Egyptian Journal of Chemistry

http://ejchem.journals.ekb.eg/

Properties of Fermented Permeate Beverages Enriched With Barley, Oat and Black Rice Extract

Fatma Samy Abd El-masked[1], Marwa Hatem El-Gendy[*2], Amr El-Sayed Metwally[1] and Mohammed AbdelRazek El Nawawy[1]

[1] *Food Science Department, Faculty of Agriculture, Ain Shams University, Cairo, Egypt*

[2] *Animal Production Division, Desert Research Center, Ministry of Agriculture., Cairo, Egypt*

Abstract

This research was conducted to produce beverages with health benefits from permeate as a by-product fortified with extracts of grains such as black rice, oats and barley at different concentrations and fermented with Lactobacillus casei and Lactobacillus. helveticus separately and comparing the suitability of these concentrates for production with examining the physical, chemical, microbiological, sensory and antioxidant properties during the storage period for 3 weeks at 4°C. It was observed that the level of antioxidants increased with the increase in the concentration of the extracts, and the concentration of antioxidants was higher in the fermented beverages fortified with black rice. It decreased during the storage period for all treatments. It was also noted that the counts of L. casei increased in all treatments and during the storage period compared to the counts of L. helveticus. The results showed that it was possible to produce fermented beverages from cereal extracts with texture, flavour, colour, and appearance from L. helveticus fermentation that has a high degree of preservation stability.

Keywords: permeate; antioxidants; cereal, L. casei ; L. helveticus

1. Introduction

Dairy by-products are characterized by their high nutritional value, so the tendency was to benefit from them, both in the food and non-food industries. And due to the increase in cheese manufacturing, the production of permeate is increasing, which is produced in the manufacture of cheese during UF process of milk [1].

Beverages are among the foods that meet consumer demands. Therefore, the market has recently witnessed a boom in the vegetable dairy products market because of its many health benefits, especially for people who suffer from health problems such as diabetes and cardiovascular [2], and those who have problems with eating dairy products and suffer from protein sensitivity and lactose intolerance [3]. The nutritional value of cereal-based foods can be enhanced by the fermentation of lactic acid bacteria, such as some strains of Lactobacillus and Bifidobacterium, which can improve starch digestibility and increase lysine availability in children [4].

Recently, interest in probiotic foods has increased worldwide, and food products fermented with probiotics, especially cereals and their extracts, are constantly developing. Because of the large distribution of cereals and their important nutritional value, interest has focused on their use in the development of novel fermented functional foods [5].

Oat (*Avena sativa* L.) has a high functional potential due to its composition. Oats are known to be an excellent source of dietary fiber, antioxidants and a well balanced protein fraction [5], [6], [7], [8]. They provide more protein, fiber, calcium, iron, zinc and essential amino acids than other whole grain crops [7], [8],[9], [10]. Oats are known to be unique among cereals as they are therapeutically active against diabetes, dyslipidemia, hypertension, inflammatory state and vascular injury [9], [10], [11].

*Corresponding author e-mail: marwaelgendy616@gmail.com; (Marwa Hatem El-Gendy).
EJCHEM use only: Received date 19 September 2022; revised date 26 September 2022; accepted date 02 October 2022
DOI: 10.21608/EJCHEM.2022.163910.7004

Rice is one of the most consumed grains in many peoples, especially East Asian countries, as a rich source of carbohydrates. Black rice (*Oryza sativa* L.) is considered an important crop due to its many health benefits as it contains a high percentage of anthocyanins [12] as well as many phenolic compounds with biological activity [13] This encouraged its use in the diet of patients with diabetes, cardiovascular disease and cancer [14]. Thus, the trend has become to develop the use of rice and its manufacture beverages from it that have health benefits.

Barley (*Hordeum vulgare* L.) is one of the most important grains known from ancient times. Due to its ability to adapt to different environmental conditions [15]. Several studies have indicated the nutritional importance of barley in reducing glycaemic index and blood cholesterol due to β-glucans [16]. It has also been used as an alternative to dairy, which is characterized by not containing lactose or cholesterol [17].

In this respect, cereals contain many components that make them achieve the concept of prebiotics. Which can be used when stimulating *L. casei* and *L. helveticus* and its biological and technological properties for application in foods, and its ease of adaptation to being applied to dairy products, to produce a synbiotic product [18], [19].

Given that the grains are relatively cheap, have high benefits with high nutritional values, in addition to the by-product permeate, the research objective was to produce beverages of functional characteristics using fermented cereals extracts as black rice, barley and oats, in order to raise the nutritional value of these products.

2. Materials and Methods

2.1. Materials

Permeate was obtained from Animal production research institute, Agricultural Research Center, Giza. it was a by-product of cheese making buffalo milk by Ultra-Filtration method. The permeate was immediately heat treated in a water bath at 85ºC for 15 min, then cooled to 4 ± 1°C.

Barley (Hordium vulgare L.), Oats (Avena sativa) and Black rice (Oryza sativa L.) grains were used in the present study. All grains were obtained from Agricultural Research Center, Giza, Egypt. These dry grains were ground using a mill (Moulinex, AR11, made in China). Then, it were packaged and stored in polyethylene bags at 4°C until use. The chemical composition of raw material used in preparing synbiotic beverages are presented in table 1.

Table 1
Chemical composition of raw material used in preparing synbiotic beverages

%	Moisture	Protein	Fat	Carbohydrate	Dietary fiber	Ash
Permeate	93.96^{A}	0.18^{D}	0.08^{C}	5.57^{D}	-	0.21^{D}
Barley	8.64^{B}	10.30^{A}	2.16^{B}	74.66^{B}	2.55^{B}	1.70^{C}
Oat	7.31^{C}	8.26^{B}	4.12^{A}	66.50^{C}	10.14^{A}	3.67^{B}
Black rice	6.19^{D}	7.31^{C}	2.70^{B}	75.50^{A}	2.89^{B}	5.42^{A}
±SE	0.167	0.123	0.262	0.357	0.074	0.004

A, B-D Mean with different letters are statistically different ($p \leq 0.05$) for column

Starter cultures (lactobacillus helveticus (LH-B02) and lactobacillus casei 01) were obtained from Chr. Hansen A/S Hørsholm, Denmark. It was activated in 10% (w/v) sterile reconstituted skim milk and incubated at 40 °C for 18 h.

Stabilizer Carboxymethyl cellulose (CMC), DPPH (1, 1 diphenyl-2-picrylhydrazyl) were obtained from Sigma-Aldrich, India., flavour, sugar and, natural colours (E124, E129 and E133) were purchased from a local market in Cairo, Egypt.

2.2. Preparation of grains extract

Barley extract: Barley milk was extracted according to the method of [20]. Where the barley was soaked in water in a ratio (2: 3 w/v) for 12 hours. Then add water to the soaked barley and mix it for a minute in the blender. Then followed by filtering the resulting barley milk to separate the barley grains from the resulting milk. The barley milk was kept at 4°C until use and analysis.

Oat extract: Oat milk was produced according to [21] method, with some modifications. Soak 100 gm of oats in enough water for 20 minutes. Then add 300 ml of water to the soaked oats and mix well until the liquid is homogeneous to preserve the β-glucan. Then filter the resulting milk and preserve it until use and analysis.

Black rice extract: The rice milk was prepared according to the method of [13], where the rice was soaked in water at room temperature for two hours at a ratio of 1:5 v/v. Then it was filtered after grinding using a blender. After that, it sterilization of rice milk at 70°C/ 15 minutes, then cooling at 4°C.

2.3. Preparation of the Inoculum

Each strain was separately activated in skimmed milk by dissolving 100 mg of freeze-dried culture in 50 ml of milk (10 g/100 g of total solids; autoclaved at 121°C for 20 min). Then the fermentation process was carried out at 37 ± 1 °C and reached pH 4.7-4.6 in 6 hours. Bacterial counts in these cultures approximately 6 Log CFU/ml.

2.4. Beverages fermented preparation

18 different types of cereal beverages fermented were prepared by mixing different mixtures of permeate and cereals milk. Each mixture contained at a fixed level 4% sugar, 4% CMC, 0.5% flavour, and fixed concentrations of red, raspberry and blue colours. The percentage of both oat and barley milk was 10, 20 and 30%, while the black rice milk had a concentration of 30, 50, and 70%. Two types of L. helveticus and L. casai 01, and the following table shows the design of the experiment as follows table (2).

2.5. Production of fermented beverages:

The permeate was initially heat treated at 85°C /15 min and cooled to 4°C. Then, sugar, colour and stabilizer were dissolved in permeate. This is followed by the addition of cereal milk concentrates with a heat treatment on 72˚C / 15 min. after then cooling to 42°C and add to flavour and then strain (L. helveticus or L. casei) were added 1% (106 cfu /g) to mixtures and incubated at 37°C/24 h, packing in sterile containers, and storage for 3 weeks at 5°C. The method of preparing synbiotic beverages is shown in Figure 1.

Table 2
Ingredients in fermented permeate cereal milk beverages

Ingredient		Barley			Oat			Rice		
		B10	B20%	B30%	O10%	O20%	O30%	R30%	R50%	R70%
Extract (ml)		10	20	30	10	20	30	30	50	70
Colour g/100ml	red	0.01025			0.01025			0.01025		
	raspberry	0.0037			0.0037			0.0037		
	blue	0.0001			0.0001			0.0001		
Sugar %		4			4			4		
Flavour %		0.5			0.5			0.5		
CMC g./L		4			4			4		
Permeate ml		Up to 100			Up to 100			Up to 100		
Culture		10^6 cfu/ml			10^6 cfu/ml			10^6 cfu/ml		

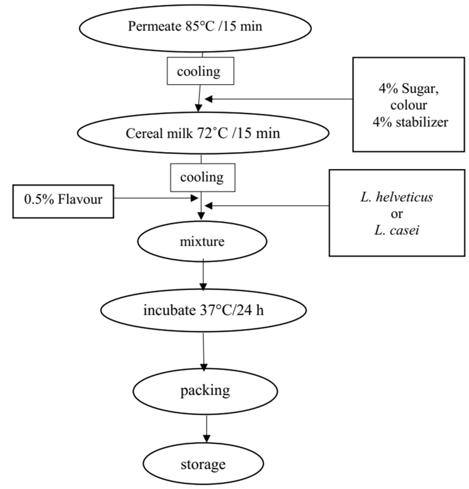

Figure 1. Flowchart of fermented beverages cereal milk production

2.6. Methods

2.6.1. Chemical analysis

pH value was measured using a pH meter (model HI 9311, HANNA Instruments, Lisbon, Portugal) at room temperature. Ash, protein contents were determined as reported in [22]. Carbohydrate was determined according to [23] by spectrophotometer (JENWAY 6105 U.V /Vis.). Total antioxidant content of products was estimated using the method of [13] using DPPH solution. And the measurement of the absorbance of the sample extracts at 517 nm using a spectrophotometer (JENWAY 6105 U.V /vis.) The antioxidant activity was calculated as the inhibition of free radical DPPH in percent (%) according to the following equation:

- Inhibition (%) = [(A517control-A517sample)/ A517control] ×100

2.6.2. Microbiological analysis

For the Lactobacillus helveticus or Lactobacillus casei MRS (Man, Rogosa and Sharpe) agar was used. The viability of Lactobacillus helveticus or Lactobacillus casei was determined at every week. After making the dilutions were plated on MRS agar to count the starting cultures present in formula according to [24]. The plates were incubated for 48 h at 37 °C, under anaerobic conditions. Considering, the safety aspects of the product, microbiological analyses measured coliforms bacterial counts (45 °C) and the presence of yeast and mold in beverages as suggested by [25].

2.6.3. Sensory evaluation

Sensory evaluation of the prepared formulas was carried out to once of week during storage 3 weeks of cold storage by panel tests of 10 judges. The maximum attainable scoring 20 point for colour, texture 10 points, flavour 20 points and 50 points overall acceptability according to [26].

2.7. Statistical Analysis

Data of the experimental were analyzed by the General Linear Model (GLM) procedure of Statistical Analysis Systems [27].

3. Results and Discussion

Chemical compositions of products fermented permeate cereal milk beverages using Lactobacillus casei or Lactobacillus helveticus are presented in table (3). The results showed that there were significant differences ($p \geq 0.05$) when comparing the two strains in the chemical composition, except for protein and ash, there were no significant differences ($p > 0.05$). However, the differences were significant ($p \geq 0.05$) in the chemical composition, except for the ash, when comparing the different concentrations of the fermented permeate cereal milk beverages.

The results indicated that with an increase in the concentration of cereal milk in the fermented permeate beverages, this led to an increase in the chemical content, and this was evident in each of the permeate beverages fermented from barley milk, oat milk and black rice milk. Where there was a difference in moisture in the different samples due to the of containing soluble fiber, which accordingly affects the level of carbohydrates and protein [28]. This was evident in the black rice milk beverage, followed by barley and oats, respectively. With the increase to concentration of beverages increase carbohydrates thus soluble fiber increased, and this was in oat milk beverages, barley milk beverages, and then black rice milk beverages, respectively. Also, Table 2 shows that there was significant ($p \geq 0.05$) increase in protein content for all beverages. On the other hand, the total protein for fermented beverages fortified with barley, oats and black rice increased with the increase in the added percentage of them. This is due to the increased concentration and decreased moisture content. These results agreed with [29], [30].

Table 3

Chemical composition of fermented permeate cereal milk beverages

		Moisture	Carbohydrate	Protein	Ash
L. casei	B10%	93.90^{b}	3.27^{c}	0.20^{bcd}	0.995
	B20%	93.80^{bc}	2.35^{g}	0.26^{ab}	0.996
	B30%	93.72^{c}	4.99^{f}	0.27^{a}	0.998
	O10%	93.55^{d}	3.02^{e}	0.20^{bcd}	0.995
	O20%	93.35^{d}	2.86^{d}	0.21^{bc}	0.997
	O30%	93.25^{d}	2.63^{d}	0.24^{ab}	0.996
	R30%	94.23^{a}	4.18^{b}	0.13^{cd}	0.996
	R50%	94.26^{a}	4.23^{f}	0.15^{bcd}	0.996
	R70%	94.08^{a}	4.48^{a}	0.18^{bcd}	0.996
L. helveticus	B10%	93.75^{b}	5.49^{c}	0.17^{bcd}	0.997
	B20%	93.32^{bc}	3.99^{g}	0.21^{ab}	0.996
	B30%	92.20^{c}	3.28^{f}	0.30^{a}	0.997
	O10%	91.35^{d}	3.53^{e}	0.18^{bcd}	0.997
	O20%	91.19^{d}	5.35^{d}	0.19^{bc}	0.997
	O30%	91.13^{d}	5.36^{d}	0.17^{ab}	0.996
	R30%	94.45^{a}	4.99^{b}	0.12^{cd}	0.996
	R50%	93.88^{a}	2.01^{f}	0.16^{bcd}	0.995
	R70%	94.06^{a}	2.23^{a}	0.18^{bcd}	0.997
±SE		0.143	0.068	0.039	0.011

Mean with different letters are statistically different ($p \leq 0.05$) for column

B10%= 10% barley milk, B20%= 20% barley milk, B30%= 30% barley milk

O10%= 10% Oat milk, O20%= 10% Oat milk, O30%= 10% Oat milk;

R30%=30% Black rice milk, R50%=50% Black rice milk, R70%= 70% Black rice milk

Table 4 shows the effect of fermented permeate cereal milk beverages with lactic acid bacteria on pH during storage. The pH of the beverage was observed to significant decrease during storage. The highest pH values were recorded in the first week, and the highest value was in beverages fermented with L. casei for oat 10% (5.94), while it was for oat 30%

fermented with L. helveticus (5.75). The pH values reduction for all treatments during storage is due to the accumulation of lactic and organic acids produced by lactic acid bacteria and its metabolic activity [30], [31]. Also, [32] found that fortification of functional beverages from whey and permeate with fruit lowers the pH value.

The results indicated in Table 5 that there were significant differences when fermenting permeate beverages fortified with cereal milk, L. casei and L. helveticus when DPPH (%) was inhibited. Also, the addition of cereal milk was significant, as the inhibition DPPH (%) increased with the level of cereal milk addition, and it was higher in black rice milk, barley milk, then oat milk. DPPH (%) inhibition beverages fortified with black rice milk by 70% were (19.97, 18.73 %) when fermentation of L. casei and L. helveticus, respectively. This is due to the higher level of antioxidants in black rice than barley and oats. In addition, results indicate significant decrease in DPPH inhibition values during storage period for all treatments was observed.

Table 4

pH values of fermented permeate cereal milk beverages during cold storage

	L. casei			*L. helveticus*		
	W1	W2	W3	W1	W2	W3
B10 %	5.02^{D}_{a}	4.80^{D}_{b}	4.68^{D}_{c}	4.75^{Da}	4.72^{D}_{b}	4.66^{D}_{c}
B20 %	4.80^{Fa}	4.64^{Fb}	4.53^{Fc}	4.64^{Fa}	4.66^{Fb}	4.59^{Fc}
B30 %	4.70^{Fa}	4.63^{Fb}	4.57^{Fc}	4.67^{Fa}	4.64^{Fb}	4.60^{Fc}
O10 %	5.94^{C}_{a}	5.85^{C}_{b}	5.50^{C}_{c}	5.34^{Ca}	5.32^{C}_{b}	5.26^{C}_{c}
O20 %	5.22_{Ba}	5.42^{B}_{b}	5.24^{B}_{c}	5.71^{Ba}	5.69^{B}_{b}	5.59^{B}_{c}
O30 %	5.43_{Aa}	5.43^{A}_{b}	5.34^{A}_{c}	5.75^{Aa}	5.82^{A}_{b}	5.63^{A}_{c}
R30 %	4.69^{Ea}	4.58^{Eb}	4.65^{Ec}	4.83^{Ea}	4.74^{Eb}	4.73^{Ec}
R50 %	4.62^{G}_{a}	4.55^{G}_{b}	4.27^{G}_{c}	4.52^{Ga}	4.49^{G}_{b}	4.40^{G}_{c}
R70 %	4.55^{G}_{a}	4.43^{G}_{b}	4.45^{G}_{c}	4.51^{Ga}	4.49^{G}_{b}	4.44^{G}_{c}
±SE	0.026	0.026	0.026	0.026	0.026	0.026

$^{A, B - G}$ Mean with different letters are statistically different ($p \leq 0.05$) for concentration

$^{a, b, c}$ Mean with different letters are statistically different ($p \leq 0.05$) for weeks

B10%= 10% barley milk, B20%= 20% barley milk, B30%= 30% barley milk

O10%= 10% Oat milk, O20%= 10% Oat milk, O30%= 10% Oat milk;

R30%= 30% Black rice milk, R50%=50% Black rice milk, R70%=70% Black rice milk

W1= 1st week, W2= 2nd week, W3= 3rd week.

Table 5

DPPH inhibition (%) of fermented permeate cereal milk beverages during cold storage

	L. casei			*L. helveticus*		
	W1	W2	W3	W1	W2	W3
B10 %	18.11_{Ea}	17.74_{Eb}	16.09_{Ec}	16.94_{Ea}	16.04_{Eb}	15.50_{Ec}
B20 %	18.60_{Ba}	18.59_{Bb}	16.96_{Bc}	17.78_{Ba}	17.64_{Bb}	16.47_{Bc}
B30 %	18.79_{Ca}	18.65_{Cb}	17.19_{Cc}	17.81_{Ca}	15.60_{Cb}	15.97_{Cc}
O10 %	18.30_{Da}	17.79_{Db}	16.34_{Dc}	17.13_{Da}	17.08_{Db}	15.48_{Dc}
O20 %	18.09_{Ea}	16.74_{Eb}	15.30_{Ec}	17.47_{Ea}	17.22_{Eb}	15.60_{Ec}
O30 %	17.54_{Fa}	16.98_{Fb}	14.89_{Fc}	16.84^{F}_{a}	16.08_{Fb}	15.88_{Fc}
R30 %	18.68_{Ba}	18.46_{Bb}	16.77_{Bc}	17.88_{Ba}	17.64_{Bb}	15.99_{Bc}
R50 %	19.43_{Aa}	18.52_{Ab}	17.16_{Ac}	17.95_{Aa}	16.97_{Ab}	16.75_{Ac}
R70 %	19.97_{Aa}	19.32_{Ab}	17.61_{Ac}	18.73_{Aa}	18.16_{Ab}	16.50_{Ac}
±SE	0.150	0.150	0.150	0.150	0.150	0.150

$^{A, B - F}$ Mean with different letters are statistically different ($p \leq 0.05$) for concentration

$^{a, b, c}$ Mean with different letters are statistically different ($p \leq 0.05$) for weeks

B10%= 10% barley milk, B20%= 20% barley milk, B30%= 30% barley milk

O10%= 10% Oat milk, O20%= 10% Oat milk, O30%= 10% Oat milk;

R30%= 30% Black rice milk, R50%=50% Black rice milk, R70%=70% Black rice milk

W1= 1st week, W2= 2nd week, W3= 3rd week.

The activity of antioxidants is affected by pH. The results showed that the highest concentration of black rice milk > barley milk > oat milk had antioxidant activity (19.97, 18.79, 17.54 %), respectively, in samples fermented by L. casei. While L. helveticus was lower in values. The higher antioxidant activity in black rice than in barley and oats may be due to the high percentage of malvidin, cyanidin-3-glucoside and anthocyanin pigment [33]. in addition to being hydrophilic pigment and could bind to protein, and thus the pigment remains in a state of protection during storage [31], [34], [35]. [36] found that starter cultures increase the solubility and extractability of polyphenolic compounds; the effectiveness of these reactions depends on the strain and the specific enzymatic activity of bacteria. The fermentative processes that occur during the ripening of beverages contribute to the release of polyphenols.

The evaluation of L. casei and L. helveticus during storage period for fermented permeate cereal milk beverages in Table 6. In general, there were significant ($p \geq 0.05$) differences when evaluating L. casei and L. helveticus. It was also noted that the populations of L. casei increased in all treatments and during the storage period compared to the populations of L. helveticus. Also, the permeate beverages fermented with L. casei and fortified with black rice milk had the highest count of those fortified with oat milk and barley milk during the storage period. There is a statistical difference ($P < 0.05$) in the L. casei populations for all treatments during the storage period. While it was found that the permeate beverages fermented with L. helveticus were less active than L. casei. However, its activity in oat milk was highest that of black rice milk and barley milk. This may be due to the ability of lactic acid bacteria to grow optimally under slightly acidic conditions when the pH is between 4.5 and 6.4 [35]. Also, the high content of carbohydrates helped to increase the growth of starter, which helped to increase their count and increase their metabolism [37], [38].

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.

Table 6

Microbiological evaluation (Log cfu/g) of fermented permeate cereal milk beverages during cold storage

	L. casei				*L. helveticus*		
	W1	W2	W3		W1	W2	W3
B10 %	6.56^{B}_{Cb}	5.75^{B}_{Cc}	7.29^{B}_{Ca}		5.50^{B}_{Cb}	6.62^{B}_{Cc}	6.77^{B}_{Ca}
B20 %	6.00^{Bb}	6.91_{Bc}	7.02_{Ba}		5.50_{Bb}	5.50_{Bc}	7.41_{Ba}
B30 %	6.29^{Cb}	6.67_{Cc}	6.92_{Ca}		5.75_{Cb}	7.50_{Cc}	7.20_{Ca}
O10 %	6.25_{Ab}	5.75_{Ac}	7.51_{Aa}		6.07_{Ab}	6.79_{Ac}	7.33_{Aa}
O20 %	6.93^{Ab}	6.14^{Ac}	7.74^{Aa}		6.77^{Ab}	6.33^{Ac}	7.53^{Aa}
O30 %	6.22_{Ab}	6.10_{Ac}	7.59_{Aa}		6.75_{Ab}	5.50_{Ac}	7.53_{Aa}
R30 %	6.70^{Bb}	7.12^{Bc}	7.77^{Ba}		6.28^{Bb}	6.27^{Bc}	7.50^{Ba}
R50 %	6.98^{B}_{Cb}	7.12^{B}_{Cc}	7.51^{B}_{Ca}		6.02^{B}_{Cb}	7.50^{B}_{Cc}	7.75^{B}_{Ca}
R70 %	6.26^{B}_{Cb}	7.00^{B}_{Cc}	7.73^{B}_{Ca}		6.00^{B}_{Cb}	7.75^{B}_{Cc}	7.75^{B}_{Ca}
±SE	0.258	0.258	0.258		0.258	0.258	0.258

$^{A, B - C}$ Mean with different letters are statistically different ($p \leq 0.05$) for concentration

$^{a, b, c}$ Mean with different letters are statistically different ($p \leq 0.05$) for weeks

B10%= 10% barley milk, B20%= 20% barley milk, B30%= 30% barley milk

O10%= 10% Oat milk, O20%= 10% Oat milk, O30%= 10% Oat milk;

R30%= 30% Black rice milk, R50%=50% Black rice milk, R70%=70% Black rice milk

W1= 1st week, W2= 2nd week, W3= 3rd week.

The results in table 7 reveal showed the sensory evaluation of the extracts of grains permeate fermented by L. casei. There was overall acceptance of all beverages, and it was significant. While there were no significant differences in terms of colour, flavour and texture. It was also noted that all products spoiled and gas formation after a week storage and this may be due to the activity of the initiator and its microbial load and gas formation in addition to the low pH. These results are in agreement with [38] that with the availability of excess amounts of lactose, galactose and citrate during the initial maturation stages of L. casei heterotrophs with sufficient substrates for gas formation. On the other hand, all beverages fermented by L. casei experienced colour changes during storage time leading to violet colour after 1st week to green colour. it may be due to L. casei demonstrating the greatest counts activity and the loss of antioxidant activity [34].

Table 7

Sensory evaluation of fermented permeate cereal milk beverages by L. casei during cold storage

	L. casei								
	B10%	B20%	B30%	O10%	O20%	O30%	R30%	R50%	R70%
	Colour (20)								
W1	20.00	20.00	20.00	20.00	20.00	20.00	20.00	19.75	20.00
	Texture (10)								
W1	8.75	9.25	9.50	9.75	9.50	9.75	10.00	10.00	9.75
Flavour (20)									
W1	20.00	19.25	19.75	19.75	19.75	19.75	19.75	20.00	020.00
Overall acceptability (50)									
W1	48.75^{AB}	48.50^{B}	49.25^{AB}	49.50^{AB}	49.25^{AB}	49.50^{AB}	49.75^{A}	49.75^{A}	49.75^{A}

$^{A, B}$ Mean with different letters are statistically different ($p \leq 0.05$) for concentration

B10%= 10% barley milk, B20%= 20% barley milk, B30%= 30% barley milk

O10%= 10% Oat milk, O20%= 10% Oat milk, O30%= 10% Oat milk;

R30%= 30% Black rice milk, R50%=50% Black rice milk, R70%=70% Black rice milk

W1= 1st week.

Table 8 shows the sensory evaluation of the extracts of grains permeates fermented by L. helveticus. In general, there was a significant improvement in the sensory characteristics, whether between treatments or during the storage period. There was overall acceptability of all treatments of rice, oats and barley during the first week, while the samples treated with rice (30%) and all treatments of oats and then barley were the most acceptable, respectively, during the 2nd week. Then there was spoiling and change in colour in the 3rd week. This is due to the low pH and increased L. helveticus activity. L. helveticus better retained the content of antioxidant activity within 2 weeks of refrigerated storage. A similar observation was confirmed by [34].

Table 8
Sensory evaluation of fermented permeate cereal milk beverages by L. helveticus during cold storage

	L. helveticus								
	B10%	B20%	B30 %	O10 %	O20 %	O30 %	R30 %	R50 %	R70 %
	Colour (20)								
W1	20.00 Aa	20.00 Aa	20.00 Ba	20.00 Aa	20.00 Aa	20.00 Aa	20.00 Aa	19.75 Ca	20.00 Ca
W2	20.00 Ab	20.00 Ab	10.00 Bb	19.75 Ab	19.75 Ab	19.00 Ab	18.00 Ab	0.00	0.00
	Texture (10)								
W1	9.25^{Ab} a	9.25^{Ab} a	9.25^{B} a	9.75^{A} a	9.50^{A} a	9.75^{A} a	10.00 Aa	10.00 Ca	9.75^{C} a
W2	6.25^{Ab} b	6.25^{AB} b	4.00^{B} b	8.50^{A} b	9.00^{A} b	8.50^{A} b	8.50^{A} b	0.00	0.00
	Flavour (20)								
W1	19.75^{C} Da	19.25^{D} a	19.50 Da	19.75^{B} Ca	19.75 Aba	19.75 Aba	19.75 Aa	20.00 Ea	20.00 Ea
W2	7.50^{CD} b	6.50^{Db}	6.00^{Db}	11.50^{B} Cb	15.50 Abb	16.00 Abb	17.00 Ab	0.00	0.00
	Overall acceptability (50)								
W1	49.00 Aba	48.50 Aba	48.75 Ba	49.50 Aa	49.25 Aa	49.50 Aa	49.75 Aa	49.75 Ca	49.75 Ca
W2	33.75 Abb	32.75 ABb	20.00 Bb	39.75 Ab	44.25 Ab	43.50 Ab	43.50 Ab	0.00	0.00

A, B- E Mean with different letters are statistically different ($p \leq 0.05$) for concentration

a, b, c Mean with different letters are statistically different ($p \leq 0.05$) for weeks

B10%= 10% barley milk, B20%= 20% barley milk, B30%= 30% barley milk

O10%= 10% Oat milk, O20%= 10% Oat milk, O30%= 10% Oat milk;

R30%= 30% Black rice milk, R50%=50% Black rice milk, R70%=70% Black rice milk

W1= 1st week, W2= 2nd week.

4. Conclusion

It can be concluded that the use of milk permeate enriched with probiotic strains and fortified with cereal extracts can give health benefits. It is recommended to produce these beverages fermented with L. helveticus that have a shelf life of two weeks. It can be given to certain groups of people, as it is rich in antioxidants, in addition to being considered a Synbiotic product. Finally, this kind of study can facilitate the development of new, fermented, non-dairy, nutritionally well-balanced food products with unique physical properties.

5. Conflicts of interest

There are no conflicts to declare.

6. References

1. Rizk, A.E., Study of production functional beverages of milk permeate fortified with fruit and herbs. Middle East Journal of Applied Sciences, 2016. 6(1): p. 155-161.
2. McMacken, M. and S. Shah, A plant-based diet for the prevention and treatment of type 2 diabetes. Journal of geriatric cardiology: JGC, 2017. 14(5): p. 342.
3. Sethi, S., S.K. Tyagi, and R.K. Anurag, Plant-based milk alternatives an emerging segment of functional beverages: a review. Journal of food science and technology, 2016. 53: p. 3408-3423.
4. do Espirito-Santo, A.P., et al., Influence of cofermentation by amylolytic Lactobacillus strains and probiotic bacteria on the fermentation process, viscosity and microstructure of gruels made of rice, soy milk and passion fruit fiber. Food Research International, 2014. 57: p. 104-113.
5. Hassan, A.A., M. Aly, and S. El-Hadidie, Production of cereal-based probiotic beverages. World Applied Sciences Journal, 2012. 19(10): p. 1367-1380.
6. Staka, A., E. Bodnieks, and A. Puķītis. Impact of oat-based products on human gastrointestinal tract. in Proceedings of the Latvian Academy of Sciences. Section B. Natural, Exact, and Applied Sciences. 2015.
7. Shah, A., et al., Newly released oat varieties of himalayan region-Techno-functional, rheological, and nutraceutical properties of flour. LWT, 2016. 70: p. 111-118.
8. Angelov, A., T. Yaneva-Marinova, and V. Gotcheva, Oats as a matrix of choice for developing fermented functional beverages. Journal of food science and technology, 2018. 55: p. 2351-2360.
9. Sangwan, S., R. Singh, and S.K. Tomar, Nutritional and functional properties of oats: An

update. Journal of Innovative Biology, 2014. 1(1): p. 3-14.
10. Tosh, S.M. and Y. Chu, Systematic review of the effect of processing of whole-grain oat cereals on glycaemic response. British Journal of Nutrition, 2015. 114(8): p. 1256-1262.
11. Londono, D., et al., Avenin diversity analysis of the genus Avena (oat). Relevance for people with celiac disease. Journal of Cereal Science, 2013. 58(1): p. 170-177.
12. Hou, Z., et al., Identification of anthocyanins isolated from black rice (Oryza sativa L.) and their degradation kinetics. Food research international, 2013. 50(2): p. 691-697.
13. Deeseenthum, S., V. Luang-In, and S. Chunchom, Characteristics of Thai pigmented rice milk kefirs with potential as antioxidant and anti-inflammatory foods. Pharmacognosy Journal, 2018. 10(1).
14. Bae, I.Y., et al., Optimized preparation of anthocyanin-rich extract from black rice and its effects on in vitro digestibility. Food Science and Biotechnology, 2017. 26: p. 1415-1422.
15. Sami Jan, M.N.K., M. Ashraf Bhat, M. Altaf Wani, Farooq Ahmad Bhat, Raihana Habib Kanth, Barley (Hordeum vulgare L.) Classification, Diversity and Health Benefits: A Review. European J. Experimental Biology, 2022. 12: p. 118.
16. Havrlentova, M., et al., β-glucans and their significance for the preparation of functional foods-a review. Czech Journal of Food Sciences, 2011. 29(1): p. 1-14.
17. Mäkinen, O.E., et al., Foods for special dietary needs: Non-dairy plant-based milk substitutes and fermented dairy-type products. Critical reviews in food science and nutrition, 2016. 56(3): p. 339-349.
18. Kumar, H., et al., Novel probiotics and prebiotics: road to the market. Current opinion in biotechnology, 2015. 32: p. 99-103.
19. Klajn, V.M., et al., Probiotic fermented oat dairy beverage: viability of Lactobacillus casei, fatty acid profile, phenolic compound content and acceptability. Journal of Food Science and Technology, 2021. 58: p. 3444-3452.
20. Gajdoš Kljusurić, J., M. Benković, and I. Bauman, Classification and processing optimization of barley milk production using NIR spectroscopy, particle size, and total dissolved solids analysis. Journal of Chemistry, 2015. 2015.
21. Bernat, N., et al., Optimisation of oat milk formulation to obtain fermented derivatives by using probiotic Lactobacillus reuteri microorganisms. Food Science and Technology International, 2015. 21(2): p. 145-157.
22. Horwitz, W., Official methods of analysis of the Association of Official Analytical Chemists. Vol. 17. 2000: The Association.
23. Albalasmeh, A.A., A.A. Berhe, and T.A. Ghezzehei, A new method for rapid determination of carbohydrate and total carbon concentrations using UV spectrophotometry. Carbohydrate polymers, 2013. 97(2): p. 253-261.
24. Tharmaraj, N. and N. Shah, Selective enumeration of Lactobacillus delbrueckii ssp. bulgaricus, Streptococcus thermophilus, Lactobacillus acidophilus, bifidobacteria, Lactobacillus casei, Lactobacillus rhamnosus, and propionibacteria. Journal of dairy science, 2003. 86(7): p. 2288-2296.
25. Laird, D.T.G.L.S.A.a.S.F.M., Microbiological Count Methods, in Standard Methods for the Examination of Dairy Products., F.J.F.E. Wehr H M, Editor. 2004, American Public Health Association: Washington, DC20001, USA. p. 187-265.
26. Fasoyiro, S., S. Babalola, and T. Owosibo, Chemical composition and sensory quality of fruit-flavoured roselle (Hibiscus sabdariffa) drinks. World Journal of Agricultural Sciences, 2005. 1(2): p. 161-164.
27. SAS. User's guide. 14.3 edn, . 2017, Statistical Analysis Systems Institute Inc.: Cary NC 27511–8000, USA. .
28. Musa, M. and S. Al-Aithy, EXTRACTION OF β-GLUCAN FROM BARLEY BRAN AND ASSESMENT OF SOME FUNCTIONAL PROPERTIES AND MOLECULAR WIEGHT. Iraqi Journal of Agricultural Sciences, 2012. 43(2): p. 100-108.
29. Abbas, M., E. Gouda, and F. Marian, Some chemical and physical properties of cereal fermented milk product. Alexandria Science Exchange Journal, 2013. 34(April-June): p. 181-188.
30. El-Sayed, H. and M.F. Ramadan, Production of probiotic-fermented rice milk beverage fortified with cactus pear and Physalis pulp. Zagazig Journal of Agricultural Research, 2020. 47(1): p. 165-177.
31. Sattar, A. and M.F. Ramadan, Properties Of Milk Permeate Synbiotic Beverages Enriched With Carrot Juice And Barley And Oat Flour. Egyptian Journal of Food Science, 2021. 49(2): p. 319-331.
32. Aamer, R. and W. El-Kholy, Production of functional beverages from whey and permeate containing kumquat fruit. Alexandria Journal of

Food Science and Technology, 2017. 14(1): p. 41-56.
33. Wijaya, C. and A. Romulo. Proximate analysis and antioxidant activity of red rice (Oryza sativa L.) Milk. in Journal of Physics: Conference Series. 2021. IOP Publishing.
34. Sánchez-Bravo, P., et al., Antioxidant and anthocyanin content in fermented milks with sweet cherry is affected by the starter culture and the ripening stage of the cherry. Beverages, 2018. 4(3): p. 57.
35. Atwaa, E., A.A. Elmaadawy, and E.A. Awaad, Production of fruit flavored probiotic rice milk beverage. Journal of Food and Dairy Sciences, 2019. 10(12): p. 453-458.
36. Merenkova, S., R. Fatkullin, and I. Kalinina, Effect of Fermentation on the Biochemical Parameters Antioxidant Capacity and Dispersed Composition of Plant Beverages Based on Barley and Hemp Seeds. Fermentation, 2022. 8(8): p. 384.
37. McSweeney, P.L., Biochemistry of cheese ripening. International journal of dairy technology, 2004. 57(2-3): p. 127-144.
38. O'Sullivan, D.J., et al., Compromised Lactobacillus helveticus starter activity in the presence of facultative heterofermentative Lactobacillus casei DPC6987 results in atypical eye formation in Swiss-type cheese. Journal of Dairy Science, 2016. 99(4): p. 2625-2640.