

Probiotic oats milk drink with microencapsulated *Lactobacillus plantarum* – an alternative to dairy products

Alternative to dairy products

471

Sangami Ravindran

Department of Food and Nutrition, Bharathiar University, Coimbatore, India, and

RadhaiSri S.

Department of Nutrition and Dietetics, PSG College of Arts and Science, Coimbatore, India

Received 16 March 2020

Revised 13 May 2020

26 June 2020

Accepted 26 June 2020

Abstract

Purpose – Interest in probiotic food products has constantly increased due to the awareness on importance of gut microbiome; an increasing demand has encouraged the development of other matrices such as cereals, vegetable and fruit juices to deliver probiotics. The purpose of this paper is to standardize and evaluate a ready to serve probiotic oats milk drink fermented with microencapsulated *Lactobacillus plantarum* to be further used as a therapeutic module.

Design/methodology/approach – Fermentation of oats milk extract with microencapsulated *L. plantarum* was subjected to various trials in making it favourable for consumption and tested for sensory characteristics, physicochemical parameters, nutrient content, viable colony count and shelf life.

Findings – Fermented oats milk drink with 3% inoculum of microencapsulated *L. plantarum* was able to achieve desirable level of 2.5×10^8 and 2.3×10^8 colony forming units (CFU)/mL for spice and strawberry flavoured drink, respectively. Antioxidant property significantly increased after fermentation showing inhibitory effect against 2,2-diphenyl-1-picryl-hydrazyl-hydrate (DPPH) assay ($p = 0.05$).

Research limitations/implications – Analysis of all the parameters were conducted only with three samples; this was the potential limitation identified in this study as large sample size always be a better representative of the results.

Practical implications – Spice and strawberry flavoured nondairy oats milk drink facilitated to be a suitable carrier for microencapsulated *L. plantarum* with good sensory attributes, low fat, moderate calorie, high fiber content, antioxidant potential and a shelf life of two-week period at 4°C.

Originality/value – The developed ready to serve, spice and strawberry flavoured nondairy oats milk drink with compactly packed functional components inclusive of beneficial probiotic organisms, β -glucan and antioxidants can be prescribed as a therapeutic food for many clinical conditions and would serve as a good probiotic option for vegans.

Keywords Probiotics, Microencapsulation, Oats milk, Nondairy – vegan, Antioxidant potential, *L. plantarum*

Paper type Research paper

Introduction

In recent times, use of nutraceuticals with functional food ingredient has been rapidly growing due to its ability to enhance the bioavailability of the loaded active ingredients,

Disclosure of interest: The authors report no conflict of interest.



Nutrition & Food Science

Vol. 51 No. 3, 2021

pp. 471-482

© Emerald Publishing Limited

0034-6659

DOI 10.1108/NFS-03-2020-0073

resulting in improved therapeutic/nutraceutical outcomes (Durazzo *et al.*, 2020). Nutraceuticals, are defined as:

Nutraceuticals are the phytochemicals if they derive from a food of vegetal origin, and as the pool of the secondary metabolites if they derive from a food of animal origin, concentrated and administered in the more suitable form (Daliu *et al.*, 2018)

Functional food products mostly resemble conventional food in terms of appearance but are composed of bioactive compounds that may offer physiological health benefits beyond nutritive functions (Sarkar, 2019). Substances that have nutritional and nutraceutical interest are antioxidants, vitamins, polyunsaturated fatty acids, dietary fibres, prebiotics and probiotics (Durazzo *et al.*, 2018). Food and beverage products are increasingly considered to be popular carriers of probiotics and drastically extend the option for regular consumption of probiotics in the human diet. Probiotic foods comprise between 60 and 70% of the total functional food market. Incorporation of probiotics into a food matrix poses several technological challenges (Wilkinson, 2018), preserving the efficacy of probiotic bacteria exhibits paramount task that need to be addressed during the development, processing and storage of any functional food products. Several factors have been claimed to be responsible for reducing the viability of probiotics including intrinsic factors such as matrix acidity, level of oxygen in products, presence of other lactic acid bacteria, water activity, presence of salt, sugar, artificial flavoring and colouring agents, processing parameters including fermentation conditions (temperature, storage conditions, packaging materials, etc.) and finally microbiological parameters such as strain of probiotics employed, rate and proportion of inoculation (Terpou *et al.*, 2019; Putta *et al.*, 2018). However, encapsulation technique seems to enhance stability, protect sensitive probiotic lactic acid bacteria from oxygen, freezing and acidic conditions during production, storage and gastrointestinal transit (Ningtyas *et al.*, 2019). *In vitro* screening to validate the strain to own putative probiotic characteristics is a mandatory regimen prior to be used in food products and therapeutic application (Food and Agriculture Organization (FAO)/World Health Organization (WHO), 2002). Recent advancements are leading to development of synbiotic foods, a selective blend of probiotics and prebiotics (Salmeron, 2017), cereal-based products have been developed mainly for the synergistic effects of the two and dietary fibers, where the prebiotic functions as “a substrate that is selectively used by host microorganisms conferring a health benefit” (Gibson *et al.*, 2017). The primordial concept of cereal-based fermented foods is implemented into development of new functional oat-based fermented beverages as healthy, fast and convenient supplementary foods (Angelov *et al.*, 2018). Oats consumption in human diet has been increased because of health benefits associated with dietary fibres, β -glucan, phytochemicals and phenolic components present. Functional component β -glucan claims the prebiotic function in the gastrointestinal tract, supporting the growth of beneficial microbial groups, slowly digestible fraction of oat starch moderates the glycemic response (Angelov *et al.*, 2018). It is essential that commercialized probiotic products which make health claims meet the minimum criterion of 1×10^6 CFU/g or mL of the product at expiration date. Accordingly, the minimum dosage of probiotic cells per day for any beneficial effect on the consumer corresponds to an intake containing 10^6 – 10^9 CFU/g or mL per day (Kechagia *et al.*, 2013), more precisely viable count of 10^8 CFU/g or mL is recommended to compensate for reduction during passage through the gut to exert therapeutic effect (Guevarra and Barraquio, 2015). This study aimed at developing a ready to serve indigenous probiotic oats milk drink fermented with microencapsulated *L.plantarum* with therapeutic properties and an alternative to dairy milk.

Materials and methods

As the developed product would be recommended for therapeutic usage after the conduct of suitable randomized control trials, the study was registered in the Clinical Trial Registry India (CTRI) with Ref ID CTRI/2018/07/014873 and obtained ethical approval with Ref ID CMMHEC/18/03. Data on clinical trial results were not presented in this current report.

Isolation and identification of probiotic strain

The organism used in this study was isolated from the fermented grain broth and screened for putative probiotic characteristic with specific *in vitro* tests as given by ICMR-DBT, 2011 using standard methodology. *In vitro* screening tests performed were resistance to gastric acidity (pH), bile acid resistance (Hassanzadazar *et al.*, 2012), antimicrobial activity against potentially pathogenic bacteria and bile salt hydrolase activity (Tanaka *et al.*, 1999). In addition to these tests lipase inhibitory activity (Mopuri and Meriga, 2014) and anti-cholesterol assay (Pant *et al.*, 2015 and Randox Laboratories, 2009) were also performed to find out the lipid metabolizing effect of the isolated strains. Polymerase chain reaction (PCR)-based techniques and 16S rRNA sequencing was carried out to identify the genus and species of the selected isolated organism.

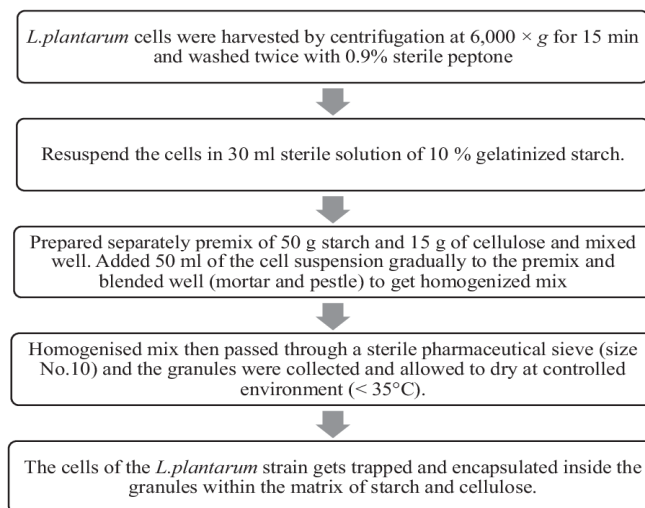
Microencapsulation of probiotic strain

Preparation and identification of microencapsulated L.plantarum. Encapsulation refers to a physicochemical or mechanical process to entrap a substance in a material to produce particles with diameters of a few nanometres to a few millimetres. The capsules are small particles that contain an active agent or core material surrounded by a coating or shell. Encapsulation shell materials include a variety of food grade polymers, such as alginate, chitosan, carboxymethyl cellulose, xanthan gum, starch, carrageenan, gelatin, pectin, fats and waxes (Shori, 2017), depending on the core material to be protected. Cultures of *L.plantarum* were obtained by inoculating the strains in Man, Rogosa and Sharpe (MRS) broth under anerobic conditions at 37°C for 36h, and the cultures were harvested by centrifugation and washed twice with sterile peptone (Cortés *et al.*, 2014). The cells of the *L.plantarum* strain were made to get trapped and encapsulated inside the granules within the matrix of starch and cellulose. Morphology of dried microcapsules were observed with a Scanning Electron Microscopy (SEM). All the equipment used for the microencapsulation procedure were cleaned using propanol and sterile environment was maintained throughout the process. Schematic flow of the process given as Figure 1.

Preparation of fermented probiotic oats milk using microencapsulated L.plantarum. Preliminary studies were conducted to evaluate the suitable method for extraction of milk from oats grain, standardization on the concentration of microencapsulated *L.plantarum* (1 to 5%) to be used for preparing the probiotic fermented oats milk drink and duration of fermentation to attain desirable colony counts of 10⁸ CFU/mL using MRS agar.

The same process of extraction of milk from oats grain as explained in a study by Sangami and Radhaisri (2018) was followed. Split oats grain was used for the milk extraction as it retains better yield than the whole grain or oat meal. Prior to soaking, the spilt oat grain was tempered in water boiled at 80°C for 3 mins and drained. The tempered oats grain was then soaked in 150 ml of sterilized hot water for 12h at room temperature, ground in a mixer grinder and the slurry filtered using a nut milk bag till the oats milk is completely extracted. The extracted milk was then sterilized using steam under pressure at 121°C for 15–20 min under 15 psi pressure (Earle and Earle, 2004). The sterilized oats milk extract was then inoculated with 3% microencapsulated *L.plantarum* granules at aseptic

Figure 1.
Schematic
representation of
microencapsulation
procedure of
L.plantarum



conditions, mixed well, closed air tight and kept at room temperature with constant shaking at intervals and allowed to ferment for 20–24 h.

Preparation of flavoured fermented oats milk with microencapsulated L.plantarum. First trial was conducted to find out the suitability of the flavours with the fermented probiotic oats milk. Among the nine different sweet flavours (almond, banana, rose, grape, orange, chocolate, pineapple, strawberry and cardamom) and spice flavour (dry roasted and ground – cumin, fennel, pepper and curry leaves), strawberry and spice-flavoured fermented oats milk drink had satisfactory palatability and was selected for further enhancement of sensory characteristics.

In the second trial, dehydrated natural fruit/vegetable powders, fruit syrup and dehydrated fruits were used instead of synthetic flavours. Strawberry flavored drink was further enhanced by replacing synthetic strawberry flavour with 2.5% of strawberry fruit syrup, 1.5% of dehydrated strawberry powder, 2.5% of cane sugar and 0.5% of stevia powder to 100 mL of the fermented oats milk. Spice flavored drink was further enhanced by adding 1% of spice mix powder (with added dehydrated onion powder) and 0.5% of salt to 100 mL of the fermented oats milk.

Sensory evaluation

Organoleptic evaluation of the strawberry and spice flavoured fermented probiotic oats milk drink with microencapsulated *L.plantarum* was performed with ten trained panelist in analyzing the colour, flavour, consistency, mouth feel, taste and overall acceptability based on a nine-point hedonic scale ranging from 1 (dislike extremely) to 9 (like extremely) (Wichchukit and Omahony, 2015). Sensory test was carried out in a laboratory setting with each panelist in an individual booth. The strawberry and spice flavoured probiotic oats milk drink was coded and set for evaluation by the panelists. Water was provided for panelists to cleanse their palate between samples.

Quality analysis

The developed strawberry and spice-flavoured fermented probiotic oats milk drinks were evaluated for quality characteristics. Association of Official Agricultural Chemists (AOAC)

protocol was used to determine all the physicochemical (pH, titrable acidity (TA), viscosity, total solids and moisture content) and nutritional (*total energy*, protein, carbohydrate, total dietary fiber) parameters except for β -glucan (Semedo *et al.*, 2015), antioxidant property by DPPH assay (Gadow *et al.*, 1997) and free fatty acid content using Gas Chromatography (GC) with flame ionization detection (FID) method (Paszczyk *et al.*, 2017). The products were also evaluated for enumeration of colony counts of *L.plantarum* (desirable range of 10^8 CFU/mL) using MRS agar and shelf-life analysis.

Packaging and storage

Studies reported that probiotic yoghurts packaged in glass bottles found to prevent oxygen diffusion and had significantly higher survival rate of probiotic organism, glass bottles also found to have the lowest Moisture Vapour Transmission Rate (MVTR), a bottle seal that adheres well to glass must be chosen to prevent moisture ingress (Fenster *et al.*, 2019). Accordingly, the developed flavoured fermented probiotic oats milk drink was packed in sterilized glass bottle, sealed immediately with airtight lid and stored refrigerated at 4°C. Refrigeration is required to help maintain probiotic viability to deliver adequate dosage throughout the shelf life and to avoid metabolic activity of the probiotic organism and spoilage (Garcia *et al.*, 2018).

Statistical analysis

Analysis of all the parameters were conducted in triplicates, mean and standard deviation was used to describe the continuous variables, for multivariate analysis Kruskal–Walli’s test followed by the Mann–Whitney U test was used to compare physicochemical, nutrient and antioxidant property of fermented and non-fermented oats milk drink and for bivariate samples, Mann–Whitney U test was used to compare sensory scores of spice and strawberry flavoured oats milk drink. Statistical analyses were conducted with IBM.SPSS statistics software 23.0 Version and significance was set at $p < 0.05$ for all analyses.

Results and discussion

Sensory properties

The mean score for all the sensory characteristics such as colour, flavor, consistency, mouth feel, taste and overall acceptability was above seven- over nine-point scale for both the strawberry and spice flavoured probiotic oats milk (Table 1). The mean total sensory score for spice flavored probiotic drink was higher (47.5) than the strawberry flavoured drink (45.4). Flavour is one of the major characteristics of nondairy symbiotic drink, and it is important to balance the oat flavor and the texture of the final product (Wang *et al.*, 2017). Interestingly both the spice and strawberry flavoured probiotic drink secured highest mean

Probiotic drink	Sensory characteristics					Overall	
	Colour	Flavour	Consistency	Mouth feel	Taste	acceptability	Total
Strawberry flavoured	7.7 ± 0.67 ^a	7.2 ± 0.63 ^a	7.4 ± 0.5 ^a	7.6 ± 0.69 ^a	7.8 ± 0.63 ^b	7.7 ± 0.82 ^a	45.4 ± 3.3 ^a
Spice flavoured	7.9 ± 0.99 ^a	7.8 ± 0.91 ^a	7.8 ± 0.78 ^a	7.6 ± 0.84 ^a	8.4 ± 0.69 ^a	8.0 ± 0.94 ^a	47.5 ± 4.0 ^a

Note: Mean (SD) values ($n = 10$) within a column with different lowercase letters are significantly different according to Mann–Whitney test ($p < 0.05$) in sensory characteristics between fermented spice flavoured and strawberry flavoured drink

Table 1.
Sensory characteristics of strawberry and spice flavoured probiotic oats milk drink

score for the taste attribute and differ significantly as well ($p = 0.03$). Study on sensory properties (Everitt, 2009) reported that a mean liking score of 7 or higher on a nine-point scale is usually indicative of highly acceptable sensory quality, and a product achieving this score could be used confidently as a good illustration of “target” quality. Thus, both the spice and strawberry flavoured probiotic oats milk drink presumed to provide physical references to illustrate the sensory quality that realistically represents the consumers’ acceptance limits.

Enumeration of colony counts

FAO/WHO 2002, recommends that probiotic bacteria should be present in the range of 10^6 – 10^9 CFU/mL or g in any food product at the time of ingestion, to exert their positive effect on the host. Viable cell concentration after the addition of the flavouring substance and sweeteners to the spice and strawberry flavoured probiotic oats drink was ensured by estimating the colony forming unit number on MRS-agar plate after incubation at 37°C for 24–48 h. The enumerated colony counts of spice and strawberry flavoured drink was 2.5×10^8 and 2.3×10^8 CFU/mL, respectively. Though a minimum viable number of 10^6 CFU per ml was suggested, a viable count of 10^8 – 10^9 CFU/mL or g was recommended to exert therapeutic effect (Zheng *et al.*, 2017). The viable counts of microencapsulated *L.plantarum* in both the flavoured probiotic oats milk drink ensured the prescribed viable count, intended to claim therapeutic effects and health benefits.

Physicochemical analysis

The physicochemical characteristics such as pH, titrable acidity (TA), total solids and viscosity are related to stability of bioactive components in plant derived products and not much difference observed with the values of all physicochemical parameters among both the flavours (Table 2). Adding probiotic starter culture will cause decrease in pH and increase in titrable acidity at the end of the fermentation period due to acid production (Tomovska *et al.*, 2016). pH and titrable acidity of the spice and strawberry flavoured probiotic oats drink was relevant with the above said and differed significantly ($p < 0.05$) than the nonfermented oats milk without the probiotic culture. This decrease in pH can be due to the lactic acid bacteria

Table 2. Physicochemical characteristics and nutrient content of the flavoured probiotic oats milk drink

Parameter	Fermented with microencapsulated <i>L.plantarum</i>		
	Non fermented (100 mL)	Spice flavoured (100 mL)	Strawberry flavoured (100 mL)
pH	4.3 ± 0.07 ^a	4.1 ± 0.10 ^b	4.1 ± 0.10 ^b
Titrable acidity (%)	0.34 ± 0.03 ^b	0.41 ± 0.01 ^a	0.40 ± 0.02 ^a
Total solids (%)	8.01 ± 0.28 ^c	10.9 ± 0.260 ^a	9.4 ± 0.52 ^b
Viscosity(cPS)	3.0 ^c	23.6 ± 1.52 ^a	20.6 ± 0.57 ^b
Moisture g (%)	91.9 ± 0.28 ^a	89.1 ± 0.26 ^b	90.6 ± 0.52 ^c
Energy (Kcals)	32.9 ± 1.8 ^b	34.6 ± 2.5 ^b	66.3 ± 3.2 ^a
Carbohydrate (g %)	7.3 ± 0.19 ^b	6.0 ± 0.25 ^c	13.9 ± 0.36 ^a
Protein (g %)	0.89 ± 0.10 ^c	1.1 ± 0.18 ^b	1.6 ± 0.11 ^a
Fat (g %)	0.37 ± 0.14 ^b	0.62 ± 0.06 ^a	0.43 ± 0.23 ^{ab}
Dietary fiber (g %)	4.4 ± 0.23 ^a	3.2 ± 0.37 ^b	2.8 ± 0.25 ^b
β-Glucan (g %)	0.86 ± 0.01 ^a	0.88 ± 0.01 ^a	0.87 ± 0.01 ^a

Note: Mean (SD) values ($n = 3$) within a row with different lowercase letters are significantly different according to Mann–Whitney’s test ($p < 0.05$) in physicochemical and nutrient content between non fermented and fermented spice and strawberry flavoured drink

producing organic acids, mainly lactic acid. The type of probiotic bacteria, fermentation time and their interaction significantly influences the pH (Gao *et al.*, 2019). Studies illustrate (Shah, 2007) that tolerable level of pH and triable acidity for the probiotic food products should range between 7–4.5 (pH) and 0.3–1.9% (TA), the products developed in the present study stayed well within the permissible range advocated and also understood that there would be a better survivability of *L.plantarum* in cereal based probiotic beverages. Usually, probiotic products with low pH containing encapsulated probiotic bacteria were more stable than those containing free probiotic microorganisms. Terpou *et al.* (2019) reported that encapsulated probiotic bacteria survived better in acidic environment throughout the entire storage period, whereas free probiotic bacteria showed a reduction in viability. Viscosity increased significantly in the fermented spice and strawberry flavoured probiotic drink than the nonfermented sample ($p = 0.03$). Reduced viscosity in the strawberry flavoured drink than the spice flavoured drink could be due to the diluting effect of sweetening agent added. Heating of cereal grains induces a series of structural changes, namely, gelatinization, where amylose dissolves, leaches, in water and increases the viscosity of the suspension (Stefano *et al.*, 2017).

Proximate nutrient analysis

Strawberry flavoured drink had significantly higher energy ($p = 0.05$) and carbohydrate ($p = 0.05$) content compared to the spice flavoured drink due to the addition of sweeteners. Subsequently, protein content shows significant increase after fermentation in both spice ($p = 0.05$) and strawberry ($p = 0.04$) flavoured drink (Table 2). Fermentation increases microbial protease activity which partially degrade and release some of the proteins from the matrix (Çabuk *et al.*, 2018). Protein hydrolysis during fermentation generates more peptides, free amino acids and thereby enhances the protein digestibility (Melini *et al.*, 2019), with *Lb.rhamnosus* SP1, *Weissella confusa* DSM 20194 and *Lb. plantarum* T6B10 strains (Lorusso *et al.*, 2018). Moreover, fermentation activates starch-hydrolyzing enzymes such as α -amylase and maltase which degrade starch into maltodextrins and simple sugars making it easy for digestion (Nkhata *et al.*, 2018). Low fat, moderate calorie and high fiber content in the flavoured drinks would favour healthy options for weight watchers and individuals with other comorbidities.

Though spice-flavoured drink had higher total dietary fiber content than the strawberry flavoured drink, no difference was seen in the soluble fiber β -glucan content present in both the fermented drinks and nonfermented oats milk ($p = 0.31$). It is clear that the functional ingredient β -glucan was unaffected by the probiotic culture *L.plantarum* as the quantity is maintained even after fermentation process. The US Food and Drug Administration and Health Canada have accepted 3g as an effective daily intake of oat β -glucan to reduce serum LDL cholesterol (Health Canada, 2010). Consuming 350 ml of the developed probiotic drink would provide the required quantity of oat β -glucan sufficient to contribute hypolipidemic effect.

Antioxidant property and free fatty acids present

Antioxidant activity/property is the ability of bioactive compounds to prevent, delay and protect against oxidation of various substrates. Phenolic antioxidants can thus play an important role in preventing deterioration of foods and maintaining their quality (Alu'datt *et al.*, 2019). Antioxidant supplements or foods containing antioxidants would greatly help in reducing oxidative damage, free radical scavenging rate and reducing activity of superoxide dismutase (SOD) and glutathione peroxidase (GPX) in the human body (Mishra *et al.*, 2015). Both the flavoured probiotic oats milk drink hold good antioxidant property as determined by inhibitory effect against DPPH free radical scavenging assay (Table 3). Strawberry and spice flavoured drink observed to have 46.4 and 43.3% of inhibitory effect

against maximum concentration (1 mg) of the flavoured probiotic oats drink, respectively. Antioxidant potential increased after fermentation process with microencapsulated *L.plantarum* showing significant difference between nonfermented oats milk and spice flavoured ($p = 0.05$), strawberry flavoured ($p = 0.05$) probiotic oats milk drink. Irrespective of the food substrate, fermentation with putative microbes results in the modification of inherent constituents, secondary metabolites, detoxification of toxic components/residues and improvement in the functionality of the food product (Adebo *et al.*, 2019; Adebisi *et al.*, 2019). Most lactic acid bacteria (LAB) have scavenging systems for oxygen free radicals; thus, probiotics from the LAB family can be potential candidates for the production of functional foods or natural antioxidant supplements. Kumar *et al.* (2010) in a study exhibited the antioxidative attributes of the intact cells of *L.plantarum* AS1 isolated from the South Indian fermented food kallappam. In addition, studies show that the phenolic acids in oat possess antioxidant properties. Phenolic fractions from fermented oats, demonstrated greater DPPH and 2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid (ABTS⁺) radical scavenging abilities than the unfermented oats (Bei *et al.*, 2017).

The scavenging effect and the antioxidant property perceived in the present study could be owed to the key ingredient oats and fermentation leads to increase in phenolic and flavonoid compounds due to microbial hydrolysis reaction, resulting in structural disintegration of cell walls leading to either liberation or synthesis of different antioxidant compounds (Panghal *et al.*, 2017). Consequently, fermentation may be a potential approach to improve oats' phenolic content and antioxidant capacity for functional foods in health promotion.

Strawberry and spice flavoured probiotic oats milk drink exhibited presence of 15 volatile compounds specifically free fatty acids. The highest area covered were by 1,54-dibromo-Tetrapentacontane (31.2%), Di-n-decylsulfone (66.1%) and the least was 2,4-Dimethylecosane (0.04%). Previous study reports that food fermentation mediated by probiotics can effectively lead to the essential fatty acids release which helps to modulate the intestinal mucosal wall and improves the barrier between pathogenic and beneficial microorganisms (Emadi-Koochak *et al.*, 2019). 1,54-dibromo-Tetrapentacontane reported to have antioxidant, antitumour, hypolipidemic and antiviral property (Enema *et al.*, 2019; Abdelhamid *et al.*, 2015).

Shelf life

Probiotic organism should not adversely affect the taste or aroma of the product or acidification during the shelf life of the product. Sensory properties have a vital role in

Table 3.
Antioxidant property of flavoured probiotic oats milk drink (DPPH free radical scavenging assay)

Concentration (mg)	Non fermented	Inhibition (%) Fermented with microencapsulated <i>L.plantarum</i>	
		Spice flavoured	Strawberry flavoured
0.2	5.4 ± 0.02 ^c	33.0 ± 0.01 ^b	35.1 ± 0.02 ^a
0.4	6.2 ± 0.02 ^c	34.4 ± 0.01 ^b	36.9 ± 0.01 ^a
0.6	9.8 ± 0.01 ^c	35.7 ± 0.01 ^b	39.4 ± 0.01 ^a
0.8	12.8 ± 0.02 ^c	38.1 ± 0.02 ^b	42.4 ± 0.02 ^a
1.0	16.1 ± 0.01 ^c	43.3 ± 0.02 ^b	46.3 ± 0.01 ^a

Note: Mean (SD) values ($n = 3$) within a row with different lowercase letters are significantly different according to Mann–Whitney's test ($p < 0.05$) in antioxidant property between non fermented and fermented spice and strawberry flavoured drink

product acceptance by consumers. Exploration of the shelf life of the developed probiotic oat drink at refrigerated temperature (4°C) was conducted for a period of 16 days with periodical observation of the sensory properties at every alternative day (2nd, 4th, 6th, 8th, 10th, 12th, 14th and 16th day). The observation signifies that the spice flavoured drink had acceptable sensory properties for 12 days and strawberry flavoured drink for 14 days. Both the products had viable cell counts in the range of 10^8 – 10^9 CFU/mL at the end of estimated shelf life period, which ensured the existence of desirable probiotic characteristics of the drink and evidenced that the addition of spices, sweeteners, salt or other flavouring substance did not affect the strain's viability during storage.

Conclusion

The isolated and microencapsulated *L.plantarum* strain used in this study possess good viability in both the spice and strawberry flavoured probiotic oats milk drink and had good shelf life period of two weeks without addition of any preservatives. The developed product remained to be stable without phase separation throughout the course of storage period, had tolerable pH and triable acidity, packed with essential proximate nutrients and other functional components such as β -glucan, antioxidant property and free fatty acids. It may act as good alternative to serve individuals with lactose intolerance, gluten or casein sensitivity/allergic reactions and to compliment probiotic option for vegans. The observations depicted that microbial fermentation may be used as suitable and inexpensive technology in formulation of health promoting foods with functional components.

References

- Abdelhamid, M.S., Kondratenko, E.L. and Lomteva, N.N. (2015), "GC-MS analysis of phytocomponents in ethanolic extract of *Nelumbo Nucifera* seeds from Russia", *Journal of Applied Pharmaceutical Science*, Vol. 5 No. 4, pp. 115-118, doi: [10.7324/JAPS.2015.50419](https://doi.org/10.7324/JAPS.2015.50419).
- Adebiyi, J.A., Njobeh, P.B. and Kayitesi, E. (2019), "Assessment of nutritional and phytochemical quality of Dawadawa (an African fermented condiment) produced from Bambara groundnut (*Vigna Subterranea*)", *Microchemical Journal*, Vol. 149, p. 104034, doi: [10.1016/j.microc.2019.104034](https://doi.org/10.1016/j.microc.2019.104034).
- Adebo, O.A., Kayitesi, E., Tugizimana, F. and Njobeh, P.B. (2019), "Differential metabolic signatures in naturally and lactic acid bacteria (LAB) fermented ting (a Southern African food) with different tannin content, as revealed by gas chromatography mass spectrometry (GC-MS)-based metabolomics", *Food Research International*, Vol. 121, pp. 326-335, doi: [10.1016/j.foodres.2019.03.050](https://doi.org/10.1016/j.foodres.2019.03.050).
- Alu'datt, M.H., Rababah, T., Alhamad, M.N., Gammoh, S., Alkhalidy, H.A., Al-Mahasneh, M.A., Tranchant, C.C., Kubow, S. and Masadeh, N. (2019), "Fermented malt beverages and their biomedical health potential: classification, composition, processing, and Bio-Functional properties", *Fermented Beverages*, pp. 369-400, Elsevier, doi: [10.1016/b978-0-12-815271-3.00009-9](https://doi.org/10.1016/b978-0-12-815271-3.00009-9).
- Angelov, A., Yaneva-Marinova, T. and Gotcheva, V. (2018), "Oats as a matrix of choice for developing fermented functional beverages", *Journal of Food Science and Technology*, Vol. 55 No. 7, pp. 2351-2360, doi: [10.1007/s13197-018-3186-y](https://doi.org/10.1007/s13197-018-3186-y).
- Bei, Q., Liu, Y., Wang, L., Chen, G. and Wu, Z. (2017), "Improving free, conjugated, and bound phenolic fractions in fermented oats (*Avena sativa* L.) with *Monascus anka* and their antioxidant activity", *Journal of Functional Foods*, Vol. 32, pp. 185-194, doi: [10.1016/j.jff.2017.02.028](https://doi.org/10.1016/j.jff.2017.02.028).
- Çabuk, B., Nosworthy, M.G., Stone, A.K., Korber, D.R., Tanaka, T., House, J.D. and Nickerson, M.T. (2018), "Effect of fermentation on the protein digestibility and levels of non-nutritive compounds of pea protein concentrate", *Food Technology and Biotechnology*, Vol. 56 No. 2, pp. 257-264, doi: [10.17113/ftb.56.02.18.5450](https://doi.org/10.17113/ftb.56.02.18.5450).

- Cortés, R.N.F., Martínez, M.G., Guzmán, I.V., Llano, S.L.A., Grosso, C.R.F. and Bustos, F.M. (2014), "Evaluation of modified amaranth starch as shell material for encapsulation of probiotics", *Cereal Chemistry Journal*, Vol. 91 No. 3, pp. 300-308, doi: [10.1094/CCHEM-06-13-0112-R](https://doi.org/10.1094/CCHEM-06-13-0112-R).
- Daliu, P., Santini, A. and Novellino, E. (2018), "A decade of nutraceutical patents: where are we now in 2018?", *Expert Opinion on Therapeutic Patents*, Vol. 28 No. 12, pp. 875-882, doi: [10.1080/13543776.2018.1552260](https://doi.org/10.1080/13543776.2018.1552260).
- Durazzo, A., D'Addezio, L., Camilli, E., Piccinelli, R., Turrini, A., Marletta, L., Marconi, S., Lucarini, M., Lisciani, S., Gabrielli, P., Gambelli, L., Aguzzi, A. and Sette, S. (2018), "From plant compounds to botanicals and back: a current snapshot", *Molecules*, Vol. 23 No. 8, p. 1844, doi: [10.3390/molecules23081844](https://doi.org/10.3390/molecules23081844).
- Durazzo, A., Nazhand, A., Lucarini, M., Atanasov, A.G., Souto, E.B., Novellino, E., Capasso, R. and Santini, A. (2020), "An updated overview on nanonutraceuticals: focus on nanoprebiotics and nanoprobiotics", *International Journal of Molecular Sciences*, Vol. 21 No. 7, p. 2285, doi: [10.3390/ijms21072285](https://doi.org/10.3390/ijms21072285).
- Earle, R.L. and Earle, M.D. (2004), "Unit operations in food processing", Web Edition. The New Zealand Institute of Food Science and Technology (Inc.), available at: www.nzifst.org.nz/resources/unitoperations/index.htm.
- Emadi-Koochak, H., Siami, Z., Zebardast, J., SeyedAlinaghi, S. and Asadollahi-Amin, A. (2019), "Effect of probiotic consumption on increasing the CD4+ T cell counts among Iranian patients living with HIV: a double-blind randomized clinical trial", *Journal of Health Research*, Vol. 34 No. 2, pp. 123-133, doi: [10.1108/JHR-04-2019-0084](https://doi.org/10.1108/JHR-04-2019-0084).
- Enema, O.J., Umoh, U.F., Thomas, P.S., Adesina, S.K. and Eseyin, O.A. (2019), "Phytochemical and antioxidant studies of leaf of *Tetrapleura Tetraptera* (Schum and thon) Taubert (Mimosaceae)", *British Journal of Pharmaceutical and Medical Research*, Vol. 04 No. 03, pp. 1865-1875, doi: [10.24942/bjpmr.2019.490](https://doi.org/10.24942/bjpmr.2019.490).
- Everitt, M. (2009), "Consumer-Targeted sensory quality", in Canovas, G.B., Mortimer, A., Lineback, D., Spiess, W., Buckle, K. and Colonna, P. (Eds), *Global Issues in Food Science and Technology*, Academic Press, pp. 117-128.
- Fenster, K., Freeburg, B., Hollard, C., Wong, C., Laursen, R.R. and Ouwehand, A.C. (2019), "The production and delivery of probiotics: a review of a practical approach", *Microorganisms*, Vol. 7 No. 3, p. 83, doi: [10.3390/microorganisms7030083](https://doi.org/10.3390/microorganisms7030083).
- Food and Agriculture Organization (FAO)/World Health Organization (WHO) (2002), "*Guidelines for the Evaluation of Probiotics in Food*", *Report of a Joint FAO/WHO Working Group on Drafting Guidelines for the Evaluation of Probiotics in Food*, FAO London, Ontario, Canada, pp. 1-11.
- Gadow, A.V., Joubert, E. and Hansmann, C.F. (1997), "Comparison of antioxidant activity of Aspalathin with that of other plant phenols of rooibos tea (*Aspalathon linearis*) α -tocopherol, BHT and BHA", *Journal of Agricultural and Food Chemistry*, Vol. 45 No. 3, pp. 632-638, doi: [10.1021/jf960281n](https://doi.org/10.1021/jf960281n).
- Gao, Y., Hamid, N., Gutierrez-Maddox, N., Kantono, K. and Kitundu, E. (2019), "Development of a probiotic beverage using breadfruit flour as a substrate", *Foods (Basel, Switzerland)*, Vol. 8 No. 6, p. 214, doi: [10.3390/foods8060214](https://doi.org/10.3390/foods8060214).
- Garcia, E.F., de Oliveira Araujo, A., Luciano, W.A., de Albuquerque, T.M.R., de Oliveira Arcanjo, N.M., Madruga, M.S., Dos Santos Lima, M., Magnani, M., Saarela, M. and de Souza, E.L. (2018), "The performance of five fruit-derived and freeze-dried potentially probiotic lactobacillus strains in apple, orange, and grape juices", *Journal of the Science of Food and Agriculture*, Vol. 98 No. 13, pp. 5000-5010, doi: [10.1002/jsfa.9034](https://doi.org/10.1002/jsfa.9034).
- Gibson, G.R., Hutkins, R., Sanders, M.E., Prescott, S.L., Reimer, R.A., Salminen, S.J., Scott, K., Stanton, C., Swanson, K.S., Cani, P.D., Verbeke, K. and Reid, G. (2017), "Expert consensus document: the international scientific association for probiotics and prebiotics (ISAPP) consensus statement on the definition and scope of prebiotics", *Nature Reviews Gastroenterology and Hepatology*, Vol. 14 No. 8, pp. 491-502, doi: [10.1038/nrgastro.2017.75](https://doi.org/10.1038/nrgastro.2017.75).

- Guevarra, R.B. and Barraquio, V.L. (2015), "Viable counts of lactic acid bacteria in Philippine commercial yogurts", *International Journal of Dairy Science and Processing*, Vol. 2 No. 5, pp. 24-28, doi: [10.19070/2379-1578-150008](https://doi.org/10.19070/2379-1578-150008).
- Hassanzadazar, H., Ehsani, A., Mardani, K. and Hesari, J. (2012), "Investigation of antibacterial, acid and bile tolerance properties of lactobacilli isolated from Koozeh cheese", *Veterinary Research Forum: An International Quarterly Journal*, Vol. 3 No. 3, pp. 181-185.
- Health Canada (2010), "Oat products and blood cholesterol lowering, summary of assessment of a health claim about oat products and blood cholesterol lowering", available at: www.hc-sc.gc.ca/fn-an/alt_formats/pdf/label-etiquet/claims-reclam/assess-evalu/oat_avoine-eng.pdf
- Indian Council of Medical Research Task Force (2011), "ICMR-DBT guidelines for evaluation of probiotics in food", *The Indian Journal of Medical Research*, Vol. 134 No. 1, pp. 22-25.
- Kechagia, M., Basoulis, D., Konstantopoulou, S., Dimitriadi, D., Gyftopoulou, K., Skarmoutsou, N. and Fakiri, E.M. (2013), "Health benefits of probiotics: a review", *ISRN Nutrition*, Vol. 2013, doi: [10.5402/2013/481651](https://doi.org/10.5402/2013/481651).
- Kumar, M., Kumar, A., Nagpal, R., Mohania, D., Behare, P., Verma, V., Kumar, P., Poddar, D., Aggarwal, P.K., Henry, C.J., Jain, S. and Yadav, H. (2010), "Cancer-preventing attributes of probiotics: an update", *International Journal of Food Sciences and Nutrition*, Vol. 61 No. 5, pp. 473-496, doi: [10.3109/09637480903455971](https://doi.org/10.3109/09637480903455971).
- Lorusso, A., Coda, R., Montemurro, M. and Rizzello, C.G. (2018), "Use of selected lactic acid bacteria and quinoa flour for manufacturing novel yogurt-like beverages", *Foods*, Vol. 7 No. 4, p. 51, doi: [10.3390/foods7040051](https://doi.org/10.3390/foods7040051).
- Melini, F., Melini, V., Luziatelli, F., Ficca, A.G. and Ruzzi, M. (2019), "Health-promoting components in fermented foods: an up-to-date systematic review", *Nutrients*, Vol. 11 No. 5, p. 1189, doi: [10.3390/nu11051189](https://doi.org/10.3390/nu11051189).
- Mishra, V., Shah, C., Mokalsh, N., Chavan, R., Yadav, H. and Prajapati, J. (2015), "Probiotics as potential antioxidants: a systematic review", *Journal of Agricultural and Food Chemistry*, Vol. 63 No. 14, pp. 3615-3626, doi: [10.1021/jf506326t](https://doi.org/10.1021/jf506326t).
- Mopuri, R. and Meriga, B. (2014), "Anti-Lipase and anti-Obesity activities of *Terminalia Paniculata* bark in high calorie diet-induced obese rats", *Global Journal of Pharmacology*, Vol. 8 No. 1, pp. 114-119.
- Ningtyas, D.W., Bhandari, B., Bansal, N. and Prakash, S. (2019), "The viability of probiotic *Lactobacillus Rhamnosus* (non-encapsulated and encapsulated) in functional reduced-fat cream cheese and its textural properties during storage", *Food Control*, Vol. 100, pp. 8-16, doi: [10.1016/j.foodcont.2018.12.048](https://doi.org/10.1016/j.foodcont.2018.12.048).
- Nkhata, S.G., Ayua, E., Kamau, E.H. and Shingiro, J.B. (2018), "Fermentation and germination improve nutritional value of cereals and legumes through activation of endogenous enzymes", *Food Science and Nutrition*, Vol. 6 No. 8, pp. 2446-2458, doi: [10.1002/fsn3.846](https://doi.org/10.1002/fsn3.846).
- Panghal, A., Virkar, K., Kumar, V., Dhull, S.B., Gat, Y. and Chhikara, N. (2017), "Development of probiotic beetroot drink", *Current Research in Nutrition and Food Science Journal*, Vol. 5 No. 3, pp. 257-262, doi: [10.12944/CRNFSJ.5.3.10](https://doi.org/10.12944/CRNFSJ.5.3.10).
- Pant, G., Simaria, C., Varsi, A.H., Bhan, P. and Sibi, G. (2015), "In vitro anti-Cholesterol and antioxidant activity of methanolic extracts from flax seeds (*linum usitatissimum L.*)", *Research Journal of Medicinal Plant*, Vol. 9 No. 6, pp. 300-306, doi: [10.3923/rjmp.2015.300.306](https://doi.org/10.3923/rjmp.2015.300.306).
- Paszczyk, B., Luczynska, J. and Tonska, E. (2017), "Fatty acid profile and trans fatty acids content in cereals and cereal bars from polish market", *Polish Journal of Natural Sciences*, Vol. 32 No. 4, pp. 733-743.
- Putta, S., Yarla, N.S., Lakkappa, D.B., Imandi, S.B., Malla, R.R., Chaitanya, A.K., Chari, B.P.V., Saka, S., Vechalapu, R.R., Kamal, M.A., Tarasov, V.T., Chubarev, V.V., Kumar, K.S. and Aliev, G. (2018), "Probiotics: supplements, food, pharmaceutical industry", in Grumezescu, A.M. and Holban, A. M. (Eds), *Therapeutic, Probiotic, and Unconventional Foods*, Elsevier, Academic Press, Cambridge, MA, pp. 15-25, doi: [10.1016/B978-0-12-814625-5.00002-9](https://doi.org/10.1016/B978-0-12-814625-5.00002-9).

- Randox Laboratories (2009), *Cholesterol (Chol) Enzymatic Endpoint Method Manual*, Randox Laboratories Pvt. Ltd.
- Salmeron, I. (2017), "Fermented cereal beverages: from probiotic, prebiotic and synbiotic towards nanoscience designed healthy drinks", *Letters in Applied Microbiology*, Vol. 65, pp. 114-124, doi: [10.1111/lam.12740](https://doi.org/10.1111/lam.12740).
- Sangami, R. and RadhaiSri, S. (2018), "Extraction and standardization of indigenous oat milk", *International Journal of Current Advanced Research*, Vol. 07 No. 8, pp. 14912-14915, doi: [10.24327/ijcar.2018.14915.2722](https://doi.org/10.24327/ijcar.2018.14915.2722).
- Sarkar, S. (2019), "Potentiality of probiotic yoghurt as a functional food – a review", *Nutrition and Food Science*, Vol. 49 No. 2, pp. 182-202, doi: [10.1108/NFS-05-2018-0139](https://doi.org/10.1108/NFS-05-2018-0139).
- Semedo, M.C., Karmali, A. and Fonseca, L.P. (2015), "A high throughput calorimetric assay of β – 1, 3-D-Glucans by congo red dye method", *Journal of Microbiological Methods*, Vol. 109, pp. 140-148, doi: [10.1016/j.mimet.2014.12.020](https://doi.org/10.1016/j.mimet.2014.12.020).
- Shah, N.P. (2007), "Functional cultures and health benefits", *International Dairy Journal*, Vol. 17 No. 11, pp. 1262-1277, doi: [10.1016/j.idairyj.2007.01.014](https://doi.org/10.1016/j.idairyj.2007.01.014).
- Shori, A.B. (2017), "Microencapsulation improved probiotics survival during gastric transit", *HAYATI Journal of Biosciences*, Vol. 24 No. 1, pp. 1-5, doi: [10.1016/j.hjb.2016.12.008](https://doi.org/10.1016/j.hjb.2016.12.008).
- Stefano, E.D., White, J., Seney, S., Hekmat, S., McDowell, T., Sumarah, M. and Reid, G. (2017), "A novel Millet-Based probiotic fermented food for the developing world", *Nutrients*, Vol. 9 No. 5, p. 529, doi: [10.3390/nu9050529](https://doi.org/10.3390/nu9050529).
- Tanaka, H., Doesburg, K., Iwaski, T. and Mierau, I. (1999), "Screening of lactic acid bacteria for bile salt hydrolase activity", *Journal of Dairy Science*, Vol. 82 No. 12, pp. 2530-2535, doi: [10.3168/jds.S0022-0302\(99\)75506-2](https://doi.org/10.3168/jds.S0022-0302(99)75506-2).
- Terpou, A., Papadaki, A., Lappa, I.K., Kachrimanidou, V., Bosnea, L.A. and Kopsahelis, N. (2019), "Probiotics in food systems: significance and emerging strategies towards improved viability and delivery of enhanced beneficial value", *Nutrients*, Vol. 11 No. 7, p. 1591, doi: [10.3390/nu11071591](https://doi.org/10.3390/nu11071591).
- Tomovska, J., Gjorgievski, N. and Makarijoski, B. (2016), "Examination of pH, titratable acidity and antioxidant activity in fermented milk", *Journal of Materials Science and Engineering A*, Vol. 6, pp. 326-333, doi: [10.17265/2161-6213/2016.11-12.006](https://doi.org/10.17265/2161-6213/2016.11-12.006).
- Wang, C., Liang, S., Wang, H. and Guo, M. (2017), "Physiochemical properties and probiotic survivability of symbiotic oat-based beverage", *Food Science and Biotechnology*, Vol. 27 No. 3, pp. 735-743, doi: [10.1007/s10068-017-0290-0](https://doi.org/10.1007/s10068-017-0290-0).
- Wichchukit, S. and Omahony, M. (2015), "The 9-point hedonic scale ranking in food science: some reappraisals and alternatives", *Journal of the Science of Food and Agriculture*, Vol. 95 No. 11, pp. 2167-2178, doi: [10.1002/jsfa.6993](https://doi.org/10.1002/jsfa.6993).
- Wilkinson, M.G. (2018), "Flow cytometry as a potential method of measuring bacterial viability in probiotic products: a review", *Trends in Food Science and Technology*, Vol. 78, pp. 1-10, doi: [10.1016/j.tifs.2018.05.006](https://doi.org/10.1016/j.tifs.2018.05.006).
- Zheng, M., Zhang, R., Tian, X., Zhou, X., Pan, X. and Wong, A. (2017), "Assessing the risk of probiotic dietary supplements in the context of antibiotic resistance", *Frontiers in Microbiology*, Vol. 8, p. 908, doi: [10.3389/fmicb.2017.00908](https://doi.org/10.3389/fmicb.2017.00908).

Corresponding author

Sangami Ravindran can be contacted at: sangamiravindran@gmail.com

For instructions on how to order reprints of this article, please visit our website:

www.emeraldgrouppublishing.com/licensing/reprints.htm

Or contact us for further details: permissions@emeraldinsight.com