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Properties of Milk Permeate Synbiotic Beverages Enriched with Carrot Juice Barley and Oat Flour

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THIS research was conducted to producing new healthy beverages from permeate as a dairy byproduct.Milk permeate was fermented with *Streptococcus thermophilus*, *Lactobacillus acidophilus* and *Bifidobacterium* BB-12along withcarrot juice, white flour of oat and barleyto compare the suitability of these substrates for the production of a novelsynbiotic beverages. Physicochemical, bacteriological, rheological,sensory properties andfunctional components (antioxidants and total phenolics) were investigated during storage period at4°C for 15 days.A viable cell countthroughout the product shelf-life was above the minimum count required in a probiotic product (more than log 6 CFU/g). The highest cell count was observed in treatments fortified withcarrot juice and oat flour (68% milk permeate + 2% oat flour+30% carrot juice). Supplementations of milk permeate with carrot juice, oat and barley flour improved the flavour, texture and overall acceptability of milk permeate synbiotic beverages. The overall results cleared that, it is possible to produce high-quality fermented milk permeate with good flavour, body,texture and appearance by adding carrot juice and oat flour (66% milk permeate + 4% oat flour + 30% carrot juice).

Keywords: Milk permeate, Probiotic, Antioxidants, Phenolic components

Introduction

Dairy industries arepresented throughout the world and produce many products, generating solid and liquid wastes. However, fast industrial growth does not only increase productivity, it also resulted in increased release of toxic substances in land or water reservoirs. This release results in destructionof environment and becomes a reason for serious health hazards for humans. These wastes can be utilized effectively as raw material for the production of other industrial products (Wong et al., 2018).

Food matrices that do not offer the best environment to ensure probiotic cell viability can be enhanced by a synbiotic combination (Fernandes Pereira and Rodrigues, 2018). The food industry must deal with many challenges, including the increasing consumer awareness and demands for more nutritious and safer food (Vita et al., 2019)

Milk permeate, which penetrate the membrane during UF process of milk has been regarded as waste product, although it contains high level of lactose, soluble proteins, vitamins and minerals. Milk permeate can be considered as a solution of nutritious significance. In this respect,cereals contain water-soluble fiber, oligosaccharides and resistant starch; have been suggested to fulfill the prebiotic concept. Cereals such as barley and oat are an important source of protein, carbohydrates, vitamins, minerals and fiber for people all over the world, and can be used as sources of non-digestible carbohydrates that besides promoting several beneficial physiological effects can also selectively stimulate the growth of *Lactobacilli* and *Bifidobacteria* present in the colon, thereby

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acting as prebiotics (Charalampopoulos et al., 2002).

Carrot is comprised with many functional food components such as vitamins (A, C, D, and K) minerals (calcium, potassium, phosphorus, sodium, and iron) and prebiotic compounds (fructo-oligosaccharide and inulin) (Alwis et al., 2016).

The term "synbiotic" is to describe a combination of synergistically acting probiotics and prebiotics. A selected component introduced to the gastrointestinal tract should selectively stimulate growth and/or activate the metabolism of a physiological intestinal microbiota,thus conferring beneficial effect to the host's health (Skalkam et al., 2016). Among probiotic and synbiotic functional foods, dairy products, especially fermented milks, are market leaders and are considered research priorities in many countries. The growing appreciation of functional foods around the world is promoting innovations in food products and stimulating the consumption of foods with nutritional and therapeutic value (Kumar et al., 2015).

The present work was carried out tostudy thebacteriological, physicochemical, sensory properties and functional properties of milk permeate synbiotic beverages enriched with carrot juice, barley and oat flour

Materials and Methods

Materials

White flour of barley and oat were obtained from King M for industrial foods company, Badr city, Egypt.The whole flourswere obtained by milling in a hammer mill (Falling Number AB, England) fitted with a sieve of 0.02 inch aperture size, whereas bran and white flour were obtained by combined debranning and dry milling using the Satake STR-100 mill.Permeate was obtained from Nestlé Egypt Company, 6th of October, Egypt. It is a by-product from the UF cow's skim milk. It was prepared at 50°C using spiral-wound module membrane supplied by APV pasilac, Denmark. The milk permeate was immediately heated in a water bath at 80°C for 10 min and cooled, and kept frozen at -20°C until use.

Freeze dried DVS ABT-5 Probio-Tec®culturescontaining *Streptococcus thermophilus*, *Lactobacillus acidophilus* and *Bifidobacterium* BB-12 were obtained from Christian Hansen Laboratory Copenhagen, Denmark, by Misr Food Additives (MIFAD), Egypt.Stabilizer (Dairy gel 3095) containing Guar gum E412, sodium carboxymethyl cellulose E466 and mono and diglyceride of fatty acid E471 (1:1:1) was obtained from the Egyptian Company for Dairy Products and Food Additives ̎EGY-DAIRY ̎ (10th of Ramadan city, Egypt).

Methods

Before treatments, good quality fresh carrot (*Daucus carota*) free from mold and insect contamination) were manually selected. After washing and peeling, fresh carrot juice was extracted using carrot juice machine (Braun AG Frankfurt Juice Extractor MP50, Germany). The juice was prepared and used in manufacture directly.The chemical composition, antioxidant activity, total phenolics and nutritional composition of raw materials used for manufacture of the synbiotic beverages are presented in Table 1.

Preparation of synbiotic beverages

Nine different formulations of fermented beverages were prepared (Table 1). All treatments were added stabilizer at 0.5%, homogenized at 60ºC, 600 Kpa and heated to 72ºC for 15sec, then rapidly cooled to 42ºC. Starter culture (0.05% direct vat set) was added, and distributed in sterile glass bottles (200 mL). All treatments were incubated at 42ºC until a pH 5.5 was reached. The synbiotic beverages of different treatments were stored at 4±1°C. Samples were taken fresh and then after 5, 10 and 15 day for examinations.

Chemical analyses

Total solids, fat, total protein (TN) contents, titratable acidity and dietary fiber of samples were determined according to AOAC (2007). The changes in pH values were measured using a laboratory pH meter with glass electrode (HANNA, Instrument, Portugal). Total volatile fatty acids (TVFA) were estimated according to Kosikowski (1984). Carbohydrate content was calculated by difference TS- (fat + protein+ ash) according to Guzmán-González et al. (1999). The caloric value was calculated by the following equation given by Chandan (2011), Calories factors (kcal/100 mL of product) = (protein × 4.27) + (fat × 8.79) + (carbohydrate × 3.87).

Determination of total phenolic content and antioxidant activity

Water extract of synbiotic beverages (10 g) was mixed with 2.5 mL distilled water andpH was adjusted to 4.0 using 1 M HCl. The beverages were then incubated in water bath (45ºC) for 10

TABLE1. Chemical composition, nutritional value of raw materials used in preparingsynbiotic beverages and Ingredients (Kg/100 Kg beverage) for synbiotic beverages

Component	Milk permeate	Carrot juice	Barley flour	Oat flour
Total solids %	5.16±0.36[c]	17.50±2.50[b]	90.41±4.32[a]	91.94±5.44[a]
Protein %	0.18±0.12[b]	0.70±0.62[b]	11.3±2.10[a]	9.73±2.94[a]
Fat %	0.09±0.04[c]	0.35±0.12[c]	2.70±0.82[b]	8.20±2.08[a]
Ash %	0.31±0.03[c]	0.52±0.60[c]	1.95±1.14[b]	3.14±1.48[a]
Total dietary fibers%	-	1.81±0.48[c]	2.94±1.34[b]	4.55±2.08[a]
Carbohydrates %	4.58±0.32[d]	14.12±1.74[c]	71.52±2.54[a]	66.32±2.08[b]
Calories (kcal/100g)	19.28±2.16[d]	60.71±4.36[c]	348.77±6.96[b]	370.28±11.94[a]
Total phenolics (mg/100g)	3.30±1.90[d]	46.52±5.47[c]	187.03±6.21[b]	94.07±5.98[a]
Antioxidant activity %	11.70±4.25[c]	35.30±5.23[b]	73.25±11.25[a]	66.28±8.64[a]
pH	6.43±0.16[a]	6.15±0.21[b]	-	-

Ingredients (Kg/100Kg beverage) for symbiotic beverage

%	Treatment								
	C	**T1**	**T2**	**T3**	**T4**	**T5**	**T6**	**T7**	**T8**
Milk permeate	100	98.00	68.00	98.00	68.00	96.00	66.00	96.00	66.00
Carrot juice	-	-	30	-	30.00	-	30	96.00	30
Barley flour	-	2.00	2.00	-	-	4.00	4.00	-	-
Oat flour	-	-	-	2.00	2.00	-	-	-	4.00

Mean (±SE). Values with small letters in the same row having different superscripts differ significantly ($p \leq 0.05$).
C: control (100% milk permeate)
T1: 98% milk permeate + 2% barley flour
T2: 68% milk permeate + 2% barley flour and 30% carrot juice
T3: 98% milk permeate +2% oat flour
T4: 68% milk permeate +2% oat flour + 30% carrot juice
T5: 96% milk permeate + 4% barley flour
T6: 66% milk permeate + 4% barley flour + 30% carrot juice
T7: 96% milk permeate + 4% oat flour
T8: 66% milk permeate + 4% oat flour+ 30% carrot juice.

min and the precipitated proteins were removed by centrifugation (10,000 rpm, 10 min, 4ºC). The supernatant was harvested and the pH was adjusted to 7.0 using NaOH (0.5 M) followed by another centrifugation (10,000 rpm, 10 min, 4ºC) to remove residual precipitated proteins and salts. The supernatant was harvested, kept refrigerated and used for subsequent analysis within 24 hr.

The total phenolic content was determined as described by Skerget et al. (2005) Absorbance at 725 nm was converted to total phenolics expressed in micrograms equivalents of gallic acid per gram (µg GAE/g) sample. Standard curves were established using various concentrations of gallic acid (5-60 µg/mL) in methanol.

Antioxidant activity as radical scavenging activity (RSA) was measured by bleaching of the purple coloured solution of 1,1-Diphenyl-2-picrylhydrazyl (DPPH) according to the method of Hatano et al. (1989). The absorbance was determined against a control at 517 nm Gülçin et al. (2004). Percentage of antioxidant activity of DPPH was calculated as follows:

$$\text{DPPH scavenging activity (\%)} = [(A0- A1)/ A0] \times 100$$

Where, A0 is the absorbance of the control reaction and A1 is the absorbance in the extract. Samples were analyzed in triplicate.

Flavour Compounds

Acetaldehyde and diacetyl insynbiotic beverages were determined as described by Less &Jago (1969). Acetaldehyde reacts with semi-carbazide to form semi-carbazone which has absorption value at wave length of 224 nm. Meanwhile diacetyl has an absorption value at wave length of 270 nm.

Rheological Measurements

Viscosity of synbiotic beverages samples was determined by the method described by Aryana (2003) using Rotational Viscometer Type Lab. Line Model 5437. Results were expressed as (mPas). Syneresis was determined by centrifugation. The amount of serum discharged after centrifugation (2000 rpm min^{-1}, 20 min, 20 g of sample) was measured and expressed as % (Liutkevičius et al., 2016).

Microbiological analysis

Microbiological analyses wereperformed on fresh samples and after 5, 10 and 15 days of cold storage. The enumeration of *S. thermophilus* was accomplished after incubation at 37ºC for 48 hr, under anaerobic conditions using M17 agar (Oxide Ltd). *L. acidophilus* counts were determined using MRS-sorbitol agar (Oxide Ltd), and the plates were incubated in anaerobic conditions at 37ºC for 72 hr (Dave & Shah 1996). *B.* BB-12 counts were enumerated according to Dinakar & Mistry (1994) using a modified MRS agar media (a mixture of antibiotics, including 2 g of neomycin sulphate, 0.3 g of nalidixic acid, 4 g of paromomycin sulphate, and 60 g of lithium chloride (NPNL, Sigma Chemical Co.) prepared in 1000 mL of distilled water, filter-sterilized, and stored at 4ºC until use. The mixture of antibiotics (5 mL) was added to 100 mL of MRS agar media. Cysteine-HCl was added at the rate of 0.05% to decrease the redox potential of the medium, and the plates were incubated in anaerobic conditions at 37ºC for 72 hr. The results were expressed as log colony-forming units per gram (log cfu/g) of sample and the viability of each culture in different samples wascalculated according to Paseephol and Sherkat (2009) as follows:

% Viability = (CFU/g after 15 days of storage/initial CFU/g) × 100.

Coliforms, moulds and yeasts were enumerated according to Feng et al. (2002) and ISO(2004), respectively.

Sensory evaluation

Sensory evaluation of synbiotic beverage samples was done by 15 trained panelists. The panelists were asked to evaluateflavour (10 points), colour (10 points), taste (10 points), and palatability (10 points) according to Bodyfelt et al. (1988).

Statistical analysis

The obtained results were evaluated statistically using analysis of variance as reported by McClave & Benson (1991). P value less than 0.05 was considered statistically significant. SPSS (Chicago, IL, USA) software window Version 20c was used.

Results and Discussion

Chemical composition of synbiotic beverages

Chemical composition of symbiotic beverage was followed as shown in Table 2. The milk permeate containing 4% barley flour + 30% carrot juice (T6) and milk permeate containing 4% oat flour + 30% carrot juice (T8) had the highest total solids content (TS) and it was different significantly ($p \leq 0.05$) from other treatments. While the control milk permeate sample exhibited the lowest TS content. The TS content of milk permeate beverages supplemented with oat flour

Table 2 shows that thetotal protein of permeate supplemented with oat and barley flour slightly increased by increasing the percentage added. On the other hand, the total protein content of all treatments insignificant changes throughout the storage periods. These results are in agreement with those obtained by Gupta et al. (2017). Also, milk permeate fortified with oat and barley flouraffectedon the fat content of the resultant product.

Supplementations of milk permeate with oat and barley flour by different concentration increased acidity of treated samples up to the end of storage period. Addition of 30% carrot juice was increment in titratable acidity with increasing the storage period up to 15 days. This increment is mainly due to conversion of lactose to lactic acid by microorganisms of the starter (Aamer et al., 2017).

Changes in pH value of synbiotic beverages of different treatments were affected by supplementation of carrot juice, oat and barley flour or storage period followed almost opposite trend to acidity. Addition of 30% carrot juice affected the pH values of the functional beverages. It can be noted that with increasing the storage period, the pH values of the prepared functional beverages significantly decreased. This may be due to the increment of acidity as well as the formation of lactic acid and acidic amino acids during the storage period(Aamer et al., 2017).

Total polyphenols and antioxidant activityof synbiotic beverages

The results in Table 2 indicated that synbiotic

TABLE 2. Gross chemical composition of synbiotic beverages during storage at 4ºC for 15 days

Storage period (day)	Treatment								
	C	T1	T2	T3	T4	T5	T6	T7	T8
Total solids									
Fresh	6.21^{d}	7.92^{cd}	11.62^{ab}	7.95^{cd}	11.65^{ab}	9.62^{bc}	13.32^{a}	9.68^{bc}	13.38^{a}
5	6.32^{d}	7.98^{cd}	11.75^{ab}	8.12^{cd}	11.77^{ab}	9.74^{bc}	13.51^{a}	9.75^{bc}	13.52^{a}
10	6.47^{d}	8.09^{cd}	11.89^{ab}	8.19^{cd}	11.91^{ab}	9.87^{bc}	13.75^{a}	9.86^{bc}	13.67^{a}
15	6.53^{e}	8.21^{d}	11.92^{b}	8.32^{d}	11.98^{b}	9.96^{c}	13.97^{a}	9.94^{c}	13.83^{a}
Fat (%)									
Fresh	0.20^{f}	0.25^{ef}	0.33^{cde}	0.36^{cde}	0.44^{bc}	0.30^{def}	0.38^{cd}	0.52^{ab}	0.60^{a}
5	0.22^{f}	0.26^{ef}	0.35^{cde}	0.39^{cd}	0.46^{bc}	0.32d^{ef}	0.41^{cd}	0.54^{ab}	0.62^{a}
10	0.23^{f}	0.27^{ef}	0.36^{de}	0.41^{cd}	0.49^{bc}	0.34^{de}	0.42^{cd}	0.55^{b}	0.65^{a}
15	0.25^{f}	0.30^{ef}	0.40^{de}	0.42^{cd}	0.51^{bc}	0.35^{def}	0.45^{cd}	0.56^{b}	0.66^{a}
Total protein (%)									
Fresh	0.34^{f}	0.66^{de}	0.82^{c}	0.63^{e}	0.79^{cd}	0.89^{bc}	1.04^{a}	0.77^{cd}	0.98^{ab}
5	0.36^{f}	0.71^{de}	0.84^{cd}	0.65^{e}	0.82^{cd}	0.92^{bc}	1.11^{a}	0.81^{cd}	1.03^{ab}
10	0.38^{f}	0.73^{de}	0.85^{cd}	0.67^{e}	0.83^{cd}e	0.95^{bc}	1.21^{a}	0.84^{cde}	1.09^{ab}
15	0.41^{e}	0.75^{d}	0.87^{c}	0.72^{d}	0.86^{c}	0.98^{b}	1.30^{a}	0.86^{c}	1.21^{a}
pH value									
Fresh	5.48^{a}	5.35^{a}	5.32^{a}	5.35^{a}	5.30^{a}	5.35^{a}	5.34^{a}	5.35^{a}	5.33^{a}
5	5.43^{a}	5.26^{ab}	5.15^{bc}	5.20^{abc}	5.10^{c}	5.22^{abc}	5.08^{c}	5.14^{bc}	5.18^{abc}
10	5.31^{a}	5.04^{b}	5.04^{b}	5.03^{b}	4.98^{b}	5.07^{ab}	5.06^{ab}	5.07^{ab}	5.05^{ab}
15	5.23^{a}	5.01^{ab}	4.98^{ab}	4.97^{ab}	4.86^{b}	5.01^{ab}	4.98^{ab}	4.96^{ab}	4.95^{ab}
Titratable acidity (lactic acid %)									
Fresh	0.56^{d}	0.59^{cd}	0.6۷^{abc}	0.58^{d}	0.6۸^{ab}	0.56^{d}	0.67^{abc}	0.60^{bcd}	0.70^{a}
5	0.59^{d}	0.62^{cd}	0.7۲^{abc}	0.66^{bcd}	0.74^{ab}	0.65^{bcd}	0.72^{abc}	0.70^{abc}	0.77^{a}
10	0.65^{d}	0.70^{cd}	0.86^{ab}	0.78^{a}	0.90^{a}	0.74^{bcd}	0.82^{abc}	0.92^{abc}	0.89^{a}
15	0.66^{e}	0.82^{e}	1.09^{ab}	0.90^{d}	1.14^{a}	0.98^{c}	1.04^{bc}	1.0۵^{bc}	1.0۸^{ab}
Total phenolic content (mg/100g)									
Fresh	4.23^{d}	80.58^{c}	96.55^{bc}	85.76^{bc}	79.24^{c}	104.34^{ab}	120.31^{a}	82.68^{c}	90.32^{bc}
5	3.28^{e}	74.82^{d}	90.3^{6b}	79.83^{cd}	74.10^{cd}	95.42^{b}	112.46^{a}	77.52^{cd}	85.28^{bc}
10	2.25^{f}	67.42^{e}	84.62^{bc}	74.25^{de}	68.66^{e}	90.20^{b}	100.04^{a}	72.33^{de}	80.14^{cd}
15	2.13^{f}	63.28^{e}	77.64^{bc}	69.59^{cde}	62.02^{e}	84.18^{b}	96.32^{a}	67.28^{de}	73.61^{cd}
Antioxidant activity (Radical Scavenging Activity %)									
Fresh	15.25^{b}	35.54^{a}	38.95^{a}	38.57^{a}	33.45^{a}	42.24^{a}	46.31^{a}	35.75^{a}	42.47^{a}
5	12.54^{c}	32.35^{b}	35.66^{ab}	35.86^{ab}	30.97^{b}	40.61^{a}	41.23^{a}	31.22^{b}	38.65^{ab}
10	10.32^{c}	29.88^{ab}	32.27^{ab}	32.62^{ab}	27.31^{b}	36.55^{ab}	38.52^{a}	27.87^{b}	35.48^{ab}
15	6.46^{d}	26.32^{bc}	28.45^{abc}	29.34^{abc}	23.53^{c}	32.86^{ab}	35.47^{a}	24.65^{bc}	31.24^{abc}

Means with small letters in the same row having different superscripts differ significantly (p □ 0.05).

C: control (100% milk permeate)

T1: 98% milk permeate + 2% barley flour

T2: 68% milk permeate + 2% barley flour and 30% carrot juice

T3: 98% milk permeate +2% oat flour

T4: 68% milk permeate +2% oat flour + 30% carrot juice

T5: 96% milk permeate + 4% barley flour

T6: 66% milk permeate + 4% barley flour + 30% carrot juice

T7: 96% milk permeate + 4% oat flour

T8: 66% milk permeate + 4% oat flour+ 30% carrot juice.

beverage containing 4%barley flour and 30% carrot juice (T6) had higher total polyphenols and antioxidant activity than the other samples. Theresults also indicated that a gradual decrement in total polyphenols and antioxidant activity was noted after 15 days of storage. Sreerupa et al. (2014) mentioned that fruit juices contained less amount of antioxidant after 28 days of storage than that found in the fresh ones. From the previous results, it can be concluded that the addition of carrot juice to milk permeate beverage can improve its content of phenolic compounds which play an important role as an antioxidant that had high antioxidant activity (Aamer et al., 2017).

Addition of carrot juice,barley and oat flour products increased both the total phenolic content and antioxidant activity compared with the control samples. The probiotic bacteria (e.g. *Bifidobacterium* spp.) hydrolyze polyphenols to aromatic acids like phenyl acetic, phenyl propionic, phenyl valeric and benzoic acids (Manach et al., 2004). Addition of carrot products improved the content of total phenolic and enhanced the antioxidant activity (Scarano et al., 2018).

Functional food is a term that is being used to define the foods which provide bioactive natural compounds to human diet beyond their basic nutrient supplying characteristics. These bioactive compounds are often subject to health- benefiting and disease preventing claims (Kris-Etherton et al., 2004). Therefore, researchers intensified their studies on the development of foods fortified with vitamins, fibers, fatty acids and the other various natural compounds to give them novel functional characteristics. Phenolics represent a significant part of natural compounds with the currently known over 8000 distinct members (Bravo,1998) which contribute to formation of sensory

TABLE 3. Flavour volatile compounds, syneresis, viscosity, total polyphenols and antioxidantactivity of synbiotic beverages during storage

Storage period (day) C	Treatments								
	T1	T2	T3	T4	T5	T6	T7	T8	
Acetaldehyde µg/g									
Fresh	4.05^{g}	9.25^{f}	24.17^{cd}	14.21^{e}	35.14^{a}	12.24^{ef}	27.13^{bc}	20.31^{d}	29.13^{b}
5	10.19^{e}	18.11^{d}	32.34^{bc}	23.33^{d}	44.83^{a}	20.14^{d}	34.05^{bc}	30.10^{c}	37.15^{b}
10	9.13^{f}	15.25^{e}	26.19^{c}	20.14^{d}	34.52^{a}	17.09^{de}	28.11^{bc}	25.17^{c}	31.27^{ab}
15	7.76^{f}	13.31^{e}	17.22^{cde}	15.18^{de}	28.76^{a}	16.18^{cde}	20.18^{c}	24.21^{b}	19.09^{cd}
Diacetyl µg/g									
Fresh	170.08^{f}	250.14^{ef}	400.18^{bc}	320.14^{d}	500.43^{a}	300.13^{de}	450.24^{ab}	350.54^{cd}	470.51^{a}
5	210.17^{f}	330.24^{ef}	430.80^{cd}	400.27^{d}	600.81^{a}	380.27^{de}	480.38^{bc}	440.15^{bc}	510.11^{b}
10	220.21^{g}	380.11^{f}	540.41^{bc}	460.11^{de}	600.45^{ab}	420.24^{ef}	580.12^{ab}	500.24^{cd}	620.13^{a}
15	250.11^{f}	300.19^{e}	480.15^{b}	410.24^{cd}	580.29^{a}	360.11^{d}	540.28^{a}	450.11^{bc}	590.22^{a}
TVFA (0.1 N of NaOH/100 g)									
Fresh	1.20^{f}	2.12^{ef}	4.92^{bc}	3.26^{de}	6.50^{a}	2.50^{def}	5.30^{ab}	3.70^{cd}	6.06^{ab}
5	2.80^{f}	4.32^{e}	6.12^{cd}	5.92^{d}	8.14^{a}	5.80^{d}	7.65^{ab}	6.30^{bcd}	7.55^{abc}
10	3.90^{c}	6.12^{bc}	7.42^{ab}	7.18^{ab}	9.32^{a}	6.96^{ab}	8.26^{ab}	7.98^{ab}	8.48^{ab}
15	5.20^{b}	7.65^{a}	9.35^{a}	8.20^{a}	10.24^{a}	8.04^{a}	9.70^{a}	8.66^{a}	9.86^{a}

Means with small letters in the same row having different superscripts differ significantly ($p \leq 0.05$).
C: control (100% milk permeate)
T1: 98% milk permeate + 2% barley flour
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T6: 66% milk permeate + 4% barley flour + 30% carrot juice
T7: 96% milk permeate + 4% oat flour
T8: 66% milk permeate + 4% oat flour+ 30% carrot juice.

(organoleptic properties) and dietary attributes of food products. Phenolics have attracted much attention of food and health scientists in recent years due to their free radical scavenging capacity, and antioxidant power (Macheix et al.,1990). Reactive oxygen species (ROS) are continuously produced in cells and they are considered as to be the main source of oxidative stress linked to structural cell damage (Apel & Hirt, 2004) and phenolics are being recognized by their ROS scavenging ability (Hatano et al.,1989). Thus, enrichment of foods with phenolics can be a promising strategy to produce functional foods exhibiting higher antioxidant activity.

Flavour compounds of synbiotic beverages

Table 3 shows the effect of supplementation ofpermeate beveragewith barley and oat flour especially at higher concentration on flavour compounds. It was found a decreasing in acetaldehyde values during the storage period, also diacetyle values increased up to 5 days for storage and then decreased until the end of storage period. This may be due to the ability of lactic organisms to hydrolysis acetaldehyde and diacetyle to acetone and other compounds (Castro et al., 2013). Total volatile fatty acids (TVFA) content (Table 4) increased in all treatments of fermented milk permeate beverages during storage periods. This could be attributed to proteolytic and lipolytic action of starter cultures during making and storage of the products. However, oat flour had higher effect than barley flour. Similar results were observed by Castro et al. (2013).

Rheological properties ofsynbiotic beverages

The supplementation of fermented milk permeate beverages withcarrot juice,barley and oat flour reduced significantly whey syneresis compared with fermented milk permeate beverages without additives (C). Itsreduction was proportional to the supplementation ratio (Table 4). These results might be due to increasing the water holding capacity by dietary fiber in the resultant products. The supplementation of fermented milk permeate beverages with carrot juice, barley and oat flour decreased significantly whey syneresis compared with controlmilk permeate beverages (C). These results may be due to increasing the water holding capacity brought by dietary fiber in the resultant curd (Akpinar et al. 2015).

TABLE 4. Syneresis and viscosity of synbiotic beverages during storage

Storage period (day)	Treatments								
	C	T1	T2	T3	T4	T5	T6	T7	T8
				Syneresis %					
Fresh	91.7^{a}	80.6^{b}	77.6^{bc}	74.6^{bcd}	70.5^{cdef}	71.4^{cde}	64.4^{ef}	65.9^{def}	61.7^{f}
5	93.2^{a}	82.3^{bc}	87.3^{ab}	78.6^{cd}	83.6^{bc}	80.5^{bcd}	85.5^{bc}	70.4^{e}	75.4^{de}
10	95.6^{a}	88.5^{ab}	90.5^{ab}	85.2^{bcd}	87.2^{bc}	85.4^{bcd}	87.4^{bc}	78.5^{d}	80.6^{cd}
15	96.8^{a}	90.4^{abc}	93.4^{ab}	87.6^{abc}	87.6^{abc}	88.2^{abc}	91.2^{abc}	82.3^{c}	85.7^{bc}
				Viscosity (mPa.s)					
Fresh	92.25^{f}	190.37^{e}	198.36^{de}	198.65^{de}	205.65^{cde}	212.21^{bcd}	230.55^{ab}	218.85^{abc}	237.22^{a}
5	113.46^{b}	230.64^{a}	234.64^{a}	245.24^{a}	249.13^{a}	262.50^{a}	286.84^{a}	266.26^{a}	292.51^{a}
10	101.61^{b}	250.42^{a}	258.48^{a}	268.37^{a}	276.38^{a}	274.64^{a}	294.47^{a}	282.64^{a}	302.38^{a}
15	81.13^{c}	160.58^{b}	167.54^{ab}	166.41^{ab}	172.77^{ab}	172.87^{ab}	198.24^{ab}	179.11^{ab}	204.58^{a}

Means with small letters in the same row having different superscripts differ significantly (p □ 0.05).

C: control (100% milk permeate)

T1: 98% milk permeate + 2% barley flour

T2: 68% milk permeate + 2% barley flour and 30% carrot juice

T3: 98% milk permeate +2% oat flour

T4: 68% milk permeate +2% oat flour + 30% carrot juice

T5: 96% milk permeate + 4% barley flour

T6: 66% milk permeate + 4% barley flour + 30% carrot juice

T7: 96% milk permeate + 4% oat flour

T8: 66% milk permeate + 4% oat flour+ 30% carrot juice.

Control fermented milk permeate beverages (C) had lower significantly viscosity compared with supplementation fermented milk permeate beverages. Fermented milk permeate beverages with oat flour increased significantly ($P \leq 0.05$) viscosity of the resultant milk permeate beverages. The increase was proportional to the level of addition. This increase could be attributed to the water hydration of oat and barley flour (Akpinar et al., 2015).

The viscosity decreased ($P \leq 0.05$) with the advance of storage period for all the treatments after 10 days compared 5 days of control beverage (C). This decrease may be explained by the bacterial enzyme action on the casein micelle matrix over time (Kosikowski, 1984). Aryana & McGrew (2007) observed gradual reduction in the apparent viscosity of probiotic yoghurt, but interaction between time interval and treatments was non-significant possibly due the activity of bacteria enzymes on the matrix of casein micelle over the time.

Microbiological evaluation ofsynbiotic beverages

Table 5 shows that *Streptococcus thermophilus* and *Lactobacillus acidophilus* counts increased gradually in all treatments up to 10 days of storage and then decreased at the end of storage period. Beverage treatments supplemented with oat flour had the highest *Streptococcus thermophilus* and *Lactobacillus acidophilus* counts. *Bifidobacterium* BB12counts increased gradually in all treatments up to the end of storage period. Beverage treatments supplemented with oat flour had the highest *Bifidobacterium* BB12 counts. The addition of oat and barley flour improved the viability of *Streptococcus thermophilus, Lactobacillus acidophilus* and *Bifidobacterium* BB12. Also, the addition of carrot juice slightly decreases the viability of *Streptococcus thermophilus, Lactobacillus acidophilus* and *Bifidobacterium* BB12 compared with treatments without carrot juice. Similar results were reported by other researchers concerning the viability and survival of *L. acidophilus* and other lactobacilli in fruit-whey beverages Sady et al. (2017). Elsanhoty and Ramadan (2018) found that the supplementation of probiotic low fat yoghurt with barley β-glucan enhanced the probiotic viability during storage.Since the starter cultures used in the present study were able to maintain the minimum therapeutic level (>6 log CFU/g) up to 15 days of storage. This improved viability of synbiotic beverages supplemented with carrot juice, oat and barley flour may have been due to the availability of nutrients in the products.

When evaluating the results in Table 5, it can be observed that the synbiotic beverages presented counts of lactic bacteria and *bifidobacterial* compatible with the Technical Regulation of Identity and Quality of Fermented Milks during the storage period. The regulation recommends the total viable *bifidobacterial*bacteria count of 6 log CFU/g of the product. On the case of functional foods, the minimum viable quantity for probiotics should be no less than 6 logsCFU/g in the daily recommendation of the product ready for consumption of 100g/day. These resultsare also in accordance with the amount recommended by several authors so that the probiotic microorganisms produce the desired physiological effect (Terpou et al.,2019).For instance, to assure that, it has been stated that the so called “minimum therapeutic” level of viable probiotic microorganisms should be at least log 6 CFU/g of viable cells throughout the product shelf-life (Neffe-Skocińska et al., 2018).

The coliform, moulds and yeasts were not detected in all samples throughout production and during storage period. This can be attributed to the good hygienic conditions practiced during processing and storage.

Sensory evaluation of synbiotic beverages

Table 6 shows the sensory evaluation of milk permeate synbiotic beverages supplemented with carrot juice, barley and oat flour. The sensory characteristics of milk permeate supplemented with barley and oat flour was markedly improved as compared to control sample. Colour and appearance and flavour of, barley and oat flour supplemented milk permeate sample showed non-significant change, whereas there was observed a significant change in body, mouthfeel and overall acceptability. Sensory evaluation as well as overall acceptability results of the milk permeate samples showed that the all synbiotic milk permeate beverages fortified with carrot juice, oat and barley flour especially T8 (4% oat flour + 30% carrot juice) were the most acceptable sample by judging panel members compared with control milk permeate beverages (C). T8 sample had the highest score among of all samples with respect to colour, appearance, body, mouthfeel and overall acceptability. Addition of carrot juice to milk permeate beverages increased the overall acceptability of the prepared functional beverages. The results agree with (Akpinar et al., 2015 and Aamer et al., 2017).

TABLE 5. Microbiological evaluation (Log cfu/g) of synbiotic beverages during storage.

Storage period (day)	Treatments								
	C	**T1**	**T2**	**T3**	**T4**	**T5**	**T6**	**T7**	**T8**
					Streptococcus thermophiles				
Fresh	5.36^{b}	6.90^{a}	6.85^{a}	7.81^{a}	7.96^{a}	7.38^{a}	7.24^{a}	7.54^{a}	7.58^{a}
5	6.04^{c}	7.23^{ab}	6.90^{bc}	7.86^{ab}	8.07^{a}	7.60^{ab}	7.30^{ab}	7.73^{ab}	7.97^{ab}
10	6.36^{c}	7.50^{ab}	7.12^{b}	7.89^{ab}	8.10^{a}	7.74^{ab}	7.46^{ab}	7.81^{ab}	8.11^{a}
15	4.42^{d}	6.11^{c}	6.34^{bc}	6.83^{ab}	7.39^{a}	6.58^{bc}	6.61^{bc}	6.79^{ab}	6.92^{ab}
Viability%	82.46^{c}	88.55^{ab}	92.55^{ab}	87.45^{b}	92.84^{a}	89.16^{ab}	91.3^{ab}	90.05^{ab}	91.29^{ab}
					Lactobacillus acidophilus				
Fresh	5.95^{c}	6.47^{bc}	6.50^{bc}	7.51^{a}	7.76^{a}	7.27^{ab}	6.98^{ab}	7.62^{a}	7.38^{ab}
5	6.69^{c}	7.14^{abc}	6.8^{bc}	7.60^{ab}	7.80^{a}	7.38^{abc}	7.14^{abc}	7.72^{a}	7.47^{abc}
10	6.07^{b}	7.29^{a}	7.15^{a}	7.81^{a}	7.92^{a}	7.50^{a}	7.20^{a}	7.86^{a}	7.65^{a}
15	5.11^{e}	5.68^{de}	5.94^{cd}	6.68^{abc}	7.11^{a}	6.39^{abcd}	6.12^{bcd}	6.76^{ab}	6.57^{abc}
Viability%	85.88^{b}	87.79^{ab}	91.38^{a}	89.07^{ab}	91.62^{a}	87.9^{ab}	87.68^{ab}	88.71^{ab}	89.02^{ab}
					Bifidobacterium. BB12				
Fresh	5.30^{b}	7.55^{a}	7.64^{a}	7.43^{a}	8.01^{a}	7.62^{a}	7.34^{a}	7.68^{a}	7.63^{a}
5	6.54^{c}	8.35^{ab}	8.48^{a}	8.01^{ab}	8.62^{a}	7.83^{ab}	7.55^{b}	7.90^{ab}	7.89^{ab}
10	6.67^{c}	8.62^{ab}	8.56^{ab}	7.97^{ab}	8.89^{a}	8.47^{ab}	7.64^{bc}	8.03^{ab}	8.07^{ab}
15	4.42^{b}	6.81^{a}	7.15^{a}	6.78^{a}	7.62^{a}	6.89^{a}	6.71^{a}	7.01^{a}	7.09^{a}
Viability%	86.35^{c}	90.20^{b}	93.59^{ab}	91.25^{ab}	95.13^{a}	90.42^{b}	91.42^{ab}	91.28^{ab}	92.92^{ab}

Means with small letters in the same row having different superscripts differ significantly ($p \le 0.05$).

C: control (100% milk permeate)

T1, 98% milk permeate + 2% barley flour

T2: 68% milk permeate + 2% barley flour and 30% carrot juice

T3: 98% milk permeate +2% oat flour

T4: 68% milk permeate +2% oat flour + 30% carrot juice

T5: 96% milk permeate + 4% barley flour

T6: 66% milk permeate + 4% barley flour + 30% carrot juice

T7: 96% milk permeate + 4% oat flour

T8: 66% milk permeate + 4% oat flour+ 30% carrot juice.

Conclusion

It can be concluded that the use of milk permeateenriched with probiotic strains and fortified with carrot juice, oat and barley flour in synbiotic beverages performs a bifidogenic function, modulating the microbial flora and conferring health benefits.The beverages from different treatments had acceptable quality and composition compared with control during cold storage for 15 days.The overall results cleared that, it is possible to produce good quality fermented milk permeate with good flavour, body & texture and appearance by adding carrot juice and oat flour (especially 66 % milk permeate + 4% oat flour + 30% carrot juice).

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TABLE 6. Sensory evaluation of synbiotic beverages during storage.

Storage period (day)	Treatments								
	C	T1	T2	T3	T4	T5	T6	T7	T8
Colour (10)									
Fresh	6.8[b]	8.1[a]	8.4[a]	8.2[a]	8.5[a]	8.2[a]	8.7[a]	8.6[a]	9.1[a]
5	6.4[b]	7.7[a]	8.0[a]	8.1[a]	8.3[a]	8.1[a]	8.3[a]	8.3[a]	8.7[a]
10	6.0[c]	7.2[b]	7.7[ab]	7.8[ab]	8.1[ab]	7.9[ab]	8.3[a]	8.1[ab]	8.4[a]
15	5.8[d]	6.9[c]	7.1[bc]	7.7[ab]	7.7[ab]	7.9[a]	8.0[a]	8.2[a]	8.3[a]
Taste (10)									
Fresh	5.9[c]	7.5[b]	7.8[b]	7.8[b]	8.2[b]	8.0[b]	8.2[b]	8.2[b]	9.2[a]
5	5.5[c]	7.2[b]	7.6[b]	7.5[b]	7.8[b]	7.7[b]	8.0[ab]	8.1[ab]	8.9[a]
10	5.0[c]	7.1[b]	7.3[b]	7.2[b]	7.5[b]	7.5[b]	7.8[ab]	7.9[ab]	8.4[a]
15	4.8[c]	6.8[b]	7.0[b]	7.1[ab]	7.3[ab]	7.3[ab]	7.6[ab]	7.7[ab]	8.4[a]
Flavour (10)									
Fresh	6.9[b]	8.2[a]	8.7[a]	8.3[a]	9.0[a]	8.4[a]	9.0[a]	8.6[a]	9.3[a]
5	6.4[b]	7.9[a]	8.3[a]	8.0[a]	8.7[a]	8.0[a]	8.7[a]	8.2[a]	9.0[a]
10	6.2[c]	7.4[b]	7.8[ab]	7.8[ab]	8.4[ab]	7.8[ab]	8.5[ab]	8.0[ab]	8.6[a]
15	4.1[b]	7.2[a]	7.6[a]	7.5[a]	8.1[a]	7.6[a]	8.2[a]	7.9[a]	8.1[a]
Acceptability(10)									
Fresh	5.5[c]	7.8[b]	8.3[ab]	8.1[b]	8.6[ab]	7.9[b]	8.5[ab]	8.6[ab]	9.5[a]
5	5.1[c]	7.5[b]	8.1[ab]	7.6[b]	8.2[ab]	7.5[b]	8.3[ab]	8.2[ab]	9.1[a]
10	4.8[c]	7.1[b]	7.7[ab]	7.4[ab]	7.8[ab]	7.2[b]	8.1[ab]	8.0[ab]	8.7[a]
15	3.5[e]	6.9[d]	7.3[bcd]	7.1[bcd]	7.5[bcd]	6.9[d]	7.7[abc]	7.8[ab]	8.4[a]
Total score (40)									
Fresh	25.10[b]	31.65[a]	33.16[a]	32.37[a]	34.26[a]	32.39[a]	34.37[a]	33.95[a]	37.03[a]
5	23.50[b]	30.2[a]	31.92[a]	31.22[a]	32.95[a]	31.41[a]	33.32[a]	32.9[a]	35.73[a]
10	22.11[c]	28.86[b]	30.49[ab]	30.14[ab]	31.81[ab]	30.49[ab]	32.56[ab]	32.00[ab]	34.16[a]
15	18.18[b]	27.81[a]	28.97[a]	29.36[a]	30.68[a]	29.67[a]	31.57[a]	31.63[a]	33.14[a]

Means with small letters in the same row having different superscripts differ significantly ($p \le 0.05$).

C: control (100% milk permeate)

T1: 98% milk permeate + 2% barley flour

T2: 68% milk permeate + 2% barley flour and 30% carrot juice

T3: 98% milk permeate +2% oat flour

T4: 68% milk permeate +2% oat flour + 30% carrot juice

T5: 96% milk permeate + 4% barley flour

T6: 66% milk permeate + 4% barley flour + 30% carrot juice

T7: 96% milk permeate + 4% oat flour

T8: 66% milk permeate + 4% oat flour+ 30% carrot juice.

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إعداد وخصائص المشروبات السينبيوتك من راشح اللبن والغنية بسلالات البروبيوتك الحيوية والمدعم بعصير الجزر ودقيق الشعير والشوفان

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تم اعداد مشروبات سينبيوتك من خلال تخمر راشح اللبن باستخدام المزرعة البكتيرية المحتوية على *Streptococcus thermophilus* و *Lactobacillus acidophilus*و *Bifidobacterium* BB-12مع عصير الجزر والدقيق الأبيض من الشعير والشوفان من أجل مقارنة وملاءمة إنتاج مشروبات سينبيوتك جديدة. تم تقييم الخصائص الفيزيائية والكيميائية والبكتريولوجية والريولوجية والحسية والمكونات الوظيفية (مضادات الأكسدة والفينول الكلي) خلال فترة التخزين لمدة ١٥ يوم على ٤ ±١ °م. محتوى أعداد الخلايا البكتيرية القابلة للحياة طوال فترة تخزين المنتج أعلى من الحد الأدنى المطلوب في منتج البروبيوتيك (أكثر من ٦ لوغاريتمات وحدة مكونة للمستعمرة/جرام). لوحظ أعلى عدد للخلايا الحية في مشروبات راشح اللبن المدعمة بعصير الجزر ودقيق الشوفان T4 (٦٨٪ راشح اللبن + ٢٪ دقيق الشوفان + ٣٠٪ عصير جزر). مشروبات السينبيوتك من راشح اللبن المدعم بعصير الجزر ودقيق الشوفان والشعير تحسنت فيها النكهة والقوام والقابلية العامة. أوضحت النتائج الإجمالية أنه من الممكن إنتاج مشروبات من راشح اللبن جيدة الجودة مع نكهة وقوام ومظهر جيد من خلال إضافة عصير الجزر ودقيق الشوفان T8 (٦٦٪ راشح اللبن + ٤٪ دقيق شوفان + ٣٠٪ عصير جزر).