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Extraction and Utilization of Tomato Peels Lycopene as Antioxidant and Natural Colorants in Beef Burger

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ABSTRACT

The tomato processing industry generates large amounts of waste, mainly tomato peels which create environmental problems. These residues are attractive sources of bioactive ingredients and pigments. Lycopene is one of the carotenoids, a fat-soluble pigment that has antioxidant properties. This study was conducted to evaluate the stability of lycopene and its antioxidant properties and its impact on these treatments. The degree of stability of lycopene was also studied during storage. After total lycopene compounds were assessed and determined by HPLC. When the colors were separated in tomato husks, it was found to contain two major components, lycopene and beta-carotene. Total phenolic compounds and total flavonoids contents were 3.75 and 0.97 mg/g, lycopene content was 3.75 mg/100g. As well as, antioxidant activity of tomato peels was 38.31% (as DPPH). In some lycopene treatments it was observed that the rate of degradation was increased by increasing pH, temperature and light by increasing the exposure time and the length of storage. The proportion of antioxidant activity of lycopene was close to the known industrial BHT. Where, the levels of crude lycopene extract 100, 200 and 300 ppm had an antioxidant activity were 0.96, 0.98 and 0.99, respectively, compared with those of BHT (1.09). In the sensory evaluation, the best percentage of natural lycopene added was 1% was preferred by the consumer. Storage for beef burger 12 weeks, the amount and remained of lycopene pigments decreased from 0.470 to 0.416 mg/100g, or from 100 to 88.51% during storage period. Meanwhile, lycopene degradation rate increased from 0.0 to 11.49%.

Keywords: Tomato peels; Lycopene; Stability; Rancimat; Antioxidant; Natural Colorants and Beef Burger

INTRODUCTION

Processing of fruits and vegetables is one of the largest manufacturing processes for waste production in the environment after sanitation. Liquid and solid waste from the fruit and vegetable industry is very large, which contains high percentages of high-value materials that can be reused again, and their economic value is not few, and such waste such as husks, peels and seeds of fruit and vegetables, stone cores and unused meat. Thus, the use of these by-products in the production of food additives or dietary supplements of nutritional importance has gained increasing attention and therefore their recovery of use is economically attractive (Gowe, 2015). Natural pigments, we can get them from some plants as well as minerals, and curiously from insects as well. Many natural pigments colors are found in bark, roots, leaves, flowers, skins, and shells of plants. Unlike the synthetic pigment, they are carcinogenic, but natural pigments are environmentally friendly and can be used in some applications (Sivakumar *et al*., 2011). Moreover, people are increasingly preferring natural pigments over synthetic colorants, which are harmless or even healthy (Azeredo, 2009). Lycopene is a red tincture characteristic of some fruits and vegetables, including ripe tomato products. It comes from a natural source of antioxidants, so it has attracted attention because of its biological and physical properties. Lycopene, a tincture that traces red carotenoids in tomatoes and tomato-based products, is a common form of beta-carotene but without provitamin A activity. Because of its proven benefits for coronary heart disease and chronic diseases, it has recently attracted considerable attention. The molecular lycopene weight is 536. 89 and its chemical formula is $C_{40}H_{56}$, 89.45% carbon and 10.51% hydrogen. Unsaturated hydrocarbon lycopene contains two unpaired conjugates (Aghel *et al*., 2011). Tomatoes (*Lycopersicon esculentum L*.) are one of the most productive and consumed vegetables with which the potato crop is involved (Kalogeropoulos *et al*., 2012). Tomatoes supply more than 85% of the total need for lycopene due to the high consumption of fresh tomatoes and their products (Amiri-Rigi and Abbasi, 2016). In the manufacture of tomatoes, large quantities of tomato residues (5-30% of the main product) are produced as food waste or by-products, mainly used as livestock feed or buried in landfills. The seeds account for about 33%, the skin 27%, the pulp 40% of the tomato pomace, while the dry matter contains about 56% of the pulp skin and 44% of the seeds (Poojary and Passamonti, 2015). Lycopene and other carotenoids are extracted from tomato pomace, especially from tomato skin, which has high concentrations of lycopene, therefore, has been thoroughly explored. (Strati and Oreopoulou, 2014).

The objective of this research is to extract lycopene from tomato peels and identify lycopene using HPLC. Stability was studied at different pH values, temperature and light extracted from lycopene. Also, an evaluation of the antioxidant activity of lycopene extracted from tomato peel on sunflower oil was performed, and a sensory evaluation of the prepared beef burger was performed using different levels of extracted lycopene.

MATERIALS AND METHODS

Raw materials: Tomato peels were obtained from Kaha Company for preservative food Kaha, Kalyobia governorate, Egypt (2017). Most chemicals (analytical grade) were purchased from El-gomhouria pharmaceuticals Co., Cairo, Egypt. 2.2-diphenyl-1-1picryl-hydrazyl (DPPH), Folin-Ciocalteu reagent, gallic acid, quercitin and HPLC grade solvents were obtained from Sigma-Aldrich Chime, Steinheim, Germany. Refined sunflower oil (antioxidant free) was purchased from Misr Company for Oil and Soap, El Mansoura, Egypt.

Preparation of tomato peels: Tomato peels were grinded and kept individually in polyethylene bags and stored in refrigerator at 5 ± 1° C until used.

Preparation of beef burgers: The control beef burger formula consisted of 60% meat, 7.10% fat, 6% water, 12% rehydrated texturized soy (1g soy: 2ml water), 5.50% fresh egg, 5.0 % fresh onion, 1.4% ground bread crust,1.5% salt and 1.5% spices according to the method described by Heinz and Hautzinger (2007). Finally, the lycopene extract (0 %, 0.5%, 1.0% and 1.5 %) were added, and there was one sample prepared as control with an artificial color (0.1). All formulations were aerobically packaged in a foam plates, wrapped with polyethylene film and stored at -18°C for 3 months. The samples were successively evaluated every week.

Gross chemical composition and minerals content: They were determined according to the methods described in the A.O.A.C. (2012). Total carbohydrates were calculated by differences according to Howard and Leonard (1963). Minerals content was determined after dry ashing according to the method described in the A.O.A.C. (2012), using atomic absorption amanitas (Perkin – Elmer, Model 3300, USA).

Determination of total phenolic compounds: It was determined using Folin-Ciocalteu reagent according to the method described by Maurya and Singh (2010).

Determination of total flavonoids: It was determined according to the method described by Jia *et al*. (1999).

Determination of antioxidant activity: The antioxidant activity of samples was determined by the 2, 2'-Diphenyl-1- picrylhydrazyl (DPPH) radical scavenging activity according to the colorimetric method of Brand-Williams *et al*. (1995). The percentage inhibition of the DPPH radical by the samples was calculated according to the formula of Yen and Duh (1994)

$$\text{Inhibition\%} = [\text{Ac (0)} - \text{AA (t)}] / \text{Ac (0)} \times 100$$

Where: **Ac (0) is the absorbance of the control at time = 0 min. AA (t) is the absorbance of the antioxidant at time =1h**

Extraction and determination of lycopene:

Lycopene was extracted and determined from tomato peels according to the method described by Ranganna (1977). The lycopene content of the sample was calculated using the relationship that an optical density (OD) of 1.0 = 3.120 µg of lycopene per ml.

$$L = \frac{3.1206 x O.D\ of\ Sx Vx Dil x 100}{1 x Wt\ of\ Sx 1000}$$

Where: **L = Lycopene (mg/100gm), S = sample, V = volume made up, Dil = dilution and Wt = weight of sample.**

Identification and quantification of lycopene: The carotenoids (lyco-red) extracted from tomato peel were identified by Knauer HPLC pump 64 according to the method reported by (Gaylek *et al*., 1987).

Stability of natural lycopene pigment extracts:

Effect of pH, A preliminary study was conducted to test the stability of lycopene extract at different pH ranged from 2.0 to 10 for 30 min and then percentage of color loss was calculated.

Effect of temperature, the stability of lycopene extracted from tomato peels against pH value, temperature and time exposed to heat was determined according to the method described by Rizk *et al*. (2014). Different media with pH value ranged from 2 to 10 at room temperature were used. To investigate the effect of heating on stability of lycopene, different temperature ranged from 30 to 100 °C for 30 min at pH 8, 10 were used.

Effect of light, A preliminary study was conducted to test the stability of lycopene extract of tomato peels the effect of exposure at dark and light condition on the stability of lycopene pigment was measured every hour for six hours under different weather conditions.

Rancimat method, the antioxidant activity was determined by the Rancimat method using Rancimat Metrohm 679 as described by Hasenhuttle and Wan (1992) and the induction period (I.P.) was conducted with Rancimat at 110ºC and calculated at 25±1ºC using the temperature coefficient of 2.2 for induction period according to the method reported by Hadorn and Zurcher (1974) and 2.5 for expired period according to the method reported by Pardun and Kroll (1972) . The antioxidant activity and increasing index were calculated using the following equation:

$$\text{Antioxidant activity} = \frac{IP\ of\ Sx100}{IP\ of\ C}$$

$$\text{Increasing index} = \frac{IP\ of\ S - IP\ of\ C}{IP\ of\ Cx100}$$

Where: ***IP of S* = induction period of sample and *IP of C* = induction period of control.**

Effect of storage period at – 18°C on natural pigment stability from tomato peels

The stability of lycopene in the beef burger with lycopene was studied during storage period at -18 C for 12 weeks according to the method described by (Ranganna, 1977).

Color measurements: The color was determined at three different points of the sample with a Chroma meter (Minolta CR 400, Minolta Camera, Co., Osaka, Japan). Corresponding L* value (lightness of color from zero (black) to 100 (white); a* value (degree of redness (0–60) or greenness (0 to -60); and b* values (yellowness (0–60) or blueness (0 to - 60) were measured for all the samples were calculated according to (Abonyi *et al.*, 2002) by the following equation:

$$L^* = \sqrt{a^2 + b^2}$$

Sensory Evaluation: Sensory attributes (color, taste, odor, texture and overall palatability) of beef burger was according to the method of Lindley *et al*. (1993).

Statistical Analysis: The statistical analysis was carried out using one-way analysis of variance (ANOVA) under significant level of 0.05 for the whole results using the statistical program CoStat (Ver. 6.400) according to Steel *et al*. (1997). To ascertain the significant among means of different samples, LSD test was applied.

RESULTS AND DISCUSSION

Chemical composition of tomato peels.

The chemical composition of tomato peels is listed in Table (1). The obtained results show that moisture, ash content and crude protein of dried tomato peels were 6.57,

4.48 and 11.67%; respectively. Meanwhile, tomato peels contained the highest value of crude fiber and carbohydrates (37.36 and 41.44% respectively). However, it contains less ash. The results obtained are consistent with those recorded El-Seesy and Hamed (1998), Elbadrawy and Sello (2016) and Ammar and Aboalfa (2017) who recorded that the protein content of tomato was 10.50 %, fat 4.04%, moisture 4.76 % and crude fiber 53.27 %, whereas, ash content ranged from 4.09 to 5.90%, and 27.29 to 78.56% for total carbohydrates, respectively on dry weight. These variations may be attributed to effect of the cultural practices in raw materials production environment, the differences of raw materials cultivars or processing.

Table 1. Gross chemical composition of tomato peels

chemical composition	Tomato peels
Moisture	6.57
Ash*	4.48
Crude protein*	11.67
Crude Fat*	5.05
Crude fiber*	37.36
Carbohydrates°°*	41.44

*** Calculated on dry weight basis.**
°° carbohydrate calculated by difference.

Minerals content of tomato peels wastes.

The data in Table (2) showed that tomato peels content of the minerals. Calcium, nitrogen, phosphorus, potassium, sodium, manganese, copper, zinc and iron were detected. The results also showed that the potassium and calcium content of tomato peels was larger amount (1660 and 1140 mg / 100 g) followed by nitrogen (260 mg / 100 g). At the same time, sodium and phosphorus content was found in moderate amounts (180 and 120 mg / 100 g). In addition, it contained the lowest amount of iron, copper, zinc and manganese. These results agree to a less extent with that of Gonzalez *et al*. (2011) who reported that the minerals content of tomato peels was 140.5 mg/100g for Ca, 157.33 mg/100g for Mg, 78.33 mg/100g for Na, 1125.0 mg/100g for K, 1.68 mg/100g for Fe and 0.53 mg/100g for Zn, respectively. The mineral content of plants can be significantly influenced by variety, location and environmental conditions (Rao, 1996).

Bioactive compounds of tomato peels.

Some bioactive compounds namely, polyphenols, flavonoids contents and antioxidant activity, as well as, lycopene content were determined, and the results are presented in Table (3). Date showed that total phenolic compounds and total flavonoids contents were 3.75 and 0.97 mg/g. As well as, antioxidant activity of tomato peels was 38.31%. These results are in close agreement with those reported by Ammar and Aboalfa (2017) who found that the level of total phenolic was 2.40 mg/g on dry weight basis. On the other hand, tomato peels are considered as a good source for lycopene which amounted to 3.75 mg/100g on dry weight basis.

Identification of lycopene for tomato peels by HPLC.

Method of HPLC has been widely applied to investigate the compositions of lycopene. Lycopene pigments extracted from tomato peels were separated and identified by HPLC are showed that in Figure (1) and Table (4). Spectral measurement and HPLC separation indicated that there are two major lycopene components for tomato peels namely lycopene which represented to be 2.282µg /ml, meanwhile, the other one was β- carotene 1.801µg/ml. The results obtained are consistent with those recorded Rizk *et al*. (2014) They showed that lycopene was the predominant pigment in tomato peels (86.13%), while phytoene and β –carotene was concentrated.

Table 2. Minerals content (mg/100g) of tomato peels *:

Sample	Minerals (mg/100g)								
	Ca	N	P	K	Na	Mn	Cu	Zn	Fe
Tomato peels	1140	260	120	1660	180	0.1	4.1	2.17	11.1

*** calculated on dry weight basis.**

Table 3. Bioactive compounds and lycopene content of tomato peels

Sample	Constituents			
	Total phenolic compounds as Gallic acid mg/g	Total flavonoids (mg/g)	Antioxidant activity % as DPPH	Lycopene mg/100g
Tomato peels	3.75	0.97	38.31	3.75

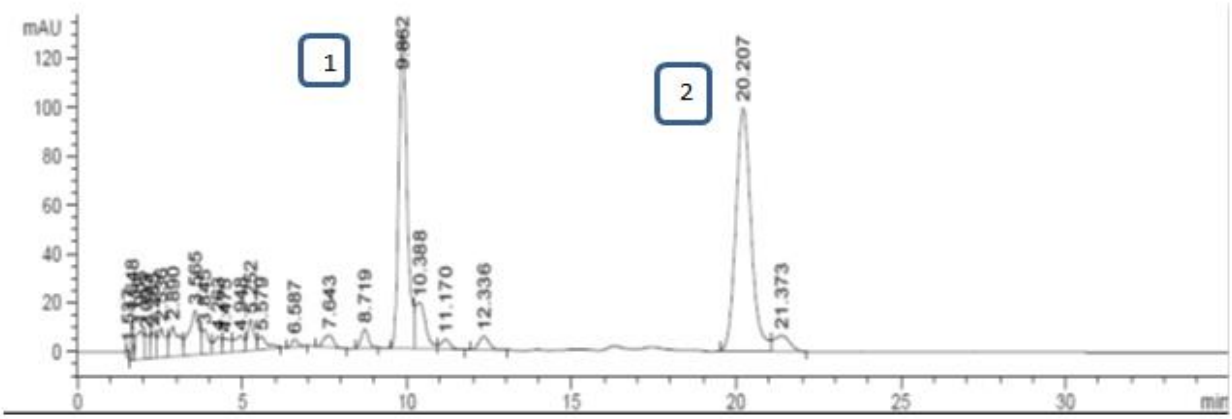

Figure 1. Identification of lycopene pigment compounds extracted from tomato peels.
1= Lycopene 2=β-carotene

Table 4. Identification of lycopene from tomato peels by HPLC

Std.	Rt	Inj	Area Std.	Area Sp.	µg /ml
Lycopene	9.8	3.33	2294.391	39.26041	2.282
β- carotene	20.2	1.67	3214.668	86.83062	1.801

Area Std :Area Stander Area Sp: Area sample Inj: injection volume Rt: Retention time

Effect of pH on retention of lycopene pigment extracted from tomato peels.

The effect of different pH values on retention of lycopene pigments derived from tomato peels was determined using the pH-differential method at nine different pH ranged from 2 to 10 at room temperature and the results are presented that in Table (5). The highest O.D value at 470 nm. observed at pH 9 and pH 10 and pH3 were 1.120 ,1.120 and 0.840, respectively) while, at pH 2 recorded the lowest O.D (0.800). As well as, decreasing of pH values from 10 to 7 caused 8.48% little degradation of lycopene content and the degradation of color was not significant till pH 6. Meanwhile, the degradation of color reached to 25.00 and 28.57 % at pH 3and pH 2, respectively. So, it could be observed that the stability of lycopene pigment was more pronounced at acidic pH values (7 to 10). While, the highest degradation was occurred at pH 2. These results are in agreement with those reported by Rizk *et al*. (2014) Which found that the highest levels of stability and the lowest levels of carotenoids degradation (Lyco red) extracted from the tomato peels were in alkaline media from 7.0 to 10.00 pH. Therefore, the lycopene extracted from tomato peels was more unstable in the acid media and more fixed at alkaline media. These results may be due to the characteristic double bonding system associated with carotenoids. The major work-related problem is the instability of carotenoids, especially toward light, heat, oxygen and acids (Oliver and Palou, 2000).

Effect of different temperature degrees on lycopene separated pigment stability under different pH

Table (6) represents the rate of degradation% and remained% of lycopene extracted from tomato peels. The high stability was observed as a result of exposing lycopene at low and moderate temperature between zero °C and 50°C, while at above 60°C, the degradation of lycopene increased gradually by increasing the temperature to 100 °C which reached to 13.24% at pH 8 and 11.87% at pH10. As well as, the remained of lycopene decreased with raising the temperature to 100 °C which reached to 86.76 % at pH 8 and 88.13% at pH10. These results are in agreement with those reported by Hamed *et al*. (2000) and Rizk *et al*. (2008) who mentioned that the lycopene is somewhat sensitive to high temperature and have a good stability at 50° C. For instance, lycopene degradation, caused by exposure to higher heat, may be one of the main causes of lycopene degradation during isomer and oxidation therapy. Isomerization converts all-trans isomers to cis-isomers Due to the additional energy input and output at an unstable level is rich in energy. The thermal processing generally causes some loss of lycopene -based foods. Heat induces isomerization of the all trans to cis forms. As the temperature increases and processing time increases the cis-isomers (Shi and Le Maguer, 2000).

Table 5. Effect of pH on retention of lycopene pigment extracted from tomato peels.

pH	Lycopene ʎ=470 nm	Remained of Lycopene %	Degradation of Lycopene %
2	0.800	71.43	28.57
3	0.840	75.00	25.00
4	0.900	80.36	19.64
5	0.960	85.72	14.28
6	0.990	88.39	11.61
7	1.025	91.52	8.48
8	1.065	95.09	4.91
9	1.120	100.00	0.00
10	1.120	100.00	0.00

Table 6. Effect of different temperature degrees on lycopene separated pigment stability under different pH:

C°/ 30min	pH 8			pH 10		
	Lycopene ʎ=470 nm	R*	D**	Lycopene ʎ=470 nm	R*	D**
Zero	1.585^{a}±0.005	100.00	0.00	1.600^{a}±0.019	100.00	0.00
30 °C	1.565^{a}±0.013	98.74	1.26	1.585^{a}±0.007	99.07	0.93
40 °C	1.540^{b}±0.008	97.16	2.84	1.565^{b}±0.005	97.81	2.19
50 °C	1.520^{b}±0.014	95.9	4.10	1.540^{c}±0.009	96.25	3.75
60 °C	1.495^{c}±0.022	94.32	5.68	1.515^{d}±0.07	94.69	5.31
70 °C	1.480^{c}±0.017	93.38	6.62	1.500^{d}±0.006	93.75	6.25
80 °C	1.440^{d}±0.008	90.85	9.15	1.470^{e}±0.004	91.87	8.13
90 °C	1.410^{e}±0.007	88.96	11.04	1.435^{f}±0.006	89.69	10.31
100°C	1.375^{f}±0.008	86.76	13.24	1.410^{g}±0.007	88.13	11.87
LSD	0.021			0.015		

(R*): Remained%. (D): Degradation %. LSD ≤ 0.05**

Effect of light on the stability or degradation of natural lycopene.

Light is one of the factors that affect the stability or degradation of lycopene. The effect of exposure at dark and light condition on the stability of lycopene pigment was measured everyone hour for six hours and the results are presented in Table (7). Data in Table (7) reveal that the samples which were exposed to light showed more reduction in the absorbance value compared with the samples kept in the dark. As well as, the remained of lycopene (94.36%) was higher in the dark than the light (93.15%). comparatively, lycopene degradation under dark condition is very low than degradation under light which reached to 5.64 % compared with light condition (6.85%). These results are consistent with Salama *et al*. (2015)

found that storage for 4 and 8 hours in the dark had almost no effect on the retention rate of the extracted lycopene.

The results showed that the reduction in percent retention was increased by increasing the time of the extracted lycopene to daylight for 4 to 8 h. Lycopene retention was observed after 8 hours in sunlight by 78.6%. However, this lycopene is lost when exposed to daylight. Thus, when storing lycopene and the food containing it, it should be stored to prevent degradation and loss in dark conditions.

Table 7. Effect of light on stability of natural lycopene pigment.

Time (Hour)	Dark			Light		
	Lycopene ʎ=470 nm	R*	D**	Lycopene ʎ=470 nm	R*	D**
Zero	1.24^{a}±0.011	100.00	0.00	1.24^{a}±0.011	100.00	0.00
1	1.23^{ab}±0.007	99.19	0.81	1.22^{b}±0.009	98.39	1.61
2	1.215^{bc}±0.007	97.99	2.01	1.195^{c}±0.011	96.83	3.62
3	1.200^{cd}±0.005	96.78	3.22	1.185^{c}±0.009	95.57	4.43
4	1.19^{de}±0.013	95.97	4.03	1.175^{cd}±0.011	94.76	5.24
5	1.185^{de}±0.006	95.57	4.43	1.160^{d}±0.004	93.55	6.45
6	1.170^{e}±0.016	94.36	5.64	1.155^{d}±0.013	93.15	6.85
LSD	0.018			0.017		

(R*): Remained%. (D): Degradation %. LSD ≤ 0.05**

Effect of crude lycopene extract addition on oxidative stability of sunflower oil

Different levels of crude lycopene extracted from tomato peels were added to sunflower oil and its oxidative stability and the induction period were compared with those of BHT (200 ppm). Data in Table (8) showed that the effect of crude lycopene extract addition with levels 100, 200 and 300 ppm on the oxidative stability of sunflower oil. The addition of BHT resulted in the highest induction period (5.05 hrs.) and increased expired period at 25ºC to 2.80 months. On the other hand, crude lycopene extracted as pro-oxidant in higher concentrations (200 and 300 ppm, respectively), which had induction periods 4.55 and 6.60 hrs., respectively compared with the lower concentration (100 ppm). Also, the levels of crude lycopene crude extract 100, 200 and 300 ppm had an antioxidant activity were 0.96, 0.98 and 0.99, respectively, comparison with BHT (1.09) (Table 8). These results are consistent with those reported Zakaria (2006), who found that lycopene concentration (200 ppm) have induction period of 4.865 hrs. while 300 ppm had 6.63 hrs., Therefore, there is an adverse relationship between concentration and induction period when lycopene added to sunflower oil. The potential reduction of the antioxidant property of lycopene is associated with the formation of a radical superoxide anion (Palozza, 1998). Lycopene also allows the formation of peroxyl radicals that are able to act as pro-oxidants and subject to the same auto-oxidation (Shi *et al*., 2002).

Table 8. Effect of crude lycopene extract addition on oxidative stability of sunflower oil.

Extracts Levels (ppm)	Oxidative stability				
	Added Weight (ppm)	Induction period at 100ºC (hours)	Expired period at 25ºC (months)	Antioxidant Activity %	Increasing Index %
Control	---	4.63	5.65	100	0.00
BHT	200	5.05	6.16	1.09	9.07
	100	4.45	5.43	0.96	-
Tomato extract	200	4.55	5.55	0.98	-
	300	4.60	5.61	0.99	-

Sensory evaluation of beef burger with natural lycopene pigment

Beef burgers was prepared with artificial color (0.1 %) and different levels of natural lycopene pigment (0, 0.5, 1.0 and 1.5 %). These treatments were sensory evaluated was assessed for color, taste, odor, texture and palatability in general. Data were analyzed statistically, and these results are clearly shown in Table (9). From Table (9), it could be clear observed that addition of natural lycopene pigment with 1.0% (T4) was more acceptable and scored the highest score for all parameters compared with other treatments. Meanwhile, the lowest score was given to T2 without addition. These results are consistent with those reported García *et al*. (2009) who found that, the addition of tomato peels as source of lycopene and fiber with 4.5% in beef hamburgers gave the highest score for overall acceptability after control sample without addition. Finally, of data can clearly conclude that addition of natural lycopene pigment with 1.0% (T4) had holds the highest score and was favored by the consumer. Therefore, this treatment was choose being the most favorite to continue the study. Selected one was stored under freezing temperature (-18 ±1 °C) then, the effect of storage on color intensity was studied every week.

Table 9. Mean organoleptic scores* for beef burger with natural lycopene pigment.

T*	Properties					
	Color	Taste	Odor	Texture	Palatability	Over all
T1	7.88^{a}±1.55	7.31^{a}±1.44	7.38^{a}±1.51	7.56^{a}±1.50	7.81^{a}±1.07	37.94^{a}±5.78
T2	7.19^{a}±1.49	7.31^{a}±1.27	7.00^{a}±1.13	7.00^{a}±1.69	7.31^{b}±1.49	35.81^{b}±5.25
T3	7.69^{a}±0.83	7.63^{a}±0.78	7.19^{a}±1.05	7.56^{a}±1.28	7.50^{a}±1.55	37.56^{a}±5.21
T4	8.25^{a}±0.70	8.06^{a}±0.74	7.56^{a}±0.86	7.81^{a}±0.62	8.00^{a}±0.76	39.69^{a}±2.55
T5	7.44^{a}±0.89	7.31^{a}±0.92	7.19^{a}±0.65	7.19^{a}±0.98	7.30^{a}±1.07	36.44^{a}±3.36

Values are mean ±SD of three replicates. Different letters in each column show significant difference. All statistical analysis was performed at P≤ 0.05. T*= treatments, scores according to a 10 point by 10 panelists. T1: (Control) beef burger colored with artificial color 0.1 %. T2: beef burger without addition. T3, T4 and T5: beef burger enhanced with 0.5%, 1% and 1.5%, respectively of natural lycopene pigment.

Color measurements of beef burger.

The natural lycopene pigment (lycopene from tomato peels) were used to enhancement of beef burger color and the results in Table (10) show that the lightness values (L*) were significantly differed between all beef burger treatments obtained using lycopene pigment in different concentrations 0, 0.5, 1.0 and 1.5% of lycopene pigment in addition of artificial color 0.1% as control sample . At first, storage period or at zero-time lightness (L*) of beef burger with lycopene pigment ranged from 45.01 to 48.31, in comparison with beef burger with artificial color which had (L*) values of (42.63). Meanwhile, the treatment (T_2) without any addition recorded 50.53. Beef burger with lycopene pigment displayed lighter less than beef burger without lycopene pigment. All beef burgers with lycopene pigment had low L* value while, the beef burger without lycopene pigment appeared to have high (L*) value. It seems that the addition of lycopene pigment, throw producing of beef burger has less enhancement and decreased the (L*) values.

On the other side, the color is darker than the beef burger with lycopene pigment can be attributed to color and the processing temperature. Meanwhile, the results in Table (10) after three months of storage, show that beef burger with lycopene pigment had a significant high in (L*) values compared to control sample. The value averaged from 41.32 to 43.32 in beef burger with lycopene pigment. While, a dramatic reduction in (L*) values (45.12) due to the lack of lycopene pigment. These results may be due to the storage temperature degrees which effect on the stability of lycopene pigment. The obtained results in Table (10) show that the beef burger with lycopene pigment had a significant difference in red value (a*). The values at zero-time averaged from 9.21 to 10.81 in beef burger with lycopene pigment, however, (T_1) with artificial color presented to be (13.88). On the other hand, a dramatic reduction in (a*) values (6.79) as a result of the absence of natural red pigments were observed. In fact, red value (a*) decreased when the (lycopene pigment) concentrations were decreased. Moreover, the obtained results after three months of storage had the same trend, and these results might be due to addition of lycopene pigment.

The dominant color for beef burger is red and thus, better representation can be provided by Hunter color b * (yellowing) to distinguish the color difference in meat burger due to its effect. the addition of lycopene pigment. Highly significant differences had been observed in the value of b * (yellowing) between beef burger with and without addition of natural red color. Chroma value or vitality in the red color of beef burger considerably affected by lycopene pigment addition and its ratio as well as storage period. Similarly, the chroma values of beef burger with addition of lycopene pigment treatments were observed in the same trends of a more vivid red color than samples of beef burger without addition of lycopene pigment which recorded (22.47) Table (10).

The hue angle values for beef burger with natural red color treatments at zero-time (73.59 -75.61) were lower than those without natural red color treatments (58.87), however, their Chroma values are very high indicating a dull color, Table (10). The overall distinct, vivid red color of the beef burger might be due to of lycopene content. The minimal color change of beef burger product produced by (lycopene pigment) suggests the appropriateness of these processes to produce high quality and palatable products. These results are consistent with those reported by García *et al.* (2009). This may be because the red pigment lycopene and its addition to the meat can turn the color towards orange, which may explain the increase in a* and b* parameters in batches containing lycopene extract, in the case of both cooked and raw beef burgers.

Table 10. Color measurements of beef burger

Treatments	Properties				
	L*	a*	b*	Chroma	Hue
	Zero time				
T1	42.63	13.88	20.41	26.84	58.87
T2	50.53	6.79	21.41	22.47	72.40
T3	48.31	9.21	31.29	32.62	73.59
T4	47.61	10.02	32.34	33.86	75.61
T5	45.01	10.81	33.62	35.32	73.61
	After three months				
T1	40.83	13.02	18.98	24.21	57.46
T2	45.12	6.45	17.06	18.24	69.29
T3	43.32	6.84	28.83	29.63	67.25
T4	42.70	7.05	27.48	28.37	72.79
T5	41.32	9.32	31.69	33.03	72.17

L*: degree of lightness a*: degree of redness b*: degree of yellowness T1: (Control) beef burger colored with artificial color 0.1 %. T2: beef burger without addition. T3, T4 and T5: beef burger enhanced with 0.5%, 1% and 1.5%, respectively of natural lycopene pigment.

Effect of storage periods at -18 °C on stability of lycopene pigment in beef burger:

Meat and meat products tend to change in color by storage. Stability of added lycopene pigment in beef burger with 1.0% during storage at – 18 °C were studied every week for 12 weeks and the results are shown in Table (11). Data reveal that the amount and remained of lycopene pigment decreased from 0.470 to 0.416 mg/100g and 100 to 88.51% during storage period and it considered not significant differences. Meanwhile, lycopene degradation rate increased from 0.0 to 11.49%. These results are disagreement with Doménech-Asensi *et al.* (2013).

Table 11. Effect of storage periods at -18 °C on stability of lycopene pigment in beef burger (per weeks)

Storage periods	Properties		
	Amount (mg/100g)	Remained of pigment %	Degradation rate of pigment %
Zero	0.470	100.00	0.00
1	0.468	99.58	0.42
2	0.465	98.94	1.06
3	0.460	97.87	2.13
4	0.458	97.45	2.55
5	0.454	96.60	3.40
6	0.450	95.75	4.25
7	0.446	94.89	5.11
8	0.438	93.20	6.80
9	0.430	91.49	8.51
10	0.426	90.64	9.36
11	0.420	89.36	10.64
12	0.416	88.51	11.49

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استخلاص واستخدام ليكوبين قشور الطماطم كمضاد للأكسدة وكملون طبيعي في البيف برجر

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ينتج عن عملية تصنيع الطماطم كميات كبيرة من المخلفات ، وخاصة قشور الطماطم التي تخلق مشاكل بيئية. هذه المخلفات هي مصادر جذابة للصبغات والمركبات النشطة بيولوجيا. الليكوبين هو واحد من الكاروتينات ، وهو صبغة قابلة للذوبان في الدهون لها خصائص مضادة للأكسدة. أجريت هذه الدراسة لتقييم ثبات اللايكوبين وخصائصه المضادة للأكسدة وتأثيره على هذه المعاملات. تمت دراسة درجة ثبات اللايكوبين أيضًا أثناء التخزين. بعد تقييم مركبات اللايكوبين الكلية وتفريدها بواسطة HPLC. عندما تم فصل الألوان في قشور الطماطم، وجد أنها تحتوي على عنصرين رئيسيين، اللايكوبين والبيتا كاروتين. وكان إجمالي المركبات الفينولية ومحتويات الفلافونويد الكلية ٣.٧٥ و ٠.٩٧ ملجم / جرام ، وكان محتوى اللايكوبين ٣.٧٥ ملجم / ١٠٠ جم. كذلك، كان نشاط مضادات الأكسدة لقشور الطماطم ٣٨.٣١ ٪ (كمادة الـ DPPH) . في بعض معاملات اللايكوبين لوحظ أن معدل التدهور زاد بزيادة درجة pH ودرجة الحرارة والضوء بزيادة وقت التعرض وطول مدة التخزين. كانت نسبة نشاط مضادات الأكسدة في اللايكوبين قريبة من مادة الـ BHT الصناعية المعروفة. حيث كانت مستويات استخراج اللايكوبين الخام ١٠٠ و ٢٠٠ و ٣٠٠ جزء في المليون لها نشاط مضاد للأكسدة وهي ٠.٩٦ و ٠.٩٨ و ٠.٩٩ على التوالي. مقارنة مع تلك المادة وهى BHT (1.09). في التقييم الحسي ، كان أفضل نسبة من اللايكوبين الطبيعي المضاف هى ١ ٪ كانت المفضلة من قبل المستهلك. وبتخزين البيف برجر البقرى ١٢ أسبوعًا، انخفضت كمية وبقايا صبغة اللايكوبين من ٠.٤٧٠ إلى ٠.٤١٦ ملجم/ ١٠٠ جم، او من ١٠٠ إلى ٨٨.٥١٪ خلال فترة التخزين. وفي الوقت نفسه ، ارتفع معدل تدهور اللايكوبين من ٠.٠ إلى ١١.٤٩ ٪.