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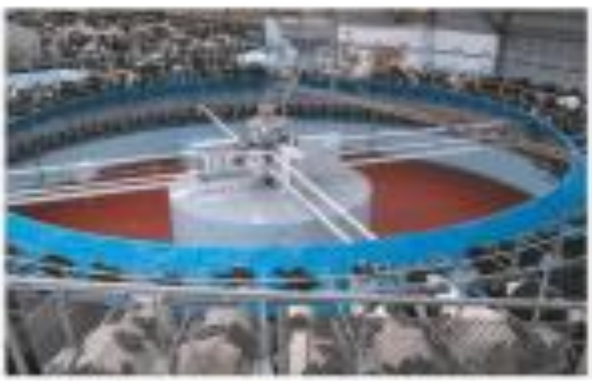


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Research Article

Improving The Properties of the Functional Frozen Bio-Yoghurt by Using Carrot Pomace Powder (*Daucus carota L.*)

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Abstract

Background and Objective: The dried carrot pomace has carotene and ascorbic acid, and is also a rich source of fiber, antioxidants, minerals including calcium, copper, magnesium, potassium, phosphorus, iron and folic acid which exhibit health promoting effects. The effect of adding carrot pomace powder (CPP), and half amount of the stabilizer/emulsifier mixture on some properties of frozen bio- yoghurt, when fresh and during storage period was studied. **Materials and Methods:** Dried carrot pomace was prepared and added in proportions of 5, 10, and 15% to frozen- bio yoghurt mixtures. Then study the effect of the addition on the physicochemical, microbial, sensory, specific weight, viscosity, freezing point, melting point, and probiotic properties of frozen bio- yoghurt. **Results:** Results indicated that, dry matter, ash, fiber, total carbohydrates and pH value were significantly increased in the mixes, while protein and fat contents were reduced significantly with the increased levels of CPP. The specific gravity, weight per gallon, freezing point, viscosity as well as flow time of the mixes containing CPP were found higher than the control. Physical properties of frozen bio-yoghurt product revealed that the specific gravity and weight per gallon were decreased whereas the overrun and melting resistances were increased. All treatments were organoleptically acceptable and the frozen bio-yoghourt product contained 10 % CPP showed superior sensory properties than the control treatment, followed by 5% and 15% CPP, in order. **Conclusion:** Generally , it could be recommended that frozen bio-yoghourt product of high quality can be made by adding CPP up to 10% as well as using half amounts of stabilizer/emulsifier mixture.

Key words: Frozen Yoghurt, By-Products, Carrot Pomace, Antioxidant, Prebiotic, Probiotics.

INTRODUCTION

Food's nutritional and potentially therapeutic value is an important consideration in the development of new value–added goods for health-conscious consumers. Frozen yoghurt has been the fastest growing product in the frozen dessert market in recent years [19,32].

Consumer interest in frozen yoghourt stems from the desirable nutritional properties attributed to the product[28]. As well to low-fat formulation frozen yoghurt supplemented with probiotic bacteria, e.g *Bifidobacteria longum* and *Lactobacillus. acidophilus,* provides additional health benefits. Health aspects attributed to the consumption of fermented dairy products supplemented with probiotic bacteria include improved lactose utilization, improved flavor and nutritional quality, and control of intestinal infections [5,26] proved that the frozen yogurt environment is not ideal for the survival of bacteria, due to the freezing process of the mixture besides the very low of temperature, a loss occurs in numbers from 1/₂ to 1 log cycle in viable counts.

Carrot (*Daucus carota L.*) is an important root vegetable which is used commonly for juice production. The juice yield in carrots is about 60–70%, and the rest is lost as carrot pomace[36]. Despite its superior residual amount of all the vitamins particularly vitamin A, B and C, minerals and dietary fiber, the carrot pomace does not find proper utilization and can become a source of environmental problem due to its high moisture content (about 88%). To extend its shelf life, carrot pomace can be dried. The dried carrot pomace has carotene and ascorbic acid, and is also a rich source of fiber, antioxidants, minerals including calcium, copper, magnesium, potassium, phosphorus, iron and folic acid which exhibit health promoting effects [44.] Regarding the nutritional quality and high fiber content of CPP, it has been used to enrich some foods including durum wheat pasta [18] and wheat flour biscuits[29.] Also, the nutritional and health importance of fiber-rich foods has been clarified[43.]

The pomace, a cause of environmental pollution may be converted to value added products, if processed properly[37]. Being fiber rich, the pomace may be incorporated into food products as inexpensive, non-caloric bulking agents for partial replacement of product component, fat or sugar, as enhancers of water and oil retention and to improve emulsion or oxidative stabilities[11,14].

The aim of this work was to study the effect of using carrot pomace powder and half the amount of stabilizer/emulsifier mixture on the physical, chemical, bacteriological, and sensory properties of the frozen bio-yoghurt product when fresh and during 90 days of storage period. In addition to low in calories and low in economic cost, while making use of carrot waste and preserving the environment from pollution.

MATERIALS AND METHODS

Study area: The study was carried out at the Department of Dairy Chemistry, Animal Production Research institute, Agriculture Research Center, Giza.

Materials:

Fresh whole raw buffalo's milk (6 % fat and 8.94 % SNF) used in this study was obtained from EL-Gemmeza Station Animal Production Research Institute, Egypt. Medium heat skim milk powder extra grade, Commercial grade of granulated sugar, vanilla (Chem. Rein 100 %) made by Boehringer Manheim GMB, Germany and carrot (*Daucus carota L.*) were made obtained from the local market. Commercial stabilizer/emulsifier mixture was obtained from MIFAD (Misr Food Additives company, Giza-, Egypt. Freeze dried starter culture (FD-DVS ABT-2) containing *Lactobacillus acidophilus, Bifidobacterium bifidum* and *Streptococcus thermophilus* was obtained from Chr. Hansen A/S, DK-2970 Horsholm, Denmark.

-Yoghurt starter culture consists of *Streptococcus thermophilus* and *Lactobacillus delbreuckii* sub sp. *bulgaricus* was obtained from Cairo MIRCEN Culture Collection Center, Faculty of Agriculture, Ain Shams University.

Methods

Preparation of carrot pomace powder (CPP):

The carrot pomace obtained after juice extraction was dried in an air-oven at 45°C during for 30 h. The dried sample was finely grounded using a grinder and passed through a 35-mesh sieve (0.40 mm).

Preparation of frozen bio- yoghourt:

frozen yoghurt products were prepared according to[20]. All frozen bio-yoghurt mixes produced were standardized to contain 4% fat, 15% sugar, 12% milk solids not fat, and commercial stabilizer/emulsifier mixture. Stabilizer/ emulsifier mixture was added at a ratio of 0.35 % for control treatment and 0.175 % for CPP treatments (T_1, T_2, and T_3). Then adding carrot pomace powder (CPP) at a rate of 5, 10 and 15% for T_1, T_2 and T_3 respectively. 4 mixtures were prepared separately, as shown in Table (2).

Table (1): The chemical composition of carrot pomace powder used in the preparation of frozen bio-yoghurt formula (g/100g)

Character assessed	Carrot pomace powder
Moisture%	4.08
Protein %	(TNx6.25) 1.85
Fat %	0.51
Ash %	3.57
Dietary Fiber %	78.6
Carbohydrates %	11.39
pH value*	7.89
Fe (mg/100 g)	3042
Zn (mg/100 g)	2927
Cu (mg/100 g)	398
Total caroteine (mg/100 g)	200
Total flavonoid (mg/100 g)	22.53
Total phenols (mg/100 g)	43.12
Ascorbic acid	32.95

*Determined in 20% aqueous solution (w/v).

Table (2): Mix composition of frozen bio-yoghurt recipes fortified with carrot pomace powder (kg/100 kg mix)

Ingredients	Treatments			
	Carrot pomace powder(CPP)			
	Control	T_1	T_2	T_3
Sugar	15	15	15	15
Stabilizer& Emulsifier	0.35	0.175	0.175	0.175
Cream(60% fat)	1.07	0.475	0.620	0.760
Raw milk (6%fat)	77.38	71.42	65.24	59.05
Skim milk powder (95% T.S)	3.200	4.930	5.965	7.015
Starter culture(1 YC: 1 ABT)	3	3	3	3
Carrot pomace powder	0	5	10	15

*$T_{1, 2, 3}$ treatments with fortified carrot pomace powder 5, 10, 15% respectively.

Ingredients for each treatment were divided into two portions (A) and (B). In portion (A) mixing the 50% from whole buffaloes milk and dissolving them in the skim milk powder , sugar, Stabilizer/ emulsifier mixture and Carrot pomace powder (CPP).Then homogenization at 250 kg/cm2 , heated at 85°C/10 min and cooled at 5°C ± 1 for 12 hr. . In Portion (B) The other 50% pasteurized whole buffaloes milk in all treatments were inoculated with 3% starter (1.5% yoghurt culture YC +1.5% ABT-2 probiotic starter, incubated at 42 ˚C until the pH value reached to 4.6 . The resultant yoghurt were put in the refrigerator at 5°C ± 1.Then, mix each two portions (A) and (B) in all treatments and put before freezing machine in in a horizontal batch freezer system.

The frozen products were packaged in plastic cups (80 ml) and hardened at -26˚ C before storage at -18˚C for 90 days.

Analysis:

Dry matter, fat, total nitrogen, ash and fiber were determined according to[2]. The pH value was measured electrometrically using Lab. pH meter with a glass electrode, Hanna digital pH meter. Denmark. Specific gravity of the frozen yoghourt mixes and the final frozen product were measured according to[43]. The weight per gallon (kg) of frozen yoghourt mixes and the final frozen products were calculated according to[25]. Freezing point of frozen yoghourt mixes were measured as described in[16] report, using Digital thermometer (Digitemp D 200/20, Germany). Whipping ability of frozen yoghourt mixes was determined as mentioned by[4] using electric mixer. The overrun percent was calculated as mentioned by[42]. Melting resistance of resultant frozen yoghourt was examined according to[37]. The viscosity of frozen yoghourt mixes was carried out as described by[39] using Brookifield DV- E viscometer. The mineral contents were determined according to the method described by the[2] using atomic absorption spectrometer. β- Carotene was determined as described by[7]**.** *L. acidophilus* count was determined according to[10] using MRS-Salcin agar. The plates were incubated at 37˚ C for 48 hrs. Yoghurt organisms*, L. delbrueckii subsp. bulgaricus* was determined using MRS–agar[35]. *S. thermophilus,* diluted samples were plated on M17 agar (Oxoid Ltd) and incubated at 37°C for 48-72 h under aerobic conditions[35]. *B. bifidum* was enumerated according to[10] using the modified MRS agar supplemented with 0.05% L. cysteine-HCl. The antibiotic mixture NPNL (neomycin sulphate, paramonycin sulphate, nalidixic acid and lithium chloride) as selective agent was sterilized by filtration through 0.22 µm millipore filter (Gelman Sci., England) then added to the medium at a rate of 50 ml/L medium just before pouring the plates. L. cystein-HCl solution sterilized by filtration (0.05% final concentration) was also added. The plates were anaerobically incubated at 37˚C for 48 hrs using anaerogen shachets.

Total flavonoids content of cream samples were determined as mentioned by[24]. Total phenolic compounds were determined according to[46].

Sensory evaluation

The organoleptic properties of frozen yoghourt were evaluated according to[3].

Statistical analysis

The data obtained were statistical analyzed according to statistical analyses system user's Guide[36].

RESULTS AND DISCUSSION

Data displaying in Table (3) are the chemical properties of bio- frozen yoghourt with mixes with fortified carrot pomace powder (CPP). The results shown that the fortifying with CPP led to significant differences in the dry matter (DM) contents of the product. Content of ash, fiber, carbohydrates and pH value were increased significantly (P< 0.001) while the protein and fat content were decreased significantly by increasing the level of CPP added. This is mainly due to the differences in chemical composition of both raw materials (Table, 1).

Table (4) indicated the effect of adding CPP and stabilizer/ emulsifier on some properties of frozen bio-yoghourt mixes. With regard to freezing point of frozen bio-yoghourt mixes, data indicating significantly gradual increase in the freezing point was associated with increased CPP ratio in product mix. Control treatment showed the lowest freezing point among all treatments. The higher freezing point in treatments contained CPP could be due to its lower lactose contents. These differences were associated directly with the rate of CPP added in the mixes and could, also, be attributed to the differences in the chemical composition of the materials used (Table, 1).

Freezing point is affect by the amount, type and molecular weight of the solutes in the mix [2914].

Data of Table (4) showing also tended significantly increase (p<0.001) in the flow time and viscosity of bio- frozen yoghourt mixes. The differences in flow time and viscosity values of control and treatments with CPP could be due to the differences in composition of both materials (Table, 1). CPP contains a high amount of fiber which able to gelatinize and therefore, increase the viscosity and flow time under the processing conditions of frozen yoghourt mix. Similar results were found by[13,14].

Table (3): Chemical composition of functional frozen bio- yoghurt mixes with fortified carrot pomace powder (CPP)

Properties		Treatment			
		Control	T_1	T_2	T_3
Dry matter	%	32.18^{d}	36.61^{c}	41.16^{b}	45.48^{a}
Total protein	%	4.740^{a}	4.136^{b}	3.892^{c}	3.63^{d}
Fat	%	4.07^{a}	3.47^{b}	3.44^{b}	3.42^{b}
Ash	%	1.051^{d}	1.218^{c}	1.375^{b}	1.556^{a}
Fiber	%	0.0^{d}	3.93^{c}	7.84^{b}	11.78^{a}
Carbohydrates	%	22.319^{d}	24.056^{c}	24.613^{b}	25.094^{a}
pH value		6.26^{d}	6.3^{c}	6.35^{b}	6.38^{a}

T1,2,3 treatments with fortified carrot pomace powder 5,10,15% respectively. The letter possess the factor of the level of CPP. The means with the same letter at any position did not significantly differ (p>0.05).

Table (4): Properties of physical bio-frozen yoghurt mixes with fortified carrot pomace powder (CPP)

Property	Treatments			
	Control	T_1	T_2	T_3
Freezing point ˚C	-3.17 ^{d}	-3.01 ^{a}	-3.09 ^{b}	-3.14 ^{c}
Flow time (sec.)	91.53 ^{d}	96.35 ^{c}	98.74 ^{b}	111.21 ^{a}
Viscosity (CP)	850^{d}	1060 ^{c}	1270 ^{b}	1420 ^{a}
Specific gravity	1.1336^{c}	1.1727^{b}	1.2000^{a}	1.2010^{a}
Weight (Kg)/ gallon	4.2997^{c}	4.4480^{b}	4.5517^{a}	4.5553^{a}

See details under Table (3).

That could be due to that DF had desirable functional properties, such as providing texture, gelling, thickening, emulsification, and stabilization in DF-enriched foods[32,12]. Therefore, DF research, particularly in the growing nutraceutical industry, has gained a lot of attention recently[23]**.** As appeared from the Table (4), the specific gravity and weight per gallon of frozen yoghourt mix, data indicating the gradual increase in both two properties ($p<0.001$) as the portion of CPP increase especially T_3. As seems from the results illustrated in figure (1) the whipping ability of bio-frozen yoghourt mixes with fortified CPP. All treatments indicated higher whipping ability at the beginning. The present figure clearly shows that the CPP had better whipping ability than control especially T_3. This is mainly due to functional (technological) properties like foaming and emulsifying of carrot pomace powder [41,11]**.**

The data shown in Table 5 showed some physical properties of bio- frozen yoghurt product fortified with different ratios of CPP. It turns out that the freezing time is reduced in all treatments to which carrot pomace is added compared to the control, and the freezing time is reduced by increasing the proportion of CPP added.

Concerning the melting resistance as affect by adding the CPP. Data in table (5) reveal that, in all treatments as CPP increased, melting resistance increased significantly, so that the treatment of 100% CPP exhibited the highest melting resistance at any certain experimental time ($p<0.001$).

We also note that there is an inverse relationship between freezing time and milting resistance of bio-frozen yoghourt. I think that the increase in the resistance to melting and the decrease in the freezing time in the treatments by the increase in the percentage of adding carrot bagasse is due to the increase in the percentage of fibers.

As appeared from Table 5, opposite to those occurred in the mix, both specific gravity and weight per gallon of resultant bio-frozen yoghourt were gradually increased by Increasing the ratio of addition ($p<0.001$). In terms of overrun, it was found that with an increase in the percentage of adding CPP, the overrun improved until the percentage of adding 10%, while it started to decrease at the percentage of adding 15%, but the percentage was equal to the control and there are no significant differences between T_3 and control.

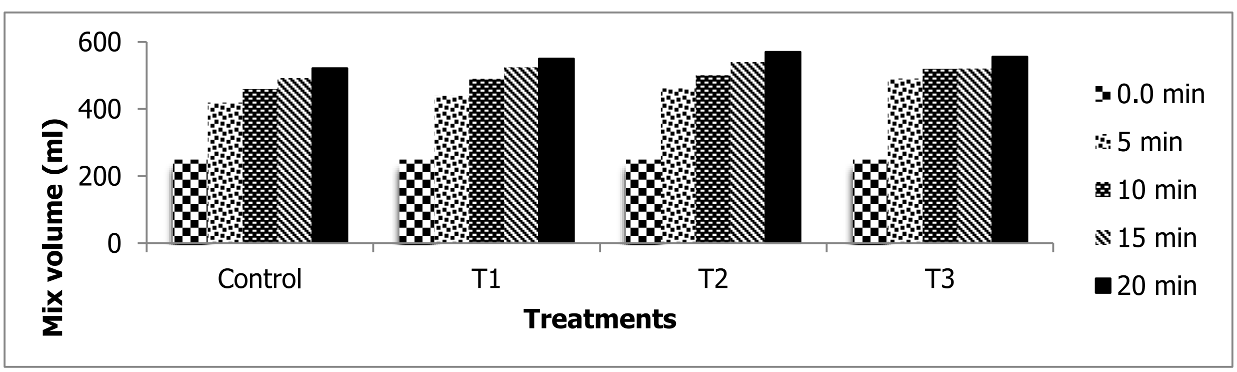

Fig (1): Whipping ability of functional bio-frozen yoghurt mixes with fortified carrot pomace powder (CPP).

Table (5): Properties of functional frozen bio- yoghurt with fortified carrot pomace powder (CPP)

Properties	Treatments			
	Control	T_1	T_2	T_3
Freezing time (min)	18.1^{a}	16.2^{b}	15.3^{c}	13.9^{d}
Melting after %				
10 min	16.83^{a}	16.01^{b}	15.54^{c}	15.14^{d}
20 min	43.49	40.81	40.67	32.98
30 min	65.18	61.85	60.11	46.87
40 min	80.03	77.64	75.29	73.44
50 min	91.36	89.7	87.41	84.68
60 min	100	98.31	94.02	90.30
Overrun %	69.95^{c}	70.20^{b}	72.84^{a}	69.76^{c}
Specific gravity	0.6670^{d}	0.6890^{c}	0.6943^{b}	0.7102^{a}
Weight per gallon	2.53^{d}	2.6134^{c}	2.6335^{b}	2.6938^{a}

See details under Table (3).

Probiotic analysis:

Table (6) presented the counts of *L. acidophilus*, *L. bulgaricus*, *S. thermophiles* and *B. bifidum* BB-12 during storage. After one day manufacture, treatments of frozen yoghourt with fortified CPP had higher counts of all Total viable bacterial counts than the control. The fiber of inulin was increased mineral absorption in the gut and can be used as an energy source by probiotic bacteria[33], which might explain the increased counts of *L. acidophilus*, *L. bulgaricus*, *S. thermophiles* and *B. bifidum* BB-12 of treatments by added[8]. Also, the counts of Probiotic bacteria increased with the increase in the proportion of CPP.

Regarding the storage period, it was observed that the counts of probiotic bacteria increased up to 30 days old. While all treatments except the control had higher than 10^6 (cfu/ml) viable cells of probiotic bacteria during storage Also, may the high fiber content in CPP seems to act as a protection of probiotic cells over the storage period[1].

The higher fiber contents in CPP treatments resulted in higher counts of probiotic cells over the storage period[1,26]. In addition, the presence of carrot pomace in the frozen yogurt product may be act as a prebiotic ingredient and stimulate the growth of these probiotic bacteria. These results were approximately similar to that found by[28]. The Food and Agriculture Organization/ World Health Organization report[16] stated tha**t**
probiotic foods with health claims must contain per gram at least 10^6 – 10^7 cfu at the time of consumption to exhibit their potential function.

Furthermore, it was found that the fortification of frozen bio- yogurt product with CPP, will extended its shelf life, because CPP was rich in phenolic and antioxidants compounds, which act as anti-microbial substances and increased the counts of probiotic bacteria. Similar trends were found by[22].

Sensory evaluation:

The sensory quality attributes of bio- frozen yoghourt samples with CPP as a are declared in table (7). The results revealed that adding with CPP up to 10% enhanced the sensory attributes of bio- frozen yoghourt samples. Bio- frozen yoghourt flavor and color were slightly affected in treatments with adding CPP showing lowest flavor score in T_3.

On the other hand, body and texture of bio-frozen yoghourt were improved with adding CPP. As for the melting resistance, as previously observed from Table 5, the CPP improved the melting resistance. T_2 exhibited highest organoleptic scores compared to other treatments including the control. Furthermore, bio-frozen yoghurt can be produced by fortifying it with CPP while reducing the added percentage of stabilizer/emulsifier by half.

Also, Table 7 showed that the sensory properties of all treatments in frozen organic yoghurt fortified with carrot pomace decreases significantly with the length of storage period.

Cost calculations

Table (8) estimates the cost of different bio-frozen yoghourt recipes with CPP. The total costs for treatments represent that bio-frozen yoghourt can be manufactured with up to 2.39, 2.37 reduction in the cost by adding 5, 10 % CPP respectively.

Conclusion:

In conclusion, the use of CPP in preparing the frozen bio-yoghourt product improved its nutritional quality and technological properties, due to the presence of antioxidant compounds (Flavonoids and carotenoids), dietary fibers, vitamins and minerals in CPP. Moreover, CPP may be act as a prebiotic ingredient and stimulating the growth of the vital microorganisms, especially probiotic bacteria, during the storage period.

Addition of CPP will, also, reduce the total costs of the resultant products by 2.37 % compared to the traditional manufacturing method. For all of the above, it can be produce this function frozen bio-yogurt to : use it as a nutraceutical and healthy product , minimize the environmental pollution , reduce the amount of stabilizer and emulsifier added and finally to be exploited in treating some of medical cases e.g. reducing the risk of constipation, cardio-vascular diseases, type-2 diabetes mellitus and colon cancer etc.

Table (6): Total viable bacterial counts of (cfu/ml) of functional frozen bio- yoghurt mixes with fortified carrot pomace powder (CPP) and during frozen storage at -20°C up to 3 months

Treatments	Frozen yoghurt after (day)			
	1	30	60	90
	S. thermophilus count (cfu/ml)*			
Control	5.8×10^6	4.1×10^6	1.2×10^5	3.6×10^4
T_1	6.0×10^6	6.3×10^6	6.0×10^6	5.7×10^6
T_2	6.3×10^6	1.2×10^7	6.8×10^6	6.4×10^6
T3	6.5×10^6	3.2×10^7	1.8×10^7	6.1×10^6
	L. bulgaricus count (cfu/ml)			
Control	6.1×10^6	4.2×10^6	2.6×10^5	5.8×10^4
T_1	6.3×10^6	6.9×10^6	6.2×10^6	4.1×10^6
T_2	6.6×10^6	2.6×10^7	6.1×10^6	5.3×10^6
T3	6.7×10^6	3.1×10^7	6.4×10^6	5.5×10^6
	L. acidophilus LA-5 (cfu/ml)			
Control	6.0×10^6	4.0×10^6	1.9×10^5	5.2×10^4
T_1	6.1×10^6	6.9×10^6	5.8×10^6	5.8×10^6
T_2	6.3×10^6	1.4×10^7	6.1×10^6	5.1×10^6
T3	6.4×10^6	2.2×10^7	6.8×10^6	5.3×10^6
	B. bifidum BB-12 (cfu/ml)			
Control	4.3×10^6	3.9×10^6	6.3×10^5	1.1×10^3
T_1	4.6×10^6	4.9×10^6	4.1×10^6	2.9×10^6
T_2	4.9×10^6	5.4×10^6	4.3×10^6	3.0×10^6
T3	5.1×10^6	5.5×10^6	4.4×10^6	4.3×10^6

See details under Table (3)
*Cfu/ml: Colony-forming unit/milliliter

Tale (7): Organoleptic properties of functional frozen bio- yoghurt mixes with fortified carrot pomace powder (CPP)

Treatment*	Storage period (month)	Flavor (45)	B &Tex (35)	MP** (10)	Appearance (10)	Total Score (100)
Control	Fresh	43	34	9	10	96^{Ac}
	1	41	33	8	10	92^{Bb}
	2	39	31	9	9	88^{Cc}
	3	38	30	8	9	85^{Dc}
T_1	Fresh	43	35	10	10	98^{Ab}
	1	43	34	9	10	96^{Ba}
	2	41	34	9	9	93^{Cb}
	3	38	32	9	9	88^{Db}
T_2	Fresh	44	35	10	10	99^{Aa}
	1	43	34	9	10	96^{Ba}
	2	42	34	9	9	94^{Ca}
	3	40	32	8	9	89^{Da}
T_3	Fresh	42	31	9	8	90^{Ad}
	1	40	30	9	7	86^{Bc}
	2	39	29	8	6	82^{Cd}
	3	38	28	8	6	80^{Dd}

**Melting properties
See details under Tables 3and4.

Table (8): Cost of different low fat bio-frozen yoghourt ingredients with fortified carrot pomace powder (CPP) (L.E/100kg mix)

Ingredients	Price L.E/Kg	Treatments			
		Control	T_1	T_2	T_3
Sugar	13.0	195	195	195	195
Stabilizer/emulsifier	100	35	17.5	17.5	17.5
Carrot pomace powder	-	-	-	-	-
Skim milk powder	70	224	345.1	417.55	491.05
Raw milk (6%fat)	15	1083.32	999.88	913.36	885.75
Cream (60% fat)	100	107	47.5	62.0	76.0
Total cost (L.E)	-	1644.32	1604.98	1605.41	1665.3
Cost reduction%	-	0.0	2.39	2.37	-1.28

$T_{1,2,3}$ treatments with fortified carrot pomace powder 5,10,15% respectively.

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