional properties, impact of processing and bioavailability. Phenolic Compounds Biological Activity. Education Technology in Rijeka, Croatia, 1-24.

Nour V, Trandafir I & Ionica ME. 2013. Antioxidant compounds, mineral content and antioxidant activity of several tomato cultivars grown in Southwestern Romania. Notulae Botanicae Horti Agrobotanici Cluj-Napoca, 41, 136-142.

Nour V, Trandafir I & Ionica ME. 2014. Evolution of antioxidant activity and bioactive compounds in tomato (*Lycopersicon esculentum* Mill.) fruits during growth and ripening. Journal of Applied Botany and Food Quality, 87.

Nour V, Ionica ME & Trandafir I. 2015. Bioactive compounds, antioxidant activity and color of hydroponic tomato fruits at different stages of ripening. Notulae Botanicae Horti Agrobotanici Cluj-Napoca, 43, 404-412.

Pastori M, Fander PH, Boscoboinik D & Azzi A. 1998. Lycopene in association with α-tocopherol inhibits at physiological concentrations proliferation of prostate carcinoma cells. Biochemical and Biophysical Research Communication, 250, 582-585.

Pellegrini N, Colombi B, Salvatore S, Brenna OV, Galaverna G, Rio DD & Brighenti F. 2007. Evaluation of antioxidant capacity of some fruit and vegetable foods: efficiency of extraction of a sequence of solvents. Journal of Science and Food Agriculture, 87, 103-111.

Periago JM, Alonso GJ, Jacob K, Olivares BA, Bernal JM, Iniesta DM & Ros G. 2009. Bioactive compounds, folates and antioxidant properties of tomatoes (*Lycopersicum esculentum*) during vine ripening. International Journal of Food Science and Nutrition, 60, 694-708.

Raffo A, La Malfa G, Fogliano V, Maiani G & Quaglia G. 2006. Seasonal variations in antioxidant components of cherry tomatoes (*Lycopersicon esculentum* cv. Naomi F1). Journal of Food Composition and Analyses, 19, 11-19.

Robards K, Prenzler PD, Tucker G, Swatsitang P & Glover W. 1999. Phenolic compounds and their role in oxidative processes in fruits. Food Chemistry, 66, 401-436.

Singh V, Mishra S, Singh J & Rai AK. 2016. Phenolic content and antioxidant activity of solvent extracts of mahua (*Madhuca longifolia*) flowers and Fruit. Nutrafoods, 16, 1017.

Sukrasno N & Yeoman MM. 1993. Phenylpropanoid metabolism during growth and development of *Capsicum frutescens* fruits. Phytochemical, 32, 839-844.

Valverde GV, Gonzalez NI, Alonso GJ & Periago MJ. 2013. Antioxidant bioactive compounds in selected industrial processing and fresh consumption tomato cultivars. Food and Bioprocessing Technology, 6, 391-402.

Zhishen J, Mengcheng T, & Jianming W. 1999. The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. Food Chemistry, 64, 555-559.