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DEVELOPMENT OF LYCOPENE CONTENT IN TOMATOES AT COLOUR BREAK AND VARIATION OF IT DURING STORAGE AND PROCESSING

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ABSTRACT

Lycopene is a natural pigment present in tomatoes and is responsible for red colour, Lycopene content in tomatoes is varied based on the variety, climatic condition, soil type and rainfall pattern in cultivated areas and also on the maturity stage of the fruit. So also, it is varied with the stage of harvest, harvesting season, storage condition, storage period and different types of processing. Harvesting at the colour break stage and storing at low temperatures (22^0 C – 30^0C) is favourable for lycopene development rather than storing at high temperatures. Ethylene gas treatment can be used to enhance the lycopene content and it accelerates the ripening process in tomatoes. Processing at high temperatures for long periods and exposure to bright lights causes to decrease the lycopene content.Further, processing of tomatoes increases the lycopene bioavailability and many health benefits according to the results of previous studies. Processing waste of tomato skin and pericarp can be used as a lycopene source for the food industry as well as to the formation of many tablets which are enriched with many vitamins and nutrients. Extracted lycopene can be used as a natural food colorant for artificial colorants in avoiding harmful effects. There is an emerging future trend in preparing many products by increasing the lycopene content in the natural product on the well-being of the consumers.

1. Introduction

Tomato (*Lycopersiconesculentum*) is one of the commonly traded vegetables, cultivated throughout the world. It contains important nutrients such as vitamin A, C, minerals and carotenoids along with lycopene (Eboigbe and Edemevughe, 2019). The most abundant carotenoid in ripe tomatoes is lycopene and it comprises approximately 80% to 90% of all pigments present. (Curl, 1961; Ilahy *et al.*, 2011;Thakur, B.R *et al.*, 1996). Recent researches have revealed that lycopene is a very effective natural antioxidant (Mascio *et al.*,1989). Lycopene is found predominantly in tomatoes and the content of lycopene varies widely among tomato varieties and increases dramatically during ripening (Clinton, 2009; Ajlouni *et al.*, 2001). Lycopene content in tomatoes after harvesting has been reported to be dependent upon the ripeness of the fruit at the time of harvesting (Sadler *et al.*, 1990) and also it presents in fresh tomato as trans configuration and due to the occurrence of isomerization and oxidation during tomato processing,a high amount of lycopene gets degraded. This isomerization converts trans isomers into cis isomers. (Shi and Maguer, 2000). It has been suggested that the bioavailability of cis-lycopene in tomatoes is higher than that of all trans-lycopene (Choksi and Joshi, 2007).

Functional food is defined as foods that provide health benefits beyond basic nutrition by the International Food Information Council (IFIC). Considering the composition and health benefits of tomatoes, they can be categorized as a functional food (Choksi and Joshi, 2007).

2. Chemical structure and properties of Lycopene

Lycopene is a carotenoid found in plant materials possessing antioxidant properties and is responsible for the red coloured pigment in fruits and vegetables. Redness is also affected by the content of lycopene present in the material and tomato can be given as an example (Suwanaruang, 2016; Khairi *et al*., 2015). The Color given by lycopene is matters the functional quality and horticultural performance of tomatoes at the stage of marketing (Ilahy *et al*., 2018).

Lycopene is confined in chloroplasts at the cellular level of tomato fruit and in the early maturity stage of tomato, green chlorophyll is the major pigment presenting in the chloroplast. At the time, when chlorophyll is reduced, lycopene is biosynthesized with different changes in the ultrastructure of the material resulting in the developing of red colour (Shi and Maguer, 2000; Harris, 1970; Khudairi, 1972; Matienco and Yedalty, 1973).

In nature, all lycopene present as trans form and can be isomerized from trans form to cis form at presenting the influence of light, heat or certain chemicals. Trans isomers of lycopene are the predominant isomer type in tomato and represent about 95.4% of total lycopene and they are most thermodynamically stable. During processing and storage of tomatoes, isomerization occurred and thus, a stable form of trans isomers is converted into an unstable form of cis isomers (Shi and Maguer, 2000; Barrett *et al*., 2001).

Chemically lycopene is an acyclic hydrocarbon, an acyclic unsaturated open chain including 13 double bonds, of which 11 are conjugated double bonds arranged linearly and possessing of molecular formula of $C_{40}H_{56}$. Methyl groups are presenting at 1, 5 position while two central methyl groups are at 1,6 position relative to each other. Colour and antioxidant properties are prevailing due to the unique structure and extended system of conjugation (Shi and Maguer, 2000). The antioxidant property given by lycopene is caused by its capacity to sequester singlet oxygen and also the ability to trap peroxyl radicals (Amany *et al*., 2009).

Due to having conjugated unsaturated double bonds in lycopene structure, it is susceptible to deterioration in the presence of light, heat and oxidants. At the industrial applications, adequate consideration should be given when the presence of chemicals, heat and light at the processing of the food matrix which includes lycopene (Shi and Maguer, 2000; Gomes *et al*, 2014). According to the main physical properties of lycopene, molecular weight, melting point, crystal and powder forms and solubility are given in table 01 (Shi and Maguer, 2000).

Table 1. Physical properties of Lycopene

Factor	**Value/ Details**
Molecular Formula	$C_{40}H_{56}$
Molecular weight	536.85 Da
Melting Point	172 – 175 ^{0}C
Crystal form	Long red needle form
Powder Form	Dark reddish brown powder
Solubility	Soluble in Chloroform, hexane, benzene, acetone, petroleum ether Insoluble in water, ethanol and methanol
Sensitivity	Sensitive to light, high temperatures, oxygen and acids

3.Variation of lycopene contents in tomatoes at the field

Lycopene contents in fresh tomatoes at the field are varied mainly on the cultivation regions of the world (Eboigbe and Edemevughe, 2019) and also depending on many internal and external factors such as tomato variety, maturity stage, soil type and environmental conditions. Some tomato varieties at the deep red stage contain more lycopene (15 mg per 100 g of fresh tomatoes) than yellow coloured tomato varieties (0.5 mg per 100 g of fresh tomatoes) (Hart and Scott, 1995; Kotikova *et al.,* 2011).

Lycopene content of high lycopene breeding lines which was grown under open field conditions during the year 2013 -2014 in Tunisia ranged from 98.8 to 280.0 mg/kg fresh weight of tomatoes. Lycopene accumulation potential in tomatoes gets varied due to having differences in temperature and prolonged heat stress might be responsible for less amount of lycopene content in tomatoes at the harvesting stage (Ilahy *et al.,* 2016).

Effect of environmental conditions on colour and lycopene content of tomato at ripening stage was analyzed and reported that lycopene synthesis is inhibited when the temperature of the fruit is exceeding 30 °C (Brandt *et al.,* 2006; Anese, *et al.,* 2002). Best Lycopene development is achieved when the temperature is between 12^0C and 21^0C (Brandt *et al.,* 2006). Further, Lycopene content of tomatoes generally ranging from 7 to 13 mg/100g based on the type of variety, climatic conditions, geographical areas and cultivation techniques (Schierle*et al*., 1997). The average value of lycopene content varied between 84.1 to 172.9 mg/kg whiles some tomato varieties have particularly high contents of lycopene in many countries (Barrett and Anthon, 2001). Some of the red tomato varieties such as “Flavour top” or “Money marker” contain around 50 mg/kg fw of lycopene when yellow varieties have around 5 mg/kg in fresh weight (Choksi and Joshi, 2007).

Significantly higher lycopene content was observed in glass house grown tomatoes (83.0 mg kg^{-1} f.w) than that growing in the field (59.2 mg kg^{-1} f.w) at a different time of harvesting (Brandt, S.,*et al*.,2003).

Lycopene content in tomatoes has been reported to be dependent upon the stage of maturity (Thompson *et al*., 2000; Fraser *et al.*, 1994; Sadler *et al*., 1990) and it was at the immature green stage, mature green stage, colour breaker stage, firm red stage and overripe stage were reported as 25, 10, 370, 4600 and 7050 μg/100 g respectively (Fraser *et al.,* 1994). When maturation was completed, lycopene content was increased from 17 mg/ 100g to 69.98 mg/100g in reddish tomatoes (Kotikova *et al*, 2011).

4. Variation of the lycopene content at the storage

Lycopene content of red colour ripe tomatoes which were stored at room temperature (22^0C) was higher than the tomatoes which were stored at refrigeration temperature (4°C) (Ajlouni*et al.,* 2001). Biosynthesis of lycopene depends upon the temperature range between 12^0C and 32^0C (Leoni,1992) and for optimizing the lycopene content, the suitable temperatures were 16 ^{0}C–18 °C and 26 °C (Turk *et al*., 1994). A similar conclusion has indicated that lycopene contents were high in ripe tomatoes stored at 20°C for 10 days (Hamauzu *et al*., 1998).

Tomatoes harvested at the colour break stage and stored at room temperature (22 °C – 23 °C) for 06 days had significantly higher lycopene content than that harvested at the mature green stage, treated with ethylene and stored for 06 days. The lycopene for tomatoes that were stored over 9 days at room temperature (22 °C – 23 °C) are given in table 02 (Thompson *et al.,* 2000). Lycopene biosynthesis was inhibited at temperatures above 30^0C and below 12^0C and stopped at temperatures above 35^0C, even though it was favored at temperatures between 16^0C to 21^0C. Lycopene biosynthesis is also influenced by the light intensity at ripening (Brandt *et al*., 2006; Leoni,1999; Dumas *et al*., 2003; Helyes*et al*., 2003). Studies on temperature effect on lycopene biosynthesis have shown that temperatures below 12°C strongly inhibited lycopene biosynthesis and temperatures above 35°C stopped this process (Dumas *et al*., 2003). Lycopene content was also found to be influenced by light intensity during ripening (Helyes *et al*., 2003).

Table 2. Lycopene content (mg/100 g) of various tomato cultivars on days 0, 6, 9, and 12 at mature green, breaker, and red ripe stages.

	Maturity stage of the cultivar	Day 0	Day 6	Day 9	Day 12
Agriset	Green	8 (11)	1598^{d} (308)	3744^{cd} (809)	2564^{f} (349)
	Breaker	942^{b} (415)	4574^{b} (1030)	3174^{d} (854)	4276^{bcde} (1152)
	Red	4154^{c} (856)	ND	ND	ND
Solar Set	Green	7 (5)	2502^{cd} (552)	4803^{b} (602)	3638^{def} (620)
	Breaker	1084^{b} (321)	5636^{a} (1008)	4267^{bc} (797)	4489^{abcde} (1380)
	Red	4419^{bc} (742)	ND	ND	ND
Suncoast (og)	Green	8 (9)	3326^{c} (323)	4847^{b} (1786)	4257^{bcde} (842)
	Breaker	1210^{ab} (218)	6207^{a} (1807)	6178^{a} (678)	4571^{abcde} (1694)
	Red	5274^{ab} (998)	ND	ND	ND
FL7692D (og)	Green	7 (6)	2015^{d} (362)	4528^{bc} (630)	3699^{cdef} (1036)
	Breaker	1511^{a} (216)	4589^{b} (917)	4616^{bc} (784)	3077^{de}(1148)
	Red	5560^{a} (597)	ND	ND	ND

[a-f] The day 0 samples of the four varieties were compared at each maturity stage within the column, while both maturity stages of the day 6, day 9, and day 12 samples were compared within the column. Means

The rate of lycopene synthesis in non-hydroponic tomatoes was higher than the tomatoes grown in the hydroponic cultivar. Tomatoes at a similar degree of maturity were used from two different cultivations such as hydroponic and non-hydroponic. Different samples of tomatoes of two cultivation types were equally divided and stored at room temperature (22°C) and refrigerated temperature (4°C) for 21 days and changes in lycopene contents and colour were measured. Results revealed that the lycopene and colour value was increased with the storage period. Initially, lycopene contents were 36.15±4.17 µg/g fresh weight for both hydroponic and non-hydroponic tomato fruits and it was continuously increased with the storage while recording maximum lycopene content at 14th day of storage at 22°C and reached up to 89.75±4.51 µg/g fresh weight and 115.13±2.08 µg/g fr. wt. for hydroponic and non-hydroponic cultivars, respectively. At 4°C, the increment in lycopene content in non- hydroponic tomatoes was less than the same sample stored at 22°C for the same storage period (Ajlouni *et al*., 2001).

Lycopene content of tomato fruits which were provided with 100% of optimal water supply at the field was less than that of the tomato samples supplied with 50% of optimal water at the field (Brandt *et al*., 2003) and showed variation in lycopene content in tomatoes subjected to water stress compared to no water stress tomatoes at storage (Khairi and Takahashi, 2013) while moderated water stress tomatoes that cultivated in a greenhouse and hydroponically maintained, contains high lycopene (Khairiet al., 2015). Tomatoes were hydroponically grown in a greenhouse and harvested fruit samples were stored in different temperatures such as at 10,15,20,25 and 30^{0}C in cool incubators for 07 days and analyzed for lycopene content and colour variation (table 03). Results revealed that the color of the tomatoes which were stored above 10^{0}C had increased after the storage and the lycopene content of tomatoes had increased at storage when fruits are stored above 20^{0}C (Takahashi *et al.,* 2018).

Table 3. Effect of storage temperatures and duration on lycopene content of the tomato fruits

Storage temperature (^{0}C)	Lycopene content (mg/100 g FW) after storage		
	2 days	4 days	7 days
5	18.4 ±1.2 a	13.8 ±2.1 b	14.5 ±0.7 c
10	17.5 ±1.5 a	16.7 ±1.0 ab	16.8 ±1.7 bc
15	16.4 ±1.7 a	17.2 ±0.9 ab	17.9 ±1.7 abc
20	22.9 ±2.4 a	23.4 ±1.9 a	25.7 ±0.3 a
25	23.6 ±0.8 a	23.6 ±1.8 a	24.3 ±2.4 ab
30	23.3 ±3.7 a	21.9 ±4.1 ab	24.1 ±3.0 ab

* Values are means ± standard deviation (n=4). Different letters indicate a significant difference ($p<0.05$) according to one-way ANOVA followed by Tukey-Kramer test in within columns

Tomato samples were taken for the study from cultivations that maintained irrigation systems with moderate water stress and normal irrigation and the harvested tomatoes were stored in different temperatures (10^{0}C, 15^{0}C, 25^{0}C and 30^{0}C) until reached to mature stage. Thereafter, the lycopene content was analyzed using a spectrophotometer. The initial lycopene content of water stress tomatoes and normal irrigated tomatoes were 7.024 mg/100g and 6.459 mg/ 100g respectively. Lycopene of water stress tomatoes showed an increasing trend during storage and the highest lycopene content was obtained at 12thday storage at 25 ^{0}C temperature. Lycopene content was increased at a temperature above 15^{0}C for both types of tomatoes obtained from water stress and normal irrigated conditions. After 8th day in storage, lycopene content tends to get reduced. Lycopene content was high for water stress tomatoes than that of no water stress while on storage at all temperatures aforementioned. Test of optimum storage condition for having high lycopene content was done by surface and contour plot analysis and results are shown in table 4 (Khairi *et al.*, 2018).

Table 4. Regression analysis test for optimizing model

	Cons-tant	temperatures	times	Lyco-pene	Temp. Vs Temp.	Times Vs times	Lyco-pene Vs Lyco-pene	Temp. Vs times	Temp. Vs Lyco-pene	Time Vs Lyco-pene
F	0.00	0.785	0.298	0.522	0.496	0.071	0.898	0.555	0.813	0.899

5. Stability of Lycopene during tomato processing

Lycopene bioavailability in unprocessed fresh tomatoes is less than that of processed tomato products. However, different food processing methods caused to improvement of the bioavailability of lycopene by breaking down the cell walls in tomatoes. As a result that, lycopene availability is high in cooked tomatoes than in fresh tomatoes (Shi and Maguer, 2000; Gartner *et al.*, 1997; Barrett and Anthon, 2001; Stahl and Sies, 1992). Lycopene degradation and loss of colour in processed tomato products are occurred due to a number of factors like high-temperature treatment and long duration in storage etc. The main reasons for lycopene degradation during tomato processing are isomerization and oxidation of lycopene (Shi and Maguer, 2000). The heating of tomatoes at high temperatures is not favourable due to the degradation of lycopene content; because temperature negatively affects the nature and extent of lycopene breakdown. Holding time of tomatoes at high temperature during processing is also seriously affects the degradation of lycopene. The rate of Lycopene loss during heating of tomato juice is high during processing at high temperatures as well as prolonged processing (Table 5).

Table 5. Rate of Lycopene loss of tomato juice during heating

Heating temperature/ ^{0}C	Lycopene loss percentage		
	Heating for 1 min	Heating for 3 min	Heating for 7 min
90	0.6	0.9	1.1
100	0.9	1.4	1.7
110	2.2	3.2	4.4
115	2.7	4.5	7.0
118	3.7	6.0	9.1
121	4.6	7.3	10.6
124	5.5	8.5	12.5
127	6.5	9.9	14.6
130	7.4	11.5	17.1

Source: Miki and Akatsu, 1970.

During vacuum evaporation, a small amount of lycopene loss was noticed. When the tomato was processed at a high temperature within the shortest possible time, it is favorable to retain the quality of the final product particularly lycopene content (Shi and Maguer, 2000). Lycopene bioavailability of tomato paste is higher than of fresh tomato (Gartner *et al.*, 1997). Lycopene contents of fresh tomatoes and tomato products drawn from the Romanian market were evaluated and found that fresh tomato contained 12mg/ 100g of lycopene and tomato products like tomato paste, tomato boiled sauce, tomato ketchup and spaghetti sauce contained about 16, 4, 17and 16mg/100g of lycopene respectively (Alda *et al*., 2009).
When tomatoes were boiling for 1 hour with the presence of 1% corn oil, caused to increase the lycopene bioavailability in tomato juice (Cogdell, 1985). By heating the tomatoes in the presence of metallic ions (Cu^{2+}, Fe^{3+}, etc.) or oxygen, lycopene may be partially destroyed (Shi and Maguer, 2000).

Lycopene content of tomato powder varied from 1016.05 to 1181.30 μg/g and total solids and loss is around 8.07% to 20.93% when the preparation of tomato powder using spray drying technology without adding any carrier agent (Goula and Adamopoulos, 2005). Water loss has occurred during the processing of tomato products and it also may be contributed to an increase in the concentration of lycopene content compared to fresh produce (Thompson *et al.,* 2000).

6. Future Developments

Tomato skin, seeds and pericarp tissues are the byproducts that get wasted during the processing of tomato based food products and these skin and outer pericarp tissues contain more than 80% to 90% of the total amount of lycopene in tomato fruit (Shi and Maguer, 2000) and tomato skin contains 12 mg lycopene/ 100 g (wet basis) while whole mature tomato contains 3.4 mg lycopene/100 g (wet basis) (Al-Wandawi*et al.,* 1985). Tomato skin contained more lycopene (53.9 mg/ 100 g) than whole tomato pulp (11mg/100g) on the wet basis (Sharma and Maguer, 1996). Commercial products enriched with lycopene in the pharmaceutical industry are less and there is a high demand for industrial production of lycopene from tomatoes (Shi and Maguer, 2000). Processing waste of tomato skin and pericarp can be used as a lycopene source for the food industry as well as to the formation of many tablets which are enriched with many vitamins and nutrients for the well-being of human health. Therefore more attention should be focused in producing value-added products from lycopene from byproducts of tomato based industries (Shi and Maguer, 2000; Choksi and Joshi, 2007).

The main reason for the degradation of lycopene during food processing and storage is oxidation. Due to that lycopene is needed to be protected from excess heat applications, extremely acidic or basic conditions, exposure to oxygen and high intensity of lights and lipid degrading enzymes to avoid lycopene from isomerization and oxidation. Application of one of the suitable antioxidants like ethoxyquin, ascorbic acid and sodium acid pyrophosphate may be given positive results (Granado *et al.*, 1992; Clindon *et al.*, 1996; Porrini and Testolin, 1998). Isolation of lycopene from tomatoes could be efficiently carried out by the Super Critical Fluid Extraction (SCF) process (Gomez *et al.*, 2003). Lycopene can be used as a natural food colorant as an alternative to artificial colorants due to avoiding the harmful effects of artificial food colorants (Agarwal and Rao, 1998).

7. Conclusions

Throughout this review, it has been shown that Lycopene is present in tomatoes and responsible for its colour and many bio-active properties. The storage condition and harvesting stage significantly affect the amount of lycopene present in tomatoes. Processing of tomatoes or tomato puree caused to increment of lycopene bioavailability and in extreme heat treatment effect for degradation of lycopene. Immerging improvements in extraction of lycopene from plant materials and processing wastes of tomato skin and seeds and then incorporation of lycopene in the pharmaceutical industry are testing. Super Critical Fluid Extraction has been successfully carried out in the extraction of lycopene from ripe tomatoes

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