Functional Low-Fat Frozen Yoghurt with Carrot (Dascus carota L.) Puree

Khoulod Blassy[⊠]; Magdy Osman; Fawzy Abbas and Nawal Galal

Dairy Department, Faculty of Agriculture, Suez Canal University, 41522 - Ismailia, Egypt

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Abstract: The effect of addition carrot puree solids (CPS) at ratio of 1, 2 and 3% as a milk fat replacer on the characteristics of frozen yoghurt was studied. Physical, rheological, total phenolic compounds, antioxidant scavenging activity, total viable lactic acid bacteria (LAB), sensory properties and cost production of frozen yoghurt were investigated. The CPS is considered as sources for dietary fiber and natural antioxidant scavenging activity substances in low fat frozen yoghurt. Carrot frozen yoghurt contains antioxidant scavenging activity (4.52 mg TE 100 g⁻¹), total carotene (69.76 mg 100 g⁻¹), crude fibers (0.82 mg 100 g⁻¹), vitamin C (16.66 mg 100 g⁻¹), and total phenol contents (10.41 mg GAE 100 g⁻¹). Carrot puree contained high levels of many phenolic acid compounds such as of ρ -hydroxybenzoic, vanalic, catechin and ρ -coumaric acid. Low fat frozen yoghurt containing CPS exhibited significantly higher specific gravity, weight per gallon, melting resistance, apparent viscosity, total phenol contents and antioxidant scavenging activity in parallel with the increase of the added CPS. Addition of CPS significantly improved the over run (%) and decrease the viable of lactic acid bacteria of carrot frozen yoghurt. Whereas, addition of 3% CPS increased significantly the total acceptability scores with higher antioxidant scavenging activity, total phenol content and decreased the cost production by 22.51%.

Key words: Functional frozen yoghurt, antioxidant scavenging activity, carrot puree, low fat

INTRODUCTION

Yoghurt is a fermented dairy product that is produced by fermentation of live lactic acid bacteria. It contains an important nutrient such as proteins, lipids, calcium, potassium, phosphorus, magnesium, zinc and vitamins B (Yildirim *et al.*, 2014).

Frozen yoghurt or yoghurt ice cream is a complex of yoghurt and ice cream. This dessert combines the flavour of yoghurt and texture of ice cream. Consumers have a strong preference for functional foods containing specific ingredients and can reduce the threat of diseases such as cancer, heart diseases, obesity, cardiovascular diseases and lactose intolerance (Milani and Koocheki, 2011; Rezaei et al., 2015). Frozen voghurt's attractiveness to consumers include providing a low-fat replacement for ice cream and the probiotic benefits of the live cultures present in the voghurt Lactobacillus delbrueckii ssp. bulgaricus and Streptococcus thermophilus (Mahrous and Abd-El-Salam, 2014).

Carrot (Dascus carota L.) is one of the most commonly vegetables used to human nutrition. It is rich in β -carotene, to copherol, ascorbic acid, carbohydrate, (calcium, iron, phosphorous, copper. minerals potassium, manganese, sulphur) and improvement for body and texture were more pronounced than its flavour (Aly et al., 2004). It is an excellent source of vitamin A, E, B₁, B₂, C, folic acid, thiamin and riboflavin but lack in protein and fat (Hashimoto and Nagayama, 2004). Combination of carrot juice and yoghurt produce a nutritionally balanced food. The aim of this study was to explore the effect of replacement milk fat with fiber of carrot puree solids on the physical, rheological, antioxidant scavenging activity, total phenol content, total viable LAB count, sensory properties and cost production of low-fat frozen yoghurt to produce functional frozen yoghurt.

MATERIALS AND METHODS

Materials

Milk: Full cream milk powder (27% fat and 70% solid nonfat) and skim milk powder (97% T.S) low heat – spray dried $Miro^{TM}$, Sweden was obtained from the local market, Cairo, Egypt.

Starter cultures: Direct vat starter (DVS) culture containing *Streptococcus thermophilus* and *Lactobacillus delbreuckii* ssp. *bulgaricus* (YC-X11) was obtained from Chr. Hansen's Laboratories, Denmark. The culture was stored at $-18 \pm 1^{\circ}$ C until used before expired date.

Other materials: Commercial grade crystalline sugar (sucrose) was obtained from the local market. Sodium carboxy methyl cellulose (CMC) was obtained from Misr Food Additives - MIFAD - Cairo - Egypt. Carrot fresh was purchased from the local market (Eleubur market, Egypt).

Methods

Preparation of carrot puree

Carrot roots were washed with warm water after removing top, bottom and surface layers by sharp knife and soaked in hot water to inactivate pectinase and peroxidase enzymes and also to tenderize it and cut into sticks $(1\times3\times1 \text{ cm})$. The sticks were then soaked for 5 min in boiling water with a ratio of 1: 0.5 w/v carrot: water. The sticks along with soaked water were blended by Braun Power Max MX 2000 Blender (Germany) for 6 min to get fine paste. The puree was blended at 6000 rpm min⁻¹ for 5 min using Ultra Turrax Homogenizer (Germany) and kept in polyethylene bags at $-18\pm1^{\circ}$ C until used.

Preparation of frozen yoghurt

Frozen yoghurt mixes were prepared as described by Marshall and Arbuckle (1996). Five treatments of frozen yoghurt were carried out which describes the formulations of the different prepared mixes per 100 kilograms. The control full fat frozen yoghurt (T_1) was standardized (4% fat, 11% SNF, 15% sugar and 0.25% CMC) for comparison. While, other four reduced fat frozen yoghurt treatments (11% SNF, 15% sugar and 0.25% CMC) with gradual decrease of fat by 2, 1, 0 and 1% with adding 2%, 3%, 4 and 0 % CPS, respectively

for T_2 , T_3 , T_4 and T_5 treatments (Table 1). All treatments of frozen yoghurt were prepared in three replicates. Each treatment was analyzed for physiochemical, chemical, rheological, bacteriological and organoleptic characteristics at fresh (1 day) and after 15, 30 and 45 days of freezing storage at -18±1°C.

Table	(1): Com	position (Kg	100 K	g^{-1}) of frozen	yoghurt	mixes	containing	different	levels	of carrot	puree	solids
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Ingradiants	Treatments								
Ingredients	T ₁	T ₂	T ₃	T_4	T ₅				
Sugar (Kg)	15	15	15	15	15				
Stabilizer (Kg)	0.25	0.25	0.25	0.25	0.25				
Skim milk powder (Kg)	1.32	3.82	6.3	8.83	8.80				
Full cream milk powder (Kg)	14.28	10.71	7.14	3.57	3.50				
Water (l)	69.15	57.32	45.51	33.65	72.45				
Carrot puree solids (Kg)	-	12.90	25.80	38.70	-				
Total	100	100	100	100	100				

T₁: Full fat (4% fat) frozen yoghurt (FY) without carrot puree solids [CPS] (control₁).

T₂: Treatment with 3% fat FY with adding 1% CPS.

T₃: Treatment with 2% fat FY with adding 2% CPS.

T₄: Treatment with 1% fat FY with adding 3% CPS.

 T_5 : Treatment with 1% fat FY without adding CPS (control₂).

Analysis of carrot puree

The levels of total solids and crude fiber contents were determined according to AOAC (2000). Total carotenoids content was determined according to Moore *et al.* (2005), vitamin C as described by Elgailani *et al.* (2017) and pH values (Jenway pH meter with Jenway spear electrode No. 3505, Jenway limited, Gransmore Green, Felsted, Dunmow, England). The total soluble solids content was measured using with an ATAGO type refractometer (Schmidt Haensch, Germany) and the values were expressed as degree °Brix (%).

Colour measurement

The colour of CPS and resultant frozen yoghurt samples was measured using Hunter colorimeter (Hunter Ultra Scan XE, France). Measurements were recorded in L (lightness), + a (redness) and + b (yellowness) according to Commission Internationale de l'Eclairage, CIE (1976).

Determination of total phenolic compounds

The total phenol content was determined using the Folin-Ciocalteu method (Sigma-Aldrich, Germany) according to (Zilic *et al.*, 2012).

Determination of antioxidant scavenging activity

The antioxidant scavenging activity was determined by 2, 2-diphenyl-1-picrylhydrazyl (DPPH) according to Hwang and Do Thi (2014). Percent inhibition of the DPPH free radical was calculated by the following equation:

DPPH (%) = $[(A_{control}-A_{sample})/A_{control}] \times 100$

The standard curve was prepared using Trolox. Results were expressed as mg Trolox equivalents (TE) 100 g^{-1} sample. Additional dilution was needed if the DPPH value measured was over the linear range of the standard.

Analysis of phenolic acid compounds by HPLC

Phenolic compounds were extracted as described by Kim et al. (2006). HPLC analysis was carried out using Agilent Technologies 1100 series liquid chromatograph (Tedia company lnc, Ohio, USA) equipped with an auto sampler and a diode-array detector (Thermo FisherTM, USA). The analytical column was an Eclipse XDB-C18 (150 X 4.6 µm; 5 µm) with a C18 guard column (Phenomenex, Torrance, CA). The mobile phase consisted of acetonitrile HPLC grade (Sigma-Aldrich, Germany, solvent A) and 2% acetic acid (Almasria for Chemicals, Egypt) in water (v/v, solvent B). The flow rate was kept at $0.8 \text{ ml} \text{min}^{-1}$ for a total run time of 65 min and the gradient programme was as follows: 100% B to 85% B in 30 min, 85% B to 50% B in 20 min, 50% B to 0% B in 5 min and 0% B to 100% B in 10 min. Sample injection volume was 50 µl and peaks were monitored simultaneously at 280 and 320 nm for the benzoic acid and cinnamic acid derivatives, respectively. All samples were filtered through a 0.45 µm syringe filter (Sigma-Aldrich, Germany) before injection. Peaks were identified by congruent retention times and UV spectra and compared with those of the standards.

Analysis of β-carotene by HPLC

Carotenoids were extracted as described by Periago *et al.* (2004). β -carotene content was determined using HPLC. A 50 µl sample was injected into an Agilent Model 1100 HPLC (Agilent Technologies, Santa Clara, CA, USA), equipped with a quaternary pump and diode array detector using Agilent Technologies set at 461 nm. Separation was performed for 22 min per analysis on a ZORBAX Eclipse XDB-C18 column (150 x 4.6 mm ID; Agilent Technologies, Germany) at flow rate of 0.8 ml min¹. The column temperature was maintained at 20°C during the separation process. The mobile phase consisted of: A (acetonitrile and 0.1% butylhydroxy toluene [BHT]), B (acetone and 0.1% BHT) and C (water). The gradient elution as follows: in 1 min 60% A and 40% C, in 10 min 35% B and 12% C, in 12 min 100% B. Compounds

were identified by compared their retention time with standards. Data analysis was performed using Chem Station software (Agilent Technologies, Germany). All chemicals were HPLC grade and obtained from Merck (Darmstadt, Germany).

Analysis of the carrot puree frozen yoghurt mix and resultant frozen yoghurt

Physical analysis

All measurements of frozen yoghurt mixes were done in triplicates. The specific gravity and weight per gallon of both mixes and resultant frozen yoghurt was analysed as described by AOAC (2000). Freezing point of frozen yoghurt mixes was determined according to Arbuckle (1986). Overrun % was calculated as mentioned by Marshall and Arbuckle (1996). The melting rate of resultant frozen yoghurt was determined according to Segall and Goff (2002). The coulor reading characteristic was measured by Hunter colorimeter.

Viscosity of frozen yoghurt mixture

The apparent viscosity of all mixes of frozen yoghurt samples was measured using (Ametek Brookfield model LV rotary viscometer, DV-lllTM, USA) with spindle 03 and 50 rpm for different treatments. Viscosity was estimated using 200 ml of frozen yoghurt mix at 20°C. Measurements were taken every 3 min of spindle operation and at least 3 replicates performed. Values are expressed in centipoises (Cp).

Chemical analysis

Crude fiber content and acidity % were determined according to AOAC (2000). Total carotenoids content was determined according to Moore *et al.* (2005), vitamin C as described by Elgailani *et al.* (2017), antioxidant activity determination according to Hwang and Do Thi (2014), pH value and total phenolic compounds determined according to Zilic *et al.* (2012). The Phenolic acids were determined by HPLC, according to Kim *et al.* (2006).

Bacteriological properties

Eillker agar medium (Eillker et al., 1956) was used for the enumeration of total viable lactic acid

bacteria after incubation (Memmert, Germany) at 37°C for 48 h under aerobic condition.

Sensory properties

Sensory properties for the control and other treatments of frozen yoghurt were evaluated at fresh (1 day) and after 15, 30 and 45 days of storage at $-18\pm1^{\circ}$ C by staff members of the Dairy Department, Faculty of Agriculture, Suez Canal University, Egypt as described by Salama (2004).

Cost of production

The cost of production the different mixes were calculated according to Khalil and Blassey (2019). The available prices (L.E) of raw materials used in frozen yoghurt making in the Egyptian market. Full cream milk powder 68 (L.E Kg⁻¹), skim milk powder 54 (L.E Kg⁻¹), sugar 8 (L.E Kg⁻¹), CMC 65 (L.E Kg⁻¹) and carrot 2 (L.E Kg⁻¹).

Statistical analysis

Data were analyzed statistically by using CoStat Pro (2005), Version 6. 311. statistically different groups were determined by the LSD (least significant difference) test ($p \le 0.05$).

RESULTS AND DISCUSSION

Chemical properties of carrot puree (CP)

Carrot puree (CP) contented 7.75% total solids and its T.S.S 6.85%. It has a good content of fibers 0.82%. pH value of CP was 5.79. The colour reading characteristic using Hunter was recorded $L^{*50.75}$, a^{*} 32.69 and b^{*} 50.69 as shown in Table (2). So, CP had unique attractive orange colour.

Carrot and carrot puree solids are considered to be a good source of carotenoids, especially β -carotene, dietary fibers, sugars, minerals, vitamins and bioactive compounds and a small amount of fat therefore, provide many benefits for human health and nutrition (Di Giacomo and Taglieri, 2009). The variation chemical composition of CPS with other studies may be attributed to differences in th5e agricultural season and storage condition of the vegetables used in this study and coulor of carrot.

 Table (2): Chemical composition, total phenolic compounds, vitamin C, antioxidant scavenging activity and colour reading of carrot puree (average of three replicates)

Components		Carrot puree	
Moisture (%)		92.25	
T.S (%)		7.75	
T.S.S (%)		6.85	
Crude fiber contents (mg 100 g ⁻¹)		0.82	
pH value		5.79	
Total carotene contents (mg 100 g ⁻¹)		69.76	
Antioxidant scavenging activity contents (mg TE 100 g ⁻¹)		4.52	
Total phenolic compounds (mg GAE 100 g ⁻¹)		10.41	
Vitamin Contents (mg 100 g ⁻¹)		16.66	
Colour reading characteristics	<i>L</i> * 50.75	a * 32.69	b * 50.69

The CPS contained high levels of antioxidant scavenging activity (4.52 mg TE 100 g⁻¹), total phenol (10.41mg GAE 100 g⁻¹) and total carotene contents (69.76 mg 100 g⁻¹) as shown in Table (2). Carotenes contain mainly β -carotene and other antioxidants may protect humans against certain types of cancer and cardiovascular diseases and may enhance the immune system, protect against stroke, high blood pressure, osteoporosis, cataracts, arthritis, heart disease, bronchial asthma and urinary tract infections (Rafiq *et al.*, 2016). Their bright orange colour is related to the content of β -

carotene properly protected by inactivation of oxidative enzymes (Quitão-Teixeira *et al.*, 2008). Carrot puree contained high levels of ρ -hydroxybenzoic (1115.04 µg 100 g⁻¹) as shown in Table (3) and Figs. (1 a, b). These results are in agreement with Ahmad *et al.* (2017). Carrot puree contained 54.06 mg 100 g⁻¹ of β -carotene (Figs. 2a, b). These results suggest that the CPS can be used as functional ingredient with high dietary fibers diets. These results are in agreement with Susiloningsih *et al.* (2016). However, the CPS confirmed the high antioxidant scavenging activity especially β -carotene of the CPS.

Table (3): Identification of phenolic acid compounds in carrot puree samples determined by HPLC

Compounds	Peak number	Retention time (min)	Carrot puree (μg 100 g ⁻¹)
ρ-Hydroxybenzoic	4	12.082	1115.04
Catechin	6	16.672	86.89
Syringic	9	20.108	30.78
Vanillic	10	28.008	171.77
Ferulic	12	31.703	72.61
Sinapic	13	33.301	30.22
ρ-Coumaric	15	35.782	47.91
Apigenin-7-glucoside	18	41.234	33.79
Rosmarinic	19	49.968	7.26
Cinnamic	20	51.755	30.81



Fig. (1 a, b): HPLC chromatogram of standards (a) and carrot puree (b) phenolic acid compounds



Fig (2 a, b): HPLC chromatogram of standards (a) and carrot puree (b) β-carotene

Physical properties of carrot frozen yoghurt mixtures

The specific gravity and weight per gallon values of carrot frozen yoghurt mixes increased significantly with adding CPS (Table 4) as a result of higher solids not fat and lower fat contents of these mixes. These results are in agreement with El-Kholy (2018).

Freezing point depression (FPD) is a critical parameter in ice cream production as it influences the initial and gradual growth of the mean size of the formed ice crystals and also their native thermodynamic instability (Hartel, 2001). The freezing point of carrot

frozen yoghurt mixes (T_2 , T_3 and T_4) treatments decreased significantly (p ≤ 0.05) compared with control (T_1 and T_5) treatments might be due to high ash content and fiber of carrot puree solids (Table 4). El-Samahy *et al.* (2015) stated that when fat was removed from ice cream and replaced with nonfat milk solids or other dissolved substances, the freezing point was lowered. The use of vegetable in the manufacturing of ice cream mix added some sugars and minerals which caused more FPD as compared to low fat ice cream mix without using fruit. Similar finding was found by Khalil and Blassey (2019).

 Table (4): Effect of using different percentages of carrot puree solids on physical properties and total viable lactic acid bacterial count of carrot frozen yoghurt mixture

Developed properties -	Treatments								
r nysicar properties	T ₁	T ₂	T ₃	T ₄	T ₅				
Specific gravity	$1.087^d\pm0.00$	$1.118^{c} \pm 0.00$	$1.134^b\pm0.00$	$1.155^a\pm0.00$	$1.077^{e} \pm 0.00$				
Weight per gallon (kg)	$4.941^d\pm0.00$	$5.082^{c} \pm 0.00$	$5.154^b\pm0.00$	$5.250^a\pm0.00$	$4.895^{e} \pm 0.00$				
Freezing point °C	$-1.77^{d} \pm 0.01$	$-1.85^{\circ} \pm 0.01$	$-1.95^{b} \pm 0.01$	$-2.02^{a} \pm 0.01$	$-1.62^{e} \pm 0.00$				
Viscosity (Cp)	$779^d \pm 1.52$	$937^{c} \pm 2.64$	$1628^b\pm2.00$	$1767^{a} \pm 2.64$	$602^{e} \pm 2.51$				
LAB (Log cfu g ⁻¹)	$8.00^{a} \pm 0.09$	$7.59^b\pm0.05$	$7.35^{\circ} \pm 0.04$	$7.14^{d} \pm 0.09$	$7.96^{a}\pm0.03$				

Values are means \pm standard deviations of triplicate determinations

Means with the same row with different superscript (a, b, c...) are significantly different (p≤0.05)

The apparent viscosity of frozen yoghurt mix affects the body and texture of the finished product (Minhas *et al.*, 2002). The apparent viscosity of frozen yoghurt mix increased gradually as the ratio of fat substitution of CPS increased whereas, control low fat FY (T_5) showed the lowest apparent viscosity (Table 4). The data also indicated that the mixes with the addition of 4% CPS (T_4 treatment) had a higher viscosity, compared with other mixes. This might be due to the higher fiber content in carrot, which is responsible for gel forming viscous, as well as particles size and high-water holding capacity of fiber (Soukoulis *et al.*, 2009) and (Milani and Koocheki, 2011).

Bacteriological properties of carrot frozen yoghurt mixtures

Addition of 1, 2 and 3% of CPS in carrot frozen yoghurt decreased the total viable lactic acid bacterial count significantly ($p\leq0.05$) as compared with control T_1 and T_5 treatments (Table 4). The decrease in total viable LAB counts maybe attributed to the high content of phytochemicals of CPS added (Leong and Shui, 2002) during yoghurt processing. However, other studies (Radiati *et al.*, 2016) stated that no significant variation of LAB counts have been found between carrot juice frozen yoghurt and control.

Properties of resultant carrot frozen yoghurt Physical properties of carrot frozen yoghurt

Incorporation of air into frozen yoghurt mix during the pre-freezing process decreased the specific gravity of frozen yoghurt and weight per gallon as compared with its mixes values (Table 5). The specific gravity and weight per gallon were significantly (p \leq 0.05) higher for T₂, T₃ and T₄ carrot frozen yoghurt treatments compared with control $(T_1 \text{ and } T_5)$ treatments. While, T₄ treatment recorded the highest specific gravity and weight per gallon compare to all treatments (Table 5). In addition, full fat frozen yoghurt T_1 treatment tended to have higher overrun % than low fat without CPS. The increase of specific gravity depends on the formula component as well as mix ability to hold the air bubbles and overrun percent in the resultant FY (Khalil and Blassey, 2019). Overrun can be defined as the percent of FY expansion due to the incorporation of air into matrixes of FY mix during the freezing process (Cruz et al., 2009). Carrot frozen yoghurt made with CPS (T_2 , T_3 and T_4) treatments showed significantly ($p \le 0.05$) higher overrun than that of low-fat FY without CPS (T_5) treatment which can be attributed to the high viscosity mix containing CPS

(Table 5). Addition of 3% CPS causes decreased of overrun% probably due to its higher viscosity and decreased air bubbles stability. These results are in agreement with Erkaya *et al.* (2012) for ice cream containing cape gooseberry.

The meltdown rate is an important characteristic of ice cream. Slow meltdown and good shape retention are some of the ideal quality parameters of ice cream (Bahramparvar and Mazaheri-Tehrani, 2011). The increase in the melting resistance of carrot frozen voghurt was proportional ($p \le 0.05$) to the amount of CPS used (Fig. 3). Melting resistance of carrot frozen yoghurt was expressed as the loss in weight present of the initial weight of the tested formula during 60 min. The control FY (T_5) treatment had the highest melting rate than other frozen yoghurt treatments made with replacement of fat replacer by CPS. These results are in agreement with El-Kholy and Abbas (2015), they stated that the melting resistance was related to viscosity and freezing point of the FY mix. On the other hand, the initiation of fluid release was slower significantly for T_4 , T_3 and T_2 treatments, respectively. Generally, as the mix viscosity increased, the resistance of ice milk to melting increases (Salama, 2004). A lower freezing point of ice cream was reported to produce a softer but more unstable product during frozen storage (Eisner et al., 2005). These results are in agreement with Goh et al. (2008) who reported that the differences in melting resistance are mainly due to the differences in the freezing points of the treatments.

Colour is an important parameter to the quality of food products because of its association with factors such as freshness, ripeness, desirability and food safety. It is often a primary consideration for consumers when purchasing decisions (Arscott making and Tanumihardjo, 2010). All FY containing CPS exhibited orange colour of variable intensity according to the CPS ratio used. The colour of CFY was also evaluated by instrumental measurement of colour parameters (Fig. 4 a, b). Low fat FY with CPS treatments $(T_2, T_3 \text{ and } T_4)$ had lower L^* value indicating darker colour, higher redness (higher a^* values) and higher negative b^* (referring to blueness) than both full fat and low-fat FY $(T_1 \text{ and } T_5)$ treatments without CPS. All treatments of CFY with CPS appeared increases in L^* value and decreased in a^* and b^* values during storage period from fresh (1 day) until 45 days. These results are in agreement with El-Samahy et al. (2015).

Table (5): Effect of using different percentages of carrot puree solids on physical properties of carrot frozen yoghurt

Dhysical properties	Treatments									
r nysicar properties	T ₁	T ₂	T ₃	T_4	T ₅					
Specific gravity	$0.652^d \pm 0.00$	$0.697^{c} \pm 0.00$	$0.722^{b} \pm 0.00$	$0.750^{a} \pm 0.00$	$0.229^{e} \pm 0.00$					
Weight per gallon (kg)	$2.966^{d} \pm 0.01$	$3.169^{c} \pm 0.00$	$3.284^{b} \pm 0.01$	3.412 ^a 0.00	$1.041^{e} \pm 0.02$					
Overrun (%)	$66.10^{a} \pm 0.82$	$60.16^{b} \pm 0.40$	$56.95^{\circ} \pm 0.64$	$53.73^d {\pm}~0.48$	$43.31^{e} \pm 1.30$					

Values are means \pm standard deviations of triplicate determinations

Means with the same row with different superscript (a, b, c...) are significantly different ($p \le 0.05$)



Fig. (3): Effect of using different percentages of carrot puree solids on the melting rate of carrot frozen yoghurt



Fig. (4 a, b): Effect of using different percentages of carrot pure solids on colour parameters (L^* , a^* and b^*) for fresh (1 day) [a] and after 45 days of freezing storage (b) of carrot frozen yoghurt

Chemical analysis of carrot frozen yoghurt

The acidity % of CFY decreased slightly as percentage of CPS increased. While, in both control (T_1 and T_5) the acidity increased gradually compared with the other treatments, this may be due to the effect of low pH value in CPS add to CFY but T_1 was recorded the highest acidity during storage period at -18±1°C. On the other hand, pH value for T_4 treatment increased significantly (p≤0.05) as compared to control FY (T_1 and T_5) treatments (data are not shown).

Carotenoids (especially β -carotene) are important for human nutrition because of their provitamin A and antioxidant capacity (Agabriel *et al.*, 2007). The total carotene content of CPS fortified treatments (T_2 , T_3 and T_4) was increased significantly (p \leq 0.05) as the ratio of CPS increased compared with FY control (T_1 and T_5) treatments (Fig. 5). Due to low levels of total carotene content in milk, it would be useful for the human diet with supplementation of CPS, which is rich in total carotene. Generally, total carotene contents were decreased in all CFY samples and control during freezing storage. These results are in agreement with Dutta *et al.* (2005) who reported that the total carotene content losses significantly when increase freezing storage period.



Fig. (5): Effect of using different percentages of carrot puree solids on total carotene (mg 100 g⁻¹) of carrot frozen yoghurt

Vitamin C content of CPS fortified treatments increased significantly ($p \le 0.05$) as CPS increased in all treatments (T_2 , T_3 and T_4) compared with frozen yoghurt control (T_1 and T_5) treatments (Fig. 6). These results are in agreement with Jhansi and Sucharitha (2013) who stated that carrot ice cream recorded highest ascorbic acid content compare with the control ice cream. This result can be explained by the higher content of vitamin C in raw carrot compared to milk. On the other hand, it is clear that the value of vitamin C decreased during 30 days of freezing storage period at $-18\pm1^{\circ}$ C and stable until 45 days of storage period in all treatments. This which was probably due to the fact that ascorbic acid being sensitive to oxygen, light and heat. It was easily oxidized in presence of oxygen by both enzymatic and non-enzymatic catalyst. These results are in agreement with Jan and Masih (2012).



Fig. (6): Effect of using different percentages of carrot puree solids on vitamin C of carrot frozen yoghurt during storage period

The addition of dietary fiber to dairy products is one of the fields of functional foods. Carrots are high in dietary fibers which play an important role in human health and diets. Dietary rich fiber foods are associated with the prevention, reduction and treatment of some diseases such as diverticular and coronary heart diseases (Dundar and Gocmen, 2013). It can import functional properties to foods such as increasing the water retention capacity which improves the viscosity of the products (Rezaei *et al.*, 2015). Some fruits or vegetables mixtures improve the nutrition value of frozen yoghurt and provide a desirable taste for consumers and it plays a considerable role in frozen yoghurt consumption also, increases the consumer acceptability (Çakmakçi *et al.*, 2012).

Crude fiber content of CPS fortified CFY increased significantly ($p \le 0.05$) as CPS ratios increased in all treatments (T_2 , T_3 and T_4) compared to FY control (T_1 and T_5) treatments (Fig. 7). Addition of CPS at different ratios of carrot frozen yoghurt improved the viscosity. These results are in agreement with Khalil and Blassey (2011) who reported that the improve of viscosity may be due to thickening effect of the soluble fiber due to the three-dimensional confirmation of the hydrated biopolymers. In addition, to soluble fiber can modify and improve the texture and sensory properties of food because of its ability to bind with water, form

gels and its thickening properties (Soukoulis *et al.*, 2009).On the other hand, it is clear that crude fiber contents were stable during freezing storage period at $-18\pm1^{\circ}$ C when fresh until 45 days in all treatments.

Phenolic acid compounds content of carrot frozen yoghurt

Phenolic compounds are secondary plant metabolites, mainly composed of an aromatic ring bearing one or more hydroxyl groups, playing on oxidative stress. Phenolic compounds can be divided into different subgroups, such as phenolic acids, flavonoids and tannins. It has been reported that carrots are rich in phenolic acids, such as ρ -hydroxybenzoic, syringic, cateachin, vanillic, ferulic, ρ -coumaric, cinnamic, apigenin-7- glucoside and sinapic. These compounds were identified in the orange carrot (Alasalvar *et al.*, 2001; Ahmad *et al.* 2017).

The phenolic acid compound contents of CFY (T_2 , T_3 and T_4) treatments were higher than that of control (T_1 and T_5) treatments. The increased ratios of CPS added to FY increased the phenolic acid compound contents of CFY treatments. T_4 treatment was recorded highest phenolic acid compound contents compared with all treatments after 45 days of freezing storage period at -18±1°C (Table 6 and Fig. 8).







Fig. (8): HPLC chromatogram of phenolic acid compounds of carrot frozen yoghurt (T_4 treatment)

Table (6): Concentration of phenolic acid compound contents in carrot frozen yoghurt after 45 days of freezing storage period

Compounds	Peak	Retention time	Treatments (μg 100 g ⁻¹)					
I	number	(min)	T ₁	T ₂	T ₃	T ₄	T ₅	
<i>p</i> -Hydroxybenzoic	4	12.082	39.63	45.12	63.57	212.62	29.95	
Catechin	6	14.672	21.85	43.19	92.62	99.35	15.63	
Syringic	9	18.279	0.78	5.29	5.51	6.14	ND	
Vanillic	10	28.008	ND	6.42	20.72	39.31	ND	
Ferulic	12	31.703	ND	8.94	21.62	25.17	ND	
Sinapic	13	33.301	ND	3.49	4.50	5.57	ND	
<i>p</i> -Coumaric	15	35.287	ND	15.32	22.78	30.45	ND	
Apigenin-7-glucoside	18	41.234	ND	8.06	9.31	12.81	ND	
Rosmarinic	19	4 ⁹ .968	ND	3.85	5.52	6.24	ND	
Cinnamic	20	51.755	ND	3.90	10.90	15.63	ND	

ND: not detected.

 ρ -Hydroxybenzoic is found in a great amount in plant cell walls and it was the major phenolic acid compound. ρ -Hydroxybenzoic was recorded 39.63, 45.12, 63.57, 212.62 and 29.95 µg 100 g⁻¹ in T₁, T₂, T₃, T₄ and T₅ treatments, respectively. While the highest ratio was recorded in T₄ treatment. It potential to decrease cardiovascular problems related to aging such as hypertension, atherosclerosis and dyslipidemia (Juurlink *et al.*, 2014). Also, it is the primary bioactive component in preventing urinary tract infections thus prevents the growth of *Escherichia coli* on the urinary tract (Vattem *et al.*, 2005).

Catechin was recorded 21.85, 43.19, 92.62, 99.35 and 15.63 μ g 100 g⁻¹ in T₁, T₂, T₃, T₄ and T₅ treatments, respectively. While the highest ratio was recorded in T₄ treatment. The functional activity of catechin refer to prevent of some diseases such as cancer, obesity, diabetic, cardiovascular, Alzheimer's disease, infection (like influenza), improved antiviral activities, hepatoprotective and neuroprotective effects. Catechin is known to have dual actions in relation to reactive oxygen species as an antioxidant and a pro-oxidant (Isemura, 2019).

Syringic acid was recorded 0.78, 5.29, 5.51 and 6.14 μ g 100 g⁻¹ in T₁, T₂, T₃ and T₄ treatments, respectively. While the highest ratio in T₄ treatment.

Syringic acid is an excellent compound to be used as a therapeutic agent in various diseases (diabetes, brain ischemia, decreased the blood pressure, neuro and liver damage) and this reaction due to the presence of methoxy groups onto the aromatic ring at positions 3 and 5 in syringic acid. Also, it can improve the nonenzymatic antioxidants (vitamin C and vitamin E), stimulate the brain to send signaling for immune cells to prevent inflammation processes and it improves insulin secretion (Srinivasulu *et al.*, 2018). Vanillic was recorded 6.42, 20.72 and 39.31 μ g 100 g⁻¹ in T₂, T₃ and T₄ treatments, respectively. The highest ratio was recorded in T₄ treatment. While, it was absent in T₁ and T₅ (control) treatments.

Vanillic acid is known as Chinese medicine. It is a benzoic acid derivative that is used as a flavoring agent. It is an oxidized form of vanillin. Vanallic acid can be prevent the harmful effects of chronic stress on cognitive function and activation memory, reduced colitis, regulation of chronic intestinal inflammation, enhanced the of activity lymphocyte and improve function of immune system in human and protective of liver inflammation (Kim *et al.*, 2010; Singh *et al.*, 2015).

Ferulic acid was recorded 8.94, 21.62 and 25.17 μ g 100 g⁻¹ in T₂, T₃ and T₄ treatments, respectively. The highest ratio was recorded in T₄ treatment. While, it was absent in T₁ and T₅ (control) treatments. Ferulic acid (4-hydroxy-3-methoxy cinnamic acid) is a phenolic compound, it contains carboxylic acid group which it binds to the lipid bilayer to provide some protection against lipid peroxidation. Ferulic acid exhibits anticarcinogenic effect especially colon and skin carcinogenesisby polycyclic aromatic hydrocarbon (Srinivasan *et al.*, 2007; Batista, 2014).

Sinapic acid was recorded 3.49, 4.50 and 5.57 μ g 100 g⁻¹ in T₂, T₃ and T₄ treatments, respectively. The highest ratio was recorded in T₄ treatment. It was not found in T₁ and T₅ treatments. Sinapic acid (3,5-dimethoxy-4-hydroxycinnamic acid) is a bioactive phenolic acid and has the potential to attenuate various chemically induced toxicities and widespread in the plant kingdom (citrus and berry fruits, vegetables, cereal grains, oilseed crops, and some spices and medicinal plants) and as such is common in the human diet. It is known to exhibit antioxidant, anti-inflammatory, anticancer, antimutagenic, antiglycemic,

neuroprotective, anti-anxiety activity and antibacterial activities. In another study, sinapic acid was reported to have the potential to selectively kill the pathogenic bacteria leaving beneficial lactic acid bacteria alive. For these reasons they have been suggested this compound for potential use in food processing, cosmetics and the pharmaceutical industry (Nićiforović and Abramovič, 2014; Chen, 2016).

ρ-Coumaric was recorded 15.32, 22.78 and 30.45 μ g 100 g⁻¹ in T₂, T₃ and T₄ treatments, respectively. The highest ratio was recorded in T₄ treatment While, it was absent in T1 and T5 (control) treatments. The functional of ρ-coumaric acid are reducing oxidative stress and inflammatory reactions. It has good potential to be used as a skin-lightening active ingredient in cosmetics (Boo, 2019). Chinese medicine is used ρ-coumaric as medication to treat patients with rheumatoid arthritis (Zhu *et al.*, 2018). Also, it prevents heaptotoxicity and nephrotoxicity by increased antioxidant enzymes and prevent cancer disease by reduce the free radical (Ekinci Akdemir *et al.*, 2017).

Apigenin-7-glucoside was recorded 8.06, 9.31 and 12.81 μ g 100 g⁻¹ in T₂, T₃ and T₄ treatments, respectively. The highest ratio was recorded in T₄ treatment. It was not found in T₁ and T₅ treatments. Apigenin (4,5,7-trihydroxyflavone) is one of the most widely distributed in the plant kingdom and one of the most studied phenolics. Apigenin is present principally as glycosylated in significant amount in vegetables (parsley, celery, and onions), fruits (orange), herbs (chamomile, thyme, oregano and basil) and plant-based beverages (tea). It is effects in diabetes, amnesia and Alzheimer's disease, obesity disease Parkinson's disease, depression and insomnia, anti-inflammatory and antioxidant function (Hadrich and Sayadi, 2018; Salehi *et al.*, 2019).

Rosmarinic acid was recorded 3.85, 5.52 and 6.24 μ g 100 g⁻¹ in T₂, T₃ and T₄ treatments, respectively. The highest ratio was recorded in T₄ treatment. It was not found in T₁ and T₅ treatments. Alagawany *et al.* (2017) and Taram *et al.* (2018) stated that rosmarinic acid is an ester of caffeic acid and 3,4-dihydroxyphenylactic acid and has a number of beneficial biological activities, commonly used as an herbal spice in food and it had a positive effect treatment on headaches, stomach problems, antimicrobial, immunomodulatory, antidiabetic, anti-allergic, anti-inflammatory, hepato- and renal protectant, Alzheimer's disease and beneficial effects during skin affections.

Cinnamic acid was recorded 3.90, 10.90 and 15.63 μ g 100 g⁻¹ in T₂, T₃ and T₄ treatments, respectively. The highest ratio in T₄ treatment. It was not found in T₁ and T₅ treatments. Cinnamic acid is antimicrobial natural preparations involving cinnamon. cinnamic molecules related with medicinal application, for example on anticancer, antituberculosis, antimalarial, antifungal,

antimicrobial, antiatherogenic and antioxidant activities and it was found to be much more active against the tuberculosis causing bacterium (*Mycobacterium tuberculosis*) (Guzman, 2014).

Determination of phenolic compounds and antioxidant scavenging activity

Substitution of milk fat with CPS increased the total phenolic compounds and antioxidant scavenging activity this increase was proportional to the replacement ratio as compared with the control FY (T₁ and T₅) treatments. Total phenolic compounds and antioxidant scavenging activity of carrot frozen yoghurt significantly ($p \le 0.05$) increased with the increase of the concentration of CPS added and fermentation time in CFY. Therefore, T₄ treatment recorded was highest levels of total phenolic compounds and antioxidant scavenging activity and total phenol contents. Thus, using CPS could improve total phenolic compounds and antioxidant scavenging activity of FY (Figs. 9 a, b). During freezing storage period at -18±1°C total phenolic compounds and antioxidant scavenging activity of fresh samples of all treatments with CPS and control tended to be decreased. Similar findings were reported by Rizk et al. (2014) and El-kholy (2018) for ice cream made with added tomato peel, cactus pear pulp and powder doum fruit as a natural antioxidant source

Bacteriological properties of carrot frozen yoghurt Total viable lactic acid bacterial count

The total viable LAB count in control (T_1 and T_5) treatments was significantly (p \leq 0.05) higher than that recorded in the carrot frozen yoghurt (T_2 , T_3 and T_4) treatments (Fig. 10). The redaction of total LAB count in the CFY treatments may be due to the addition of CPS. These results are in agreement with Aly *et al.* (2004) who stated that the counts of LAB markedly decreased with adding carrot juice to yoghurt.

During freezing storage period, the counts of LAB significantly ($p \le 0.05$) decreased gradually in all treatments. This decrease may be due to the destructive of low temperature in the bacterial cells. Furthermore, this decrease may due to the residual oxygen from the air incorporated during overrun process (Ordonez *et al.*, 2000).

In compare with the LAB counts of the carrot frozen yoghurt mix, the counts of LAB in all CFY treatments decreased from 7.0-8.0 to 5.0-7.0 Log cfu g^{-1} during 45 days of storage period (Table 4 and Fig.10).

Bakirci and Kavaz (2008) and El-Kholy (2018) reported that the total bacterial counts of frozen yoghurt have been decreased during storage period at $-18\pm1^{\circ}$ C, it could be attributed to the effect of fruits and vegetables addition. Furthermore, the high content of phytochemicals may be decrease the total bacterial count (Matter *et al.*, 2016).



Fig. (9 a, b): Effect of using different percentages of carrot puree solids on (a) the antioxidant scavenging activity and (b) total phenol content of carrot frozen yoghurt during storage period



Fig. (10): Effect of using different percentages of CPS in the total viable LAB count of carrot frozen yoghurt

Sensory properties of carrot frozen yoghurt

It was clear that FY made with addition different ratios of CPS (T_2 , T_3 and T_4) treatments gained significantly higher (p ≤ 0.05) overall acceptability scores more than control frozen yoghurt (T_1 and T_5) treatments. The highest total acceptable scores and organoleptic characteristics were observed in the highest ratios of CPS (T_4) added to frozen yoghurt except for flavour whereas T_3 treatment recorded highest flavour score (data are not shown). The addition of CPS improved body, texture, colour and overall acceptability for the treatments which achieved the highest total scores compared with control FY treatments. These results are in agreement with Agarwal and Prasad (2013).

The overall acceptability scores of all carrot frozen yoghurt treatments decreased significantly ($p\leq 0.05$) up to 30 days of freezing storage period. These

results are in agreement with Agarwal and Prasad (2013).

The addition of CPS to carrot frozen yoghurt increased mouth feel thickness, tended to be creamier than control FY (T_1 and T_5) treatments. It is concluded that the low-fat frozen yoghurt containing 3% CPS and 1% fat (T_4 treatment) had the highest organoleptic and nutritional characteristics compared with the other treatments.

Cost production of carrot frozen yoghurt

Full fat frozen yoghurt had the highest cost of production but low-fat frozen yoghurt had the lowest cost of production. T₅ had recorded 26.11% reduction of cost production. Using CPS at ratio 1, 2 and 3% in low fat frozen yoghurt decreased the production cost with 7.52, 15.12 and 22.51%, respectively as compared with T₁ (Table 7) because of its cheap price of carrots.

Table ((7): Effect	of using	different	ratios of	CPS in	n the cos	st production	of carrot	frozen	yoghurt
		0								J U

Itoms	Treatments								
Items	T ₁	T ₂	T ₃	T ₄	T ₅				
Cost production (L.E)	1247.72	1153.93	1059.08	966.88	921.90				
% Reduction of the cost compared with full fat one		7.52	15.12	22.51	26.11				

CONCLUSION

Carrot (Dascus carota L.) vegetable can be used as a functional ingredient in frozen yoghurt due to its carotenoids, natural antioxidant scavenging activity, fibers content in addition to its nutritive constituent and health promoting characteristics. The carrot puree had unique characteristics and attractive colour which make it suitable to be applied in frozen yoghurt making. Carrot puree solids improved rheological characteristics, antioxidant scavenging activity and the total acceptability of the functional low-fat frozen yoghurt. Therefore, carrot puree solids can be used successfully in making low-fat frozen yoghurt to give highest overall acceptability comparable with that full fat frozen voghurt except flavour. The addition of 3% carrot puree solids was the best treatment except for the flavour results. So, we can recommend to enhance its flavour by adding fruits for improve the flavour of resultant frozen yoghurt.

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اليوجهورت المجمد الوظيفي قليل الدسم مع بوريه الجزر

خلود إبراهيم محمد بلاسي ، مجدي محمد عبد المنعم محمد عثمان ، فوزي محمد عباس ، نوال جلال عبد الحليم محمد قسم الألبان، كلية الزراعة، جامعة قناة السويس، الإسماعيلية، مصر

تم دراسة تأثير إضافة جوامد الجزر الكلية بنسب ١ و٢ و٣% من تركيب المخلوط المستخدم على الخواص الفيزيائية والكيميائية والمركبات الفينولية الكلية ونشاط المواد المضادة للأكسدة والعدد الكلى لبكتريا حامض اللاكتيك الحية والخصائص الحسية وتكلفة إنتاج اليوجهورت المجمد منخفض الدهن. حيث يتميز الجزر بأنه مصدر للألياف الغذائية وغني بالمواد المضادة للأكسدة الطبيعية مما يؤهله للاستخدام لتدعيم اليوجهورت منخفض الدهن. أوضحت النتائج إن إضافة جوامد الجزر الكلية أدت إلى زيادة كلاً من الوزن النوعي، الوزن بالجالون، اللزوجة الظاهرية، المقاومة للانصهار، المركبات الفينولية الكلية ونشاط المواد المضادة للأكسدة مقارنة بعينة الكنترول بالجالون، اللزوجة الظاهرية، المقاومة للانصهار، المركبات الفينولية الكلية ونشاط المواد المضادة للأكسدة في المنتج كاملة الدهن ومنخفضة الدهن. كما أظهرت النتائج انخفاض في كلاً من نقطة التجمد، الربع مقارنة بالكنترول كامل الدسم. يوامد الجزر الكلية أدت إلى خفض العدد الكلى لبكتريا حمض اللاكتيك الحية في الزبادي المجمد خلال التخرين المجمد. ووامد الجزر الكلية أدت إلى خفض العد الكلى لبكتريا حمض اللاكتيك الحية في الزبادي المجمد خلال التخزين المجمد. والم الجزر الكلية أدت إلى خفض العد الكلى لبكتريا حمض اللاكتيك الحية في الزبادي المجمد خلال التخرين المجمد. والحسية فيما حد الكلي المرتيات عمض اللاكتيك الحية في الزبادي المجمد خلال التخزين المجمد. والحسية فيما عدا الطعم مع خفض تكلفة الإنتاج بنسبة تصل إلى 22.51 %. بينما إضافة البوريه بنسبة ٢% من تركيب المخلوط الكلى أعطت أفضل النتائج بالنسبة للطعم.