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Storage related changes in *Carrot incorporated low fat frozen yoghurt*

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Abstract
The storage study of optimized Carrot incorporated low fat frozen yoghurt was carried out at temperature of -18±1°C up to acceptable sensory level by the judges. The results revealed that the period of storage had a significant effect on the quality characteristics of frozen yoghurt. A significant decreasing trend was noticed for total sensory score with advancement in the storage period. Acidity of frozen yoghurt showed an increasing trend. A significant decreasing trend was noticed for lactic acid bacteria count for both frozen yoghurt during storage up to 105 days. The sample of carrot incorporated low fat frozen yoghurt and control frozen yoghurt both were free from coliform throughout period of storage. This indicated that the manufacturing, handling and packaging of frozen yoghurt were carried out under strict hygienic condition. A significant increasing trend was noticed for yeast and mold count for both frozen yoghurt during storage up to 105 days.

Keywords: Carrot, *Low fat frozen yoghurt*, Storage study

Introduction

Nearly 22 per cent of the world total milk production is produced in India, while rural populations who are smaller, marginal farmers and landless people account for 70 per cent of this production. This demonstrates the significance of the Indian dairy sector to social and economic growth. As per capita availability increased to 427 g/day in 2020–21 from 178 g/day in 1991–92 and as milk production increased to 209.96 MT in 2020–21 from 55.6 MT in 1991–92, dairy is progressing at a superior accelerated rate (NDDB and DAHD, 2021).

A composite fermented frozen dairy dessert known as frozen yoghurt combines the dietary and sensory benefits of fermented milk products with the appearance and texture of ice cream. The method involves making frozen yoghurt mix by combining all of the components with stabilizers/emulsifiers, sugar, and stirred yoghurt. The frozen yoghurt mix and stirred yoghurt combination are then placed in an ice cream freezer to be frozen (Tamime and Robinson, 2007) [27]. Three fundamental production processes may be used to manufacture frozen yoghurt: (1) direct acidification, also known as the "one stream method," (2) indirect acidification, sometimes known as the "two streams method," and (3) mixing method (Westerbeek, 1995; Hui, 1993) [29, 10]. Given its low fat content (regular frozen yoghurt has a fat content ranging between 3.5 per cent and 6 per cent) and lower concentration of lactose, which solely depends on the type and length of the fermentation period, it can be considered a healthy substitute for ice cream for those suffering from obesity, lactose intolerance, and cardiovascular diseases (Marshall *et al.*, 2003; Tamime and Robinson, 2007) [14, 27].

Live strains of the bacterial species *Lactobacillus delbrueckii*, subsp. *bulgaricus*, and *Streptococcus thermophilus* must be present in yoghurt. Although frozen yoghurt is not regulated in the majority of legal systems, including those in the United States and Canada, it should still comply with certain standards. A quantity of milk is typically heated to 85 °C for 15 minutes before the yoghurt cultures are added. The combination is then allowed to produce acid and flavour over an incubation period at roughly 42 °C. The remaining pasteurized elements of the mixture are then combined with this cultured material. Milk fat of 2 per cent, 14 per cent milk solids not fat (MSNF), 0.35 per cent stabilizer, 68.65 per cent water and 15 per cent sugar are typical compositions of the finished product. This would be the outcome of mixing 20 per cent non-fat, plain, unsweetened yoghurt at 12.5 per cent MSNF with 80 per cent of a sweet mixture that had 2.5 per cent fat, 14.4 per cent MSNF, 0.44 per cent stabilizer and 18.75 per cent sugar. Some regulatory authorities have specified a minimum criterion for acidity of 0.30 per cent for final titratable acidity, expressed as lactic acid. Other nations choose to establish a standard for the amount of live bacteria in a product at a certain point after manufacturing, such as 10^7cfu/g (Goff and Hartel, 2013) [15].

Carrots are a root vegetable that are produced, exported, and eaten in many different nations. The *apiaceae* family is notable for having the most valuable member—carrots. This is as a result of a variety of elements, such as nutritional quality, phytochemical content, antioxidant capacity, and health advantages (Leja *et al.*, 2013) [13]. Carrot roots may be extensively processed into nutrient-rich finished products such juice, dry powder, concentrate, tinned, candy, gazrailla, preserve and pickle in addition to the traditional uses for salad and curry preparation in India. The *Gaajar Kaa Halwaa* carrot dish is the most well-known variant in north India. Grated carrots are boiled in milk till the whole combination is firm, and then butter and nuts are added (Sharma *et al.*, 2012) [23]. The ultimate product acceptability is significantly influenced by the quality of the carrot. High peel to core ratio, high total soluble content, high carotenoid concentration, and low fiber content are all desirable characteristics for carrots. Processing factors including cooking times, temperatures, and how long shredded carrots are blanched all have a significant effect on the final product quality. The formation of a product with a better flavour is facilitated by cooking it for a longer time at a lower temperature (Aneja *et al.*, 2002) [2]. It would be profitable to use carrot byproducts, such as carrot pomace, which contains roughly 50 per cent β-carotene, to add to baked goods like cakes and biscuits and to make a variety of useful items (Singh *et al.*, 2006) [25]. The production of frozen yoghurt made with carrots enhances customer's health.

Materials and Methods

The experiment studies were conducted in the Dairy Technology Department, SMC College of Dairy Science, Kamdhenu University, Gandhinagar, Gujarat, India.

Milk and Skim milk procured from Anubhav Dairy of SMC College of Dairy Science KU, were used for the preparation of *Carrot incorporated low fat frozen yoghurt*. Milk was standardized at 2.5% fat and 14.5% SNF. Good quality Carrot powder, Stabilizer-emulsifier mixture and vanilla flavour and orange colour were procured from market for development of frozen yoghurt. Skim milk powder (max. 1.0% fat, 3.5% moisture, 35% protein, 7% ash and 52% carbohydrates) of Amul was procured from the local market of Anand. Good quality commercial grade cane sugar, procured from the local market of anand was used for development of frozen yoghurt. The Direct Vat freeze-dried starter culture which was used for manufacture of frozen yoghurt was bought from Christen Hansen, Denmark (Exact *Dahi* 2).

All glassware such as conical flask, beakers, volumetric flaks, measuring jars were cleaned using detergents and sterilized using hot air oven set at 160-180 ºC temperature for 2 hours before use. High purity commercially available chemicals/ media were used in the investigation for different analyses. The packaging material (i.e. polystyrene cup) having 100 ml capacity with lids were purchased from local market of anand.

Method for Preparation of Frozen yoghurt

The frozen yoghurt was made by utilizing "Direct Acidification method" given by Gohil (2021) [8] by freezing in a batch ice cream freezer. Fresh milk procured from Anubhav Dairy, was standardized to 2.5% fat and 14.5% MSNF using skim milk and skim milk powder respectively. The standardized milk was heated to 45 ºC and dry ingredients (viz. sugar, SMP, carrot powder and stabilizer-emulsifier mixture) were added with vigorous agitation and the mixture is further heated to 70±2 °C. The mix at 70±2 °C was subjected to homogenization (I stage: 2500 psi; II stage: 500 psi) in a cleaned and sterilized homogenizer. The homogenized mixes were pasteurized at 85 °C for 15 min and cooled immediately to 42±2 °C. at this temperature, the mix was inoculated with DVS starter culture inoculated at a level of 7 g/100 kg frozen yoghurt mix and blended at low speed for 5 min for complete mixing of culture. The mix was then delivered to a sterilized stainless-steel container and covered with lid and incubated at 42±2 °C temperature until required acidity (0.60%LA) of the mix was obtained. The mix was then cooled to 4±2°C and aged overnight. The set curd was broken by blender and the colour (orange) and flavour (vanilla) were mixed and blended for 5 min. Followed by ageing and blending, for the manufacturing of various frozen yoghurt batches in batch freezer of direct expansion type (10.0 L cylinder capacity), the mixes were frozen in a batch freezer facilitated with the pressurized air incorporation. The frozen yoghurt was filled in pre-sterilized high impact polystyrene cups (100 ml) with lids. The frozen yoghurt was immediately subjected to hardening by transferring the cups in a hardening tunnel with -25±1 °C for 3 h. Frozen yoghurt in High Impact Polystyrene (HIPS) cups (100 ml) were stored in the deep freezer which maintained at -18±1 °C.

Response Surface Method (RSM) also known as design expert 13.0.5.0 software was employed for the development of carrot incorporated low fat frozen yoghurt process standardization and formulation. Frozen yoghurt was made by varying level of carrot powder (per cent), stabilizer-emulsifier mixture (per cent) and sugar (per cent) recommended by RSM to screen out the best optimized combination for most acceptable frozen yoghurt incorporated with carrot. The carrot powder level was chosen in the ranging from 2 to 6 per cent which is depending on preliminary experiments taken for carrot incorporated low fat frozen yoghurt. The stabilizer and emulsifier level was chosen depending on preliminary experiments and is ranging from 0.2 to 0.5 per cent. The sugar level ranging between 12 and 16 per cent was selected for this study depending on the results of preliminary experiments.

Optimization was done on the basis of sensory score and frozen yoghurt with 3.53% Carrot powder was found most suitable on the basis of sensory score. Based on the sensory acceptability of the product, it was decided to prepare frozen yoghurt with 3.53% Carrot powder. A control product without incorporation of Carrot powder was also prepared in the identical manner. Both the products were stored at -18±1 °C for 105 days and were analyzed periodically for sensory (i.e. flavour, body and texture, melting quality, colour and total score), chemical (i.e. titratable acidity and total solids) and microbiological (i.e. Lactic Acid Bacteria Count, Yeast and Mold Count and Coliform count) parameters. The analysis of product stopped when it was found to be unacceptable by the sensory panel.

Physico-chemical analysis

The method given by BIS (1989) was used to determine the titratable acidity in frozen yoghurt mix by taking 20.0 g sample of frozen yoghurt mix. The Mojonnier Milk Tester was used to determine the TS content in frozen yoghurt mix by using 5.0 g of melted frozen yoghurt (FSSAI, 2016). The physic-chemical analysis was performed at an interval of every 15 days.

Microbiological analysis

The Lactic Acid Bacteria (LAB) count of frozen yoghurt was determined as per the process of Downes and Ito (2001). Yeast and mold count of frozen yoghurt was determined as per the method given by IS-5403 (1999). Coliform count of frozen yoghurt was determined as per the method given by BIS (1964).

For the sample preparation exactly eleven gram of frozen yoghurt mix was aseptically weighed and transferred to 99 ml conical flask of sterilized phosphate buffer and mix it well. This is known as 1:10 dilution. After that the one ml diluent again aseptically transferred to 9 ml sterilized phosphate buffer tube. This is known 1:100 dilution. The same procedure was repeated to make required numbers of dilutions. After that 1 ml diluent was taken by sterilized pipette and transferred in to sterilized petri plates aseptically. The time gap between sample preparation and plating of sample should not more than 15 min. The petri plates containing the diluted samples were poured with melted and cooled (about 45°C) lactic count agar medium and allowed to solidify. The MRS agar and M17 agar were used to enumerate viable cell counts of *Lactobacillus* and *Streptococcus* respectively (Rybka and Kailasapathy, 1995; Kailasapathy *et al.*, 2008) [21, 12].

The Yeast and Mold count of all the samples were determined by same method as described in section above except that the medium used was Potato Dextrose Agar (PDA) along with added tartaric acid to set pH 3.5 of agar and the plates were incubated at 25±1 ºC for 3 to 5 days. The Coliform count of all the samples were determined by the method as described in above except that the medium used was violet red bile agar (VRBA), prepared as described in section above, and the plates were incubated at 37±1 ºC for 24 to 48 h.

Sensory evaluation

The frozen yoghurt samples, hardened at –25±2 °C in a deep freezer for 24 h, were softened to -13±2 °C temperature prior to serving to the trained judges (10 numbers) for sensory analysis. The judges were from the faculty of Dairy Technology Department, SMC College of Dairy Science, Anand. The frozen yoghurt samples of polystyrene (PS) cups (100 ml) were given for sensory analysis. The completely randomized and well labeled samples were used in sensory. At a time, only four samples were given for sensory analysis. The frozen yoghurt samples were judged using an ice cream scorecard (100-points) given by ADSA (Marshall *et al.*, 2013) [15].

Statistical Analysis

Completely randomized design (CRD) with 3 replications was used for data analysis.

Results and Discussion

Physico-chemical changes in frozen yoghurt during storage

The physico-chemical changes i.e. titratable acidity and total solids in frozen yoghurt are discuss as following:

Changes in titratable acidity

The impact of type of frozen yoghurt (T), period of storage (P) and their interaction (TxP) on acidity has been shown in Table 1.

Table 1: Influence of storage period on titratable acidity of control frozen yoghurt and carrot incorporated low fat frozen yoghurt

Storage period (Days)	Frozen yoghurt		Mean (period)
	Control (T0)	Experimental (T1)	
0	0.62 ± 0.03	0.61 ± 0.02	0.61^{c}
15	0.63 ± 0.02	0.62 ± 0.03	0.62^{c}
30	0.65 ± 0.02	0.63 ± 0.02	0.64^{b}
45	0.66 ± 0.02	0.64 ± 0.02	0.65^{b}
60	0.67 ± 0.03	0.65 ± 0.03	0.66^{a}
75	0.67 ± 0.02	0.65 ± 0.03	0.66^{a}
90	0.68 ± 0.02	0.66 ± 0.03	0.67^{a}
105	0.69 ± 0.02	0.66 ± 0.03	0.68^{a}
Mean (treatment)	0.66^{a}	0.64^{b}	
Source of variation	T	P	T×P
SEm±	0.005	0.009	0.013
CD (0.05)	0.013	0.027	NS
CV%	4.87		

There was a gradual increase in the acidity of both frozen yoghurts throughout the storage period of 105 days. The change in the acidity of frozen yoghurt differed significantly (P<0.05) for both treatment (T) and storage period (P). The interaction T x P was not affected significantly.

The magnitude of increase in the acidity within 105 days of storage was 10.63 per cent for T0 and 8.54 per cent for T1. This indicated that the carrot incorporated low fat frozen yoghurt (T1) was more resist against acidity rise during storage than control frozen yoghurt (T0).

Murtaza *et al.* (2004) [16] did the evaluation of ice cream quality which prepared with various blends of stabilizers/emulsifier. Ice cream was analyzed for sensory characteristics and physico-chemical properties during storage of 0, 10, 20, 30 and 40 days. The results of analysis of storage period revealed that acidity and pH were significantly affected by various blends of stabilizers/emulsifier (treatments) as well as in the storage period.

Changes in total solids

The impact of type of frozen yoghurt (T), period of storage (P) and their interaction (TxP) on total score has been shown in Table 4.19.

The total solids of the frozen yoghurt showed a gradually increase throughout the storage period. However, increase in the total solids differed significantly (P<0.05) only for the treatment (T). The storage period (P) and interaction of T x P failed to exert any significant effect on the total solids of frozen yoghurt. The data shown in Table 2 revealed that the non-significant increase in total solids of frozen yoghurt was noted throughout the period (P) of storage.

The magnitude of increase in the total solids within 105 days of storage was 6.23 per cent for T0 and 4.61 per cent for T1. This indicated that the carrot incorporated low fat frozen yoghurt (T1) was more stable to total solids during storage than control frozen yoghurt (T0). The lower rate of moisture loss of carrot incorporated low fat frozen yoghurt was might be due to interaction of carrot fiber with the moisture so that lower migration and lower loss of moisture during storage period.

Table 2: Influence of storage period on total solids of control frozen yoghurt and carrot incorporated low fat frozen yoghurt

Storage period (Days)	Frozen yoghurt		Mean (period)
	Control (T0)	Experimental (T1)	
0	32.04 ± 1.08	35.55 ± 1.20	33.80
15	32.44 ± 1.05	35.85 ± 1.12	34.15
30	32.78 ± 0.96	36.08 ± 1.14	34.43
45	33.09 ± 1.07	36.30 ± 1.35	34.70
60	33.42 ± 1.11	36.52 ± 1.26	34.97
75	33.76 ± 1.12	36.82 ± 1.22	35.29
90	34.04 ± 1.10	37.10 ± 1.23	35.57
105	34.17 ± 1.07	37.27 ±1.36	35.72
Mean (treatment)	33.22^{b}	36.44^{a}	
Source of variation	T	P	T×P
SEm±	0.236	0.472	0.668
CD (0.05)	0.683	NS	NS
CV%	3.32		

Murtaza *et al.* (2004) [16] evaluated the quality of ice cream made with various stabilizer/emulsifier blends. At 0, 10, 20, 30, and 40 days of storage, the physical, sensory and chemical properties of ice cream were examined. The examination of storage period data showed that ice cream samples with various stabilizer/emulsifier blends (treatments) and storage duration had a significant effect on moisture and total solids.

Pagthinathan (2020) [19] examined the sensory acceptability and physico-chemical composition of processed ginger incorporated ice creams. Five per cent ginger juice, five per cent ginger paste, and five per cent ginger syrup was used in the study to make different varieties of ginger ice cream. During 28 days of frozen storage at -20 °C, these ice cream samples were examined for their physico-chemical, microbiological, and sensory characteristics. The results of physico-chemical analysis revealed that total solids increased significantly ($p<0.05$) during storage.

Singh *et al.* (2014) [24] investigated the impact of frozen storage on the sensory, microbiological and physico-chemical quality of ice cream with a bakery flavour. A 10 per cent level of plain and chocolate cookies and vanilla and chocolate cakes were added to bakery-flavored ice cream, which was made, frozen for 60 days, and then periodically assessed every 15 days. The findings of the physico-chemical examination revealed that total solids, specific gravity and acidity of all ice cream samples was increased significantly ($p< 0.01$) throughout the storage period.

Results of current experiment are matching with above studies but no research is available in the literature related to the combined effect of carrot powder, stabilizer-emulsifier mixture and sugar on total solids of carrot incorporated low fat frozen yoghurt during storage.

Changes in the sensory qualities of frozen yoghurt upon storage

The average score for flavour, body and texture, melting quality, colour and appearance and total sensory score of the both frozen yoghurts as affected by the storage at deep freezer (i.e. -18±1 °C) were presented in Tables 3 to 7. The changes in the individual sensory attributes were discussed as following.

Flavour

The impact of type of frozen yoghurt (T), period of storage (P) and their interaction (TxP) on flavour score has been shown in Table 3.

The flavour score of the frozen yoghurt gradually decreased throughout storage due to reduction the intensity of flavour with the advancement of storage and also staleness observed with the progress of storage period ($P<0.05$).

The magnitude of decrease in the flavour score within 105 days of storage was 30.77 per cent for T0 and 30.34 per cent for T1. This indicated that the carrot incorporated low fat frozen yoghurt (T1) was more stable to flavour score during storage than control frozen yoghurt (T0).

Table 3: Influence of storage period on flavour score of control frozen yoghurt and carrot incorporated low fat frozen yoghurt

Storage period (Days)	Frozen yoghurt		Mean (Period)
	Control (T0)	Experimental (T1)	
0	43.38 ± 0.45	43.67 ± 1.40	43.52^{a}
15	41.57 ± 2.05	41.94 ± 1.75	41.75^{a}
30	39.85 ± 2.14	40.45 ± 1.78	40.15^{b}
45	37.88 ± 2.18	38.64 ± 1.77	38.26^{b}
60	36.10 ± 2.29	36.68 ± 2.19	36.39^{c}
75	34.22 ± 2.14	34.70 ± 3.29	34.46^{c}
90	32.13 ± 1.12	32.62 ± 1.53	32.37^{d}
105	30.03 ± 1.03	30.42 ± 1.31	30.22^{d}
Mean (treatment)	36.89	37.39	
Source of variation	T	P	T×P
SEm±	0.385	0.770	1.089
CD (0.05)	NS	2.229	NS
CV%	5.08		

The treatment (T) and the interaction (T x P) did not significantly influence the flavour score of frozen yoghurt during their storage at -18±1 °C.

No similar work has been carried out to study the influence of carrot on flavour during storage of carrot incorporated low fat frozen yoghurt.

Body and texture

The impact of type of frozen yoghurt (T), period of storage (P) and their interaction (TxP) on body and texture score has been shown in Table 4.

The score of body and texture of the frozen yoghurt showed a gradual reduction throughout storage. Such decrease in the body and texture differed significantly only for the storage period ($P<0.05$). The treatment (T) and interaction of T x P failed to exert any notable impact on the body and texture of frozen yoghurt.

Table 4: Influence of storage period on body and texture score of control frozen yoghurt and carrot incorporated low fat frozen yoghurt

Storage period (Days)	Frozen yoghurt		Mean (period)
	Control (T0)	Experimental (T1)	
0	26.60 ± 1.73	27.64 ± 1.23	27.12[a]
15	25.52 ± 1.60	26.59 ± 1.47	26.05[a]
30	24.45 ± 1.28	25.23 ± 1.14	24.84[b]
45	23.37 ± 1.41	24.14 ± 1.14	23.76[b]
60	22.11 ± 1.57	22.89 ± 1.40	22.50[c]
75	21.56 ± 0.71	21.88 ± 1.69	21.72[c]
90	20.44 ± 0.71	20.87 ± 0.91	20.65[d]
105	19.38 ± 1.07	20.23 ± 0.97	19.81[d]
Mean (treatment)	22.93	23.68	
Source of variation	T	P	T×P
SEm±	0.263	0.527	0.745
CD (0.05)	NS	1.525	NS
CV%	5.54		

The magnitude of decrease in the score of body and texture within 105 days of storage was 27.15 per cent for T0 and 26.83 per cent for T1. This indicated that the carrot incorporated low fat frozen yoghurt (T1) was more stable to body and texture during storage compared to control frozen yoghurt (T0).

The score of body and texture of the frozen yoghurt showed a gradually decreased throughout storage due to proteolysis (Amirdivani and Baba, 2011) and also iciness observed with the advancement of storage period.

No research is available in the literature related to the combined effect of carrot powder, stabilizer-emulsifier mixture and sugar on score of body and texture of carrot incorporated low fat frozen yoghurt during storage.

Melting quality

The impact of type of frozen yoghurt (T), period of storage (P) and their interaction (TxP) on melting quality score has been shown in Table 5.

Table 5: Influence of storage period on melting quality score of control frozen yoghurt and carrot incorporated low fat frozen yoghurt

Storage period (Days)	Frozen yoghurt		Mean (period)
	Control (T0)	Experimental (T1)	
0	4.50 ± 0.25	4.58 ± 0.14	4.54[a]
15	4.27 ± 0.25	4.37 ± 0.11	4.32[a]
30	4.06 ± 0.25	4.16 ± 0.15	4.11[b]
45	3.87 ± 0.25	3.97 ± 0.11	3.92[b]
60	3.67 ± 0.25	3.80 ± 0.26	3.73[c]
75	3.46 ± 0.25	3.60 ± 0.17	3.53[c]
90	3.27 ± 0.25	3.40 ± 0.26	3.33[d]
105	3.13 ± 0.25	3.40 ± 0.17	3.27[d]
Mean (treatment)	3.78	3.91	
Source of variation	T	P	T×P
SEm±	0.047	0.093	0.132
CD (0.05)	NS	0.270	NS
CV%	5.93		

The melting quality score of the frozen yoghurt showed a progressive decline during the entire storage period. Nevertheless, decrease in the melting quality differed significantly only for the storage period ($P<0.05$). The treatment (T) and interaction of T x P failed to exert any significant effect on the melting quality of frozen yoghurt. The data shown in Table 5 revealed that the significant ($P<0.05$) drop in melting quality score of frozen yoghurt was noted throughout the period (P) of storage.

The magnitude of decrease in the melting quality score within 105 days of storage was 30.37 per cent for T0 and 25.82 per cent for T1. This indicated that the carrot incorporated low fat frozen yoghurt (T1) was more stable to melting quality score during storage than control frozen yoghurt (T0).

There was a decrease in melting quality during storage due to proteolysis it directly affected the body and texture of product and because of that melting quality also got deteriorated. Rapid meltdown and flaky meltdown was observed in both frozen yoghurt with advancement of storage period.

No research are available in the literature related to the combined effect of carrot powder, stabilizer-emulsifier mixture and sugar on score of melting quality of carrot incorporated low fat frozen yoghurt during storage.

Colour and appearance

The impact of type of frozen yoghurt (T), period of storage (P) and their interaction (TxP) on colour and appearance score has been shown in Table 6.

Table 6: Influence of storage period on colour and appearance score of control frozen yoghurt and carrot incorporated low fat frozen yoghurt

Storage period (Days)	Frozen yoghurt		Mean (period)
	Control (T0)	Experimental (T1)	
0	4.25 ± 0.25	4.33 ± 0.29	4.29[a]
15	4.07 ± 0.23	4.07 ± 0.11	4.07[a]
30	3.90 ± 0.36	3.92 ± 0.25	3.91[b]
45	3.73 ± 0.31	3.73 ± 0.12	3.73[b]
60	3.58 ± 0.33	3.60 ± 0.20	3.59[b]
75	3.40 ± 0.35	3.43 ± 0.21	3.42[c]
90	3.23 ± 0.38	3.22 ± 0.20	3.22[c]
105	3.13 ± 0.34	2.98 ± 0.23	3.06[d]
Mean (treatment)	3.66	3.66	
Source of variation	T	P	T×P
SEm±	0.055	0.111	0.157
CD (0.05)	NS	0.321	NS
CV%	7.43		

The score of colour and appearance of the frozen yoghurt showed a progressive decline throughout the storage period. Such decrease in the colour and appearance significantly differed ($P<0.05$) only for the storage period (P). The treatment (T) and interaction of T x P failed to exert any notable impact on the colour and appearance of frozen yoghurt.

The magnitude of decrease in the score of colour and appearance within 105 days of storage was 26.27 per cent for T0 and 31.15 per cent for T1. This indicated that the carrot incorporated low fat frozen yoghurt (T1) was less stable to colour and appearance score during storage than control frozen yoghurt (T0).

There was a gradual decrease in score of colour and appearance of frozen yoghurt with the advancement of storage. The dullness in the colour was observed during storage. This may attributed to degradation of β-carotene molecules during the storage. The same results were reported by Park *et al*. (2015) for vanilla ice cream at -18 and -30 °C.

No research are available in the literature related to the combined effect of carrot powder, stabilizer-emulsifier mixture and sugar on score of colour and appearance of carrot incorporated low fat frozen yoghurt during storage.

Total score

The impact of type of frozen yoghurt (T), period of storage (P) and their interaction (TxP) on total score has been shown in Table 7.

The total score of the frozen yoghurt showed a progressive decline throughout the storage period. The period of storage (P) had a significant effect on the total score of frozen yoghurt (P<0.05). Whereas the treatment (T) and interaction T x P failed to impart any notable effect on the total sensory score of frozen yoghurt during their storage.

The total score is the reflection of flavour, body and texture, melting quality and colour and appearance score. The total score also reduced with the deterioration in sensory attributes during storage.

Table 7: Influence of storage period on total score of control frozen yoghurt and carrot incorporated low fat frozen yoghurt

Storage period (Days)	Frozen yoghurt		Mean (period)
	Control (T0)	Experimental (T1)	
0	93.74 ± 2.30	95.23 ± 2.76	94.48^{a}
15	90.42 ± 3.35	91.96 ± 3.19	91.19^{a}
30	87.27 ± 3.66	88.76 ± 2.83	88.02^{b}
45	83.85 ± 4.15	85.47 ± 2.94	84.66^{c}
60	80.46 ± 4.04	81.98 ± 3.32	81.22^{d}
75	77.64 ± 1.66	78.62 ± 1.32	78.13^{d}
90	74.07 ± 1.82	75.10 ± 1.70	74.59^{e}
105	70.68 ± 2.21	72.03 ± 2.16	71.36^{e}
Mean (treatment)	82.27	83.64	
Source of variation	T	P	T×P
SEm±	0.580	1.160	1.641
CD (0.05)	NS	3.357	NS
CV%	3.42		

Maximum score for bacteria (15) is added in total score No research are available in the literature related to the combined effect of carrot powder, stabilizer-emulsifier mixture and sugar on total score of carrot incorporated low fat frozen yoghurt during storage.

Changes in microbial qualities of frozen yoghurt during storage: The changes in microbial quality of control and experimental frozen yoghurt during storage stored at -18±1°C is depicted in Table 8 and 9.

Lactic acid bacteria (LAB) counts

The impact of type of frozen yoghurt (T), period of storage (P) and their interaction (TxP) on lactic acid bacteria counts has been shown in Table 8.

Table 8: Influence of storage period on lactic acid bacteria counts (log_{10}cfu/g) of control frozen yoghurt and carrot incorporated low fat frozen yoghurt

Storage period (Days)	Frozen yoghurt		Mean (period)
	Control (T0)	Experimental (T1)	
0	9.70 ± 0.14	8.06 ± 0.21	8.88
15	9.69 ± 0.15	8.04 ± 0.18	8.87
30	9.67 ± 0.15	8.03 ± 0.21	8.85
45	9.65 ± 0.16	7.99 ± 0.22	8.82
60	9.64 ± 0.16	7.96 ± 0.21	8.80
75	9.61 ± 0.13	7.94 ± 0.25	8.78
90	9.59 ± 0.15	7.90 ± 0.24	8.74
105	9.57 ± 0.17	7.88 ± 0.24	8.72
Mean (treatment)	9.64^{a}	7.97^{b}	
Source of variation	T	P	T×P
SEm±	0.039	0.077	0.109
CD (0.05)	0.112	NS	NS
CV%	2.15		

The carrot incorporated low fat frozen yoghurt had lower lactic acid bacteria count throughout the storage period as compared to control frozen yoghurt. The treatment (T) had a significant effect on the lactic acid bacteria count (P<0.05). There was a progressive decline in the lactic acid bacteria count during storage (i.e. -18±1°C) of frozen yoghurt. The extent of decrease in the lactic acid bacteria counts (as log_{10}cfu/g) from initial period to 105th day of storage for control frozen yoghurt (T0) and carrot incorporated low fat frozen yoghurt (T1) was 1.34 per cent and 2.23 per cent respectively.

It can be observed from table 8 that lactic acid bacteria count of carrot incorporated low fat frozen yoghurt are less than the control frozen yoghurt from starting period of storage. These lower counts of lactic acid bacteria count are mainly due to antimicrobial effect of carrot.

Nawal Galal *et al.* (2019) developed low-fat frozen yoghurt added with with carrot (*Dascus carota* L.) puree. In this study, the effect of addition carrot puree solids (CPS) on the frozen yoghurt characteristics was studied. The CPS added in the ratio of 1, 2 and 3 per cent as a replacer of milk fat. Rheological properties, physical properties, antioxidant scavenging activity, total phenolic compounds and total viable lactic acid bacteria (LAB) were analysed. The results showed that addition of CPS significantly increased the over run (per cent) and decrease the viablility of lactic acid bacteria of carrot frozen yoghurt.

Guirginov and Grouev (1961) examined that the *lactobacilli* counts was reduced from 10^{8}cfu/g to 5 x 10^{6}cfu/g, while the *S. salivarius* subsp. *thermophilus* counts remained stable after storing yoghurt frozen at -18/-22ºC for 2 months. Based on obtained results, they concluded that the latter microorganism was higher resistant to frozen temperatures compared to *L. delbrueckii* subsp. *bulgaricus*. Speck and Geoffrion (1980) performed a similar study but storing frozen yoghurt at -16ºC for 2 months and observed that during storage there was a scarcely perceptible reduction in LAB. Thompson and Mistry (1994) examined the survivability of LAB in yoghurt ice cream which was stored at -18ºC for 3 months and observed that storage conditions had no effects on total LAB.

However, a result of current experiment is in close agreement with above studies.

Coliform

The sample of carrot incorporated low fat frozen yoghurt and control frozen yoghurt both were free from coliform count throughout period of storage. This indicated that the manufacturing, handling and packaging of frozen yoghurt were carried out under strict hygienic condition.

Yeast and Mold Counts

The sample of carrot incorporated low fat frozen yoghurt and control frozen yoghurt both were free from coliform and yeast and mold count at beginning period of storage. This indicated that the manufacturing, handling and packaging of frozen yoghurt were carried out under strict hygienic condition. Coliform of carrot incorporated low fat frozen yoghurt and control frozen yoghurt were absent throughout the storage period.

There was a gradual increase in the yeast and mold counts of both frozen yoghurts throughout the storage period of 105 days (Table 9). The change in the yeast and mold counts of frozen yoghurt differed significantly ($P<0.05$) for both treatment (T) and storage period (P). The interaction T x P was not affected significantly. Nevertheless, according to FSSR (2015), the acceptable value of yeast and mold count in yoghurt and dahi should not be more than 100/g.

Table 9: Influence of storage period on yeast and mold counts of carrot incorporated low fat frozen yoghurt

Storage period (Days)	Frozen yoghurt		Mean (period)
	Control (T0)	Experimental (T1)	
0	4.67 ± 2.08	2.00 ± 3.00	3.34^{h}
15	12.00 ± 1.00	6.33 ± 1.53	9.17^{g}
30	20.00 ± 2.64	12.33 ± 1.53	16.17^{f}
45	27.33 ± 2.08	17.67 ± 2.08	22.50^{e}
60	32.00 ± 2.00	23.33 ± 2.52	27.67^{d}
75	39.67 ± 2.89	27.33 ± 2.31	33.50^{c}
90	48.67 ± 3.05	33.00 ± 2.64	40.84^{b}
105	52.67 ± 2.08	37.00 ± 4.36	44.84^{a}
Mean (treatment)	29.63^{a}	19.87^{b}	
Source of variation	T	P	T×P
SEm±	0.506	1.012	1.431
CD (0.05)	1.464	2.929	NS
CV%	7.82		

The increase in yeast and mold count of both frozen yoghurt may attributed to acidic condition of product. The lower count of carrot incorporated low fat frozen yoghurt compared to control frozen yoghurt may be due to antimicrobial action of carrot.

Salwa *et al.* (2004) prepared carrot yoghurt by addition of different concentration of carrot juice. Carrot yoghurt was made by mixing milk with 5, 10, 15 and 20% carrot juice prior to fermentation. The chemical, rheological, microbiological and sensory quality of yoghurt samples were analyzed during storage of three weeks at 4°C. The results of microbial analysis revealed that higher concentration of carrot juice decreased the growth of coliforms and yeast & mold counts. However, a result of current experiment is in close agreement with above studies.

Conclusions

The results obtained in the present investigation concluded that carrot incorporated low fat frozen yoghurt can be manufactured having low fat and good nutritive and therapeutic value. The developed frozen yoghurt was packaged in a polystyrene cup and stored in a deep freezer maintained at -18±1°C for up to 105 days of storage. The developed frozen yoghurt was found to be highly acceptable when freshly prepared and remain acceptable up to 90 days of storage.

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