Physiochemical Assay of Lycopene Supplemented Yoghurt

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Abstract:

Tomato (*Lycopersicumesculentum* L.) is one of the predominant lycopene source. Lycopene is an essential nutrient as it is not synthesized internally. It is a carotenoid abundantly found in tomato and tomato. It is found in red vegetables and fruits such as pink grapefruits, watermelons, tomatoes and papayas. The uptake of lycopene-rich foods can result in positive health benefits.

Objective:

To perform physiochemical assay of Lycopene supplemented yoghurt.

Methods:

Various organic solvents (i.e. hexane) were used to extract lycopene and to used in yoghurt preparation. Hexane residues were removed from the extract before being used for the product development. Chemical analysis, acidity, and viscosity and sensory analysis were carried out during storage period at intervals of 0, 7 and 14 days.

Results:

Acidity was increased due to the production of lactic and acetic acid. Acidity was increased significantly from 0.92 to 1.03%. Yoghurt was evaluated for different sensory attributes such as taste and mouth feel. A decline trend in the scores for sensory characteristics was observed during 14 days of storage period. At zero day maximum score (9) was recorded for T_3 and minimum score for (8.26) was awarded to T_0. After 0, 7 and 14 days of storage, judges observed a slightly decline due to the storage time of the yoghurt samples.

Conclusions:

The present analysis suggested that lycopene has a great antioxidant potential, which could be utilized in dairy products to increase quality and shelf life.

Key words:

Antioxidant, lycopene, yoghurt, chemical analysis, taste and mouth feel.

Introduction:

Milk and different dairy products are essential for human health due to their higher content of protein, vitamin and minerals particularly calcium, potassium, magnesium and phosphorous. These dairy products can be made more nutritious by addition of various complementary ingredients like multivitamins, minerals, fruit pulp, chocolate, and flavor and by adding spices of cinnamon, ginger, clove and different fruits and vegetable extracts. New born babies adapt themselves with the help of various bioactive and nutrient components of milk that stimulate and mature the digestive system and cellular growth that is most desirable for postnatal adaptation. Fermented milk is widely produced in many countries.[1] Yoghurt is an important food ingredient in most of societies. It is believed that consumption of yoghurt and other dairy products is very beneficial for health. The nutrient value of curd or yoghurt depends on the milk composition and substances added to it during manufacturing. Yoghurt can be manufactured from skimmed or whole milk and it can be sweetened, plain or flavored with fruit juices, cane sugar etc.[2] The bioactive ingredients are diverse in nature like phenolics and carotenoids showing considerable anti oxidative activity.[3, 4] Lycopene is an essential nutrient as it is not synthesized internally, and therefore must

be provided from food source which contain lycopene.[5] Lycopene, a carotenoid abundantly found in tomato as well as tomato derived products and its gives color to tomato. Different proceeding explored the various properties of carotenoids such as physicochemical and biological characteristics that make it natural antioxidants and coloring compounds, because it contain eighty to ninety percent of the total pigment content.[6] Lycopene is a main pigment compound of tomato with strong natural antioxidant especially against reactive species (O2 singlet) that promotes the cell injury as well senescence. The main source of lycopene in human diet are tomato and tomato based different food products. It is also a most dominant carotenoids present in human serum in America and represents the 50% of all carotenoids content of plasma.[7] Numerous clinical studies indicated the relationship between the consumption of carotenoids and degenerative diseases like cancer, macular degeneration and cardiovascular diseases, persons with higher intake of carotenoids are at lower risk of degenerative diseases. [8]

According to Baron JA *et al*., it has been proved in epidemiological studies that patients who did not drink or smoke, beta- carotene supplementation bring a 44% reduction in possibility of colorectal adenoma recurrence.[9] Hasler CM proved that among vegetables and fruits, tomato is gaining attention of the scientists due to the existence of potent anti-oxidant i.e. lycopene. Consequently, vegetables and fruits derived nutraceutical are important significance to curtail various physical threats via distinct pathways.[10] Sahan N *et al*., examined the quality i.e. physical, chemical and flavor of non-fat yoghurt that is affected by β-glucan hydro colloidal complex when it is storage. They also compared yoghurts with added β-glucan with non-fat yoghurt without the addition of β-glucan. They revealed that protein and fat contents were the same of the trial yoghurts, while ash contents were differed. Volatile fatty acids, titratable acidity, acetaldehyde, pH and tyrosine contents had not showed any significant change by the addition of β-glucan composite at any time of storage.

Tyrosine contents and titratable acidity improved radically during storage. Addition of β-glucan composite did not influence water-holding capacity and gel firmness in the yoghurts, although these variables decreased with storage time. Decrease in whey separation and increase in viscosity caused by the addition of β-glucan composite. They concluded that during sensory evaluation control yoghurts were more favored; however, in the production of non-fat yoghurt use of β-glucan composite at low levels gave acceptable sensory scores.[11] Teles CD and Flores SH deliberated the effects of addition of additives i.e. stabilizers and skim milk powder on the sensory parameters and apparent viscosity of nonfat yoghurt. In their study, they applied the response surface methodology. On sensory parameters significant effects was shown by the addition of stabilizers. It was also observed that overall acceptability of yoghurt decreased when higher concentration of guar gum and xanthan whereas higher consistency and apparent viscosity produced with the gelatin. The relationship between consistency and apparent viscosity which is a sensory evaluation and an instrumental measurement respectively was 0.72.[12]

The aim of study is to develop the youghurt with addition of lycopene for the improvement of its healthy benefits.

Methods:

Mature red colored tomatoes were procured from the local market. Fresh buffalo milk for yoghurt preparation was obtained from dairy farm. Peel and seeds were removed manually and dried. After drying, the tomato skin and seeds were ground using coffee grinder and sieved through 1.05 ± 0.10 mm for further analysis.[13] 25g of tomato peel and seeds powder

was added in conical flask along with 150 mL of solvent solution and placed for agitation in orbital shaker for 4 hours at 250 rpm. After agitation, cold double distilled water was added. The resultant solution was separated by using rotary evaporator at 37°C.[14] Milk was cooled to a temperature of 40-45°C and inoculated with 2.5% starter culture. Inoculated milk was poured in cups of 250 mL volume and labeled.Lycopene was added at concentrations of 0% (T_0), 0.25% (T_1),0.5% (T_2), 0.75% (T_3) and 1.0% (T_4) in milk.The inoculated milk was incubated at 42°C for 3 hours. The yoghurt was then cooled to a temperature of 4-6°C and was subjected to physicochemical and sensory evaluation. Acidity was determined by following the method (942-15). Direct titration method was used to determine the acidity of yoghurt samples.[15] Protein was determined through Kjeldaltech Apparatus (Model: D-40599, Behr Labor Technik, Gmbh-Germany) following the method (984-13).[15] The crude fat content in samples was determined following the method (920-39).[15] Viscosity of yoghurt samples was measured by following the method. Viscosity of the yoghurt samples was measured at 4°C with a spindle (No. 4) rotation of 60 rpm using a Brookfield viscometer (model LVDVE 230).[15]The yoghurt prepared from with different levels 0%, 0.25%, 0.5%, 0.75% and 1.0% of antioxidant extract (lycopene) was evaluated by a panel of judges for sensory evalution of yoghurt according to the procedure. The data generated during the experiments were analyzed by statistically available software Steel RG. *et al,*.[16]

Results:

The results indicated that the treatment showed non-significant effect on the acidity of yoghurt while storage days exhibited significant effect on the acidity of yoghurt. The interaction of treatment and storage time was found to be non-significant because the addition of lycopene do not have any effect on the acidity of yoghurt. The results indicated that total titratable acidity ranged from 0.92 to 1.03 among the yoghurt prepared by using different levels of lycopene during storage as shown in Table 1.

Treatments	Storage (days)			Means
	0	7	14	
T^0	0.93	0.99	1.0	0.97 ± 0.03
T^1	0.92	0.98	1.0	0.96 ± 0.03
T^2	0.92	0.99	1.01	0.97 ± 0.04
T^3	0.93	1.0	1.03	0.99 ± 0.05
T^4	0.94	0.98	1.02	0.97 ± 0.04
Means	0.92	0.99	1.01	

Table 1: Effect of lycopene on the acidity (%) of yoghurt

T_0 = yoghurt with lycopene 0%; T_1 = yoghurt with lycopene 0.25%; T_2= yoghurt with lycopene 0.5%; T_3 = yoghurt with lycopene 0.75%; T_4 = yoghurt with lycopene 1.0%

The results indicated that the treatment showed non-significant effect on the protein of yoghurt while storage days exhibited significant effect of the protein of yoghurt. The interaction of treatments and storage time was found to be non-significant for the protein of yoghurt. The results indicated that protein of the yoghurt ranged from 4.15 to 4.3 among the yoghurt prepared by using different levels of lycopene during storage (Table 2).

Treatments	Storage (days)			Means
	0	7	14	
T^0	4.27	4.23	4.15	4.21 ± 0.06
T^1	4.28	4.23	4.16	4.22 ± 0.06
T^2	4.30	4.24	4.16	4.23 ± 0.07
T^3	4.29	4.24	4.17	4.22 ± 0.06
T^4	4.29	4.25	4.16	4.22 ± 0.06
Means	4.286	4.238	4.16	

Table 2: Effect of lycopene on the protein of yoghurt during storage

T_0 = yoghurt with lycopene 0%; T_1 = yoghurt with lycopene 0.25%; T_2= yoghurt with lycopene 0.5%; T_3= yoghurt with lycopene 0.75%; T_4 = yoghurt

with lycopene 1.0

The results indicated that fat of the yoghurt ranged from 4.90 to 5.10 among the yoghurt prepared by using different levels of lycopene during storage (Table 3). It is observable that fat vary significantly among the yoghurt prepared by using different concentration of lycopene. The storage also showed significant effect on the fat exhibiting a decreasing trend as storage progressed. Minimum fat was recorded after 14 days of storage while maximum fat was observed at 0 day of storage. The fat in yoghurt decreased as a function of storage in all treatment and the lowest fat 4.91 was observed in T_3 after 14 days of storage.

Treatments	Storage (days)			Means
	0	7	14	
T^0	5.1	5.09	4.95	5.04 ± 0.08
T^1	5.05	5.04	4.99	5.02 ± 0.03
T^2	5.0	5.0	4.90	4.96 ± 0.05
T^3	5.0	5.03	4.91	4.99 ± 0.06
T^4	5.05	5.05	4.91	5.00 ± 0.08
Means	5.046	5.042	4.932	

Table 3: Effect of lycopene on the fat of yoghurt during storage

T_0 = yoghurt with lycopene 0%; T_1 = yoghurt with lycopene 0.25%; T_2= yoghurt with lycopene 0.5%; T_3 = yoghurt with lycopene 0.75%; T_4 = yoghurt with lycopene 1.0%

The results indicated that treatments showed non-significant effect on the viscosity of yoghurt while storage days exhibiting significant effect on the viscosity of yoghurt. The interaction of treatment and storage time was also found to be significant for viscosity of yoghurt (Table 4). The results indicated that viscosity of the yoghurt ranged from 1146 to 1395 centipoise(cp) among the yoghurt prepared by using different levels of lycopene during storage.

Treatments	Storage (days)			Means
	0	7	14	
T^0	1395	1290	1140	1275 ± 128.16
T^1	1385	1290	1147	1274 ± 119.08
T^2	1393	1280	1146	1273 ± 123.98
T^3	1390	1283	1149	1274 ± 120.74
T^4	1386	1289	1147	1274 ± 120.94
Means	1389.8	1286.4	1145.8	

Table 4: Effect of lycopene on the viscosity of yoghurt during storage

T_0 = yoghurt with lycopene 0%; T_1 = yoghurt with lycopene 0.25%; T_2= yoghurt with lycopene 0.5%; T_3 = yoghurt with lycopene 0.75%; T_4 = yoghurt with lycopene 1.0%

The results indicated that the scores assigned to taste and mouth feel of yoghurt ranged from 7.02 to 9.0 among the yoghurt prepared by using different levels of lycopene during storage (Table 5). It is obvious that taste and mouth feel varied significantly among the yoghurt prepared by using different concentrations of lycopene progressed. Maximum score was recorded at 0 day of storage while minimum score for taste and mouth feel was observed after 14 days of storage. The taste and mouth feel in yoghurt decreased as a function of storage in all treatments and the maximum score (9.0) was observed in T_3 at 7 day of storage. The storage showed a significant effect on taste and mouth feel exhibiting decreasing trend as storage.

Treatments	Storage (days)			Means
	0	7	14	
T^0	8.6	7.2	7.06	7.62 ± 0.08
T^1	8.4	8.26	8.06	8.24 ± 0.01
T^2	8.72	8.6	7.86	8.40 ± 0.04
T^3	8.92	9	8.32	8.74 ± 0.37
T^4	8.66	8.4	7.72	8.26 ± 0.48
Means	8.66	8.292	7.804	

Table 5: Effect of lycopene on taste and mouth feel of yoghurt

T_0 = yoghurt with lycopene 0%; T_1 = yoghurt with lycopene 0.25%; T_2= yoghurt with lycopene 0.5%; T_3 = yoghurt with lycopene 0.75%; T_4 = yoghurt with lycopene 1.0%

Discussion:

Acidity results were similar to the finding of Jogdand SB *et al.,* and Khalifa ME. *et al,.* who stated that the acidity increases with prolonged storage time. [17,18] The results of present study are also in concordance with Gueimonde M. *et al.,* who observed that the acidity of yoghurt increases with the increased storage period due to microbial activity and lactose conversion into lactic acid.[19] The protein in yoghurt decreased as a function of storage in all treatments and the lowest protein (4.15) was observed in T_0 after 14 days of storage. These results were according to the Eissa EA. *et al.,* who termed that protein decreased during the storage of cow and goat milk yoghurt.[21] The fat, viscosity, protein in yoghurt decreased as a function of storage in all treatment and the lowest fat (4.90) was observed in T_3 after 14 days of storage. These results were according to the Eissa EA. *et al.,* who described that the fat decreased during the storage of cow and goat milk yoghurt. They also stated that the viscosity, taste and flavor decreased from 1st day to 15 days of storage.[20] Current results were also according to the Hanif MS *et al.,* that the storage mean scores showed significant variations for taste score among different storage intervals, while treatments differed non significantly. The mean scores for taste decreased gradually during storage.[21] Sarkar S *et al.,* also reported similar results and found that there was little bit change in the overall acceptability of the product during storage.[23] Current results were similar to the Eissa EA *et al.,* that the flavor decreased during the storage of cow and goat milk yoghurt.[21] Results of current study were also according to Hanif MS *et al.,* who revealed that the storage means showed significant variation for flavor score among different storage intervals while treatments differed non significantly. The mean scores for flavor decreased gradually during storage.[21]

Conclusions:

The present investigation suggested that lycopene has great antioxidant potential which must be utilized in fatty products to increase quality and shelf life. However, practical applications are only possible if purified extract is used as a source of antioxidant. Overall acceptability of lycopene supplemented yoghurt was same for the treatments having natural antioxidants. This means that the yoghurt were equally acceptable with natural antioxidant .This concept of natural antioxidant will lead towards production of functional foods besides prolonging the shelf life of food product.

References:

1- Gandhi DN. Food and Industrial Microbiology: Microbiology of Fermented Dairy Products. Principal Scientist dairy Microbiology Division National Dairy Research Institute, Karnal. 2006.

2- Henson S, Masakure O, Cranfield J. The propensity for consumers to offset health risks through the use of functional foods and nutraceuticals: The case of lycopene. Food quality and preference. 2008 Jun 1;19(4):395-406.

3- Srividya, DN. and Rao, SM. Dahi a health giving diet. Ind. J. Dairy Chem. 2003, 75: 95-102.

4- Shahidi F. Nutraceuticals and functional foods: whole versus processed foods. Trends in Food Science & Technology. 2009 Sep 1;20(9):376-87.

5- Egydio JA, Moraes ÂM, Rosa PT. Supercritical fluid extraction of lycopene from tomato juice and characterization of its antioxidation activity. The Journal of Supercritical Fluids. 2010 Aug 1;54(2):159-64.

6- Shi J, Maguer ML. Lycopene in tomatoes: chemical and physical properties affected by food processing. Critical reviews in food science and nutrition. 2000 Jan 1;40(1):1-42.

7- Halliwell B, Gutteridge JM. Free radicals in biology and medicine. Oxford University Press, USA; 2015.

8- Krinsky NI, Johnson EJ. Carotenoid actions and their relation to health and disease. Molecular aspects of medicine. 2005 Dec 1;26(6):459-516.

9- Baron JA, Cole BF, Mott L, Haile R, Grau M, Church TR, Beck GJ, Greenberg ER. Neoplastic and antineoplastic effects of β-carotene on colorectal adenoma recurrence: results of a randomized trial. Journal of the National Cancer Institute. 2003 May 21;95(10):717-22.

10- Hasler CM. The changing face of functional foods. Journal of the American College of Nutrition. 2000 Oct 1;19(sup5):499S-506S.

11- Sahan N, Yasar K, Hayaloglu AA. Physical, chemical and flavour quality of non-fat yogurt as affected by a β- lucanhydro-colloidal composite during storage. Food Hydrocolloids. 2008 Oct 1;22(7):1291-7.

12- Teles CD, Flores SH. The influence of additives on the rheological and sensory properties of nonfat yogurt. International journal of dairy technology. 2007 Nov 1;60(4):270-6.

13- Xu F, Yuan QP, Dong HR. Determination of lycopene and β-carotene by high-performance liquid chromatography using sudan I as internal standard. Journal of chromatography B. 2006 Jun 21;838(1):44-9.

14- Machmudah S, Kawahito Y, Sasaki M, Goto M. Process optimization and extraction rate analysis of carotenoids extraction from rosehip fruit using supercritical CO2. The Journal of Supercritical Fluids. 2008 Apr 1;44(3):308-14.

15- Association of Official Analytical Chemists, (US). Official methods of analysis of the Association of Official Analytical Chemists. Association of Official Analytical Chemists.; 2006.

16- Steel RG, Torrie JH, Dickey DA. Principles and procedures of statistics: A biological approach. McGraw-Hill; 1997.

17- Jogdand SB, Lembhe AP, Ambadkar RK, Chopade SS. Incorporation of additives to improve the quality of dahi. Indian Journal of Dairy Science. 1991;44:459-60.

18- Khalifa ME, Elgasim AE, Zaghloul AH, Mahfouz MB. Applications of inulin and mucilage as stabilizers in yogurt production. American Journal of Food Technology. 2011;6(1):31-9.

19- Gueimonde M, Alonso L, Delgado T, Bada-Gancedo JC, de los Reyes-Gavilán CG. Quality of plain yoghurt made from refrigerated and CO2-treated milk. Food research international. 2003 Jan 1;36(1):43-8.

20- Eissa EA, Ahmed IM, Yagoub AE, Babiker EE. Physicochemical, microbiological and sensory characteristics of yoghurt produced from goat milk. Livestock Research for Rural Development. 2010;22(8):281-6.

21- Hanif MS, Zahoor T, Iqbal Z, Ihsan-ul-Haq AA. Effect of storage on rheological and sensory characteristics of cow and buffalo milk yogurt. Pakistan Journal of Food Sciences. 2012;22(2):61-70.