

Non-dairy plant-based milk products as alternatives to conventional dairy products for delivering probiotics

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Abstract

Dairy products are important to the human diet and are one of the four essential food groups. Conventional fermented cow milk products possessed a variation of bioactive components that enhance the human body's physiological processes. However, traditional milk products can be replaced by non-dairy plant-based milk which is produced mainly from fruits and seeds such as soy, coconut, almond, rice, peanut, lupin, cashew, and hemp. Fermented plant-based milk is the outcome of intense bacterial activity of the starter cultures, leading to the production of lactic acid and biologically active compounds that adding nutritional and physiological value. In addition, the growth and viability of probiotics in plant-based milk are depended on several factors such as milk composition, fermentation process, type of probiotics, storage time and temperature, acidity, and packages. Therefore, this review focuses on the growth and viability of probiotics in fermented plant-based milk products during fermentation and refrigerated storage.

Keywords: viability; probiotics; plant-based milk; fermentation; soy milk; coconut milk.

Practical Application: Plant-based milk could be used to replace traditional dairy products and improve human health.

1 Introduction

Dairy products are considered to be good for health (Lee et al., 2020). They are one of the four essential food groups (Shori et al., 2018). Furthermore, the beneficial health effects of consuming these products have been shown in many studies (Lucatto et al., 2020; Eor et al., 2020; Hadjimbei et al., 2020; Pena et al., 2021). Nowadays, as the popularity of being “vegan” is increasing, traditional milk products can be replaced by plant-based dairy which is produced mainly from nuts and cereals such as soy, almond, rice, oat, and coconut milk (Shori, 2013a). Several plant-based dairy products have a long tradition in both Eastern and Western cultures and available widely in the market (Bernat et al., 2014). Although some plant milk products contain low protein and calcium, plant milk substitutes are used to replace cow's milk in the diet because of low allergy and intolerance issues, lactose-free, cholesterol-free, and low-calorie (Mäkinen et al., 2016). This may lead to increased consumer awareness and subsequently a rise in purchase levels (Jeske et al., 2018). Actually, these products' marketing campaigns attempt to equate the healthiness of these substitutes with the original dairy product. In addition, previous studies have reported a therapeutic relationship between vegetable milk and diseases such as cancer, atherosclerosis, and inflammatory diseases with a good source of antioxidants (Bernat et al., 2014).

The fermentation of plant-based products is one of the most traditional methods for food preservation. It improves the sensory attributes of the final products as well as nutritional quality by reducing the content of sugars and enhancing the levels of thiamine, niacin, lysine (Jeske et al., 2018; Rasika et al., 2020). Now, more than five thousand various fermented food

products are consumed by people around the globe (Ryan et al., 2020; Balthazar et al., 2021). Moreover, fermentation enhanced the digestion of the proteins in boiled soybean by 45% which affected human health. This is due to the increase in the essential amino acids (Ketnawa & Ogawa, 2019).

Plant-based milk can be preserved by lactic acid fermentation which results in organic acid production and antimicrobials components (e.g., acetaldehyde and diacetyl; Chinsebu et al., 2015). Fermentation also improves the nutritional value of milk by increasing amino-acids, vitamins as well as therapeutic values such as anti-microbial, anti-tumor, anti-carcinogenic, and immunomodulation activity (Tangyu et al., 2019; Grom et al., 2020; Costa et al., 2020).

Today, demand for probiotic plant-based milk products is growing due to consumer's awareness of potential health advantages since probiotics enhance the balance and structure of microbiota and the protection against pathogenic species (Panghal et al., 2018). To assure the health benefits of fermented plant-based milk products, probiotics should meet the minimum level requirement for probiotic bacteria between 10^6 and 10^7 cfu/mL until the expiry date (Shori et al., 2018). Therefore, this review focuses on the growth and viability of probiotics in fermented plant-based milk products during fermentation and refrigerated storage.

2 Probiotics and health effects in plant-based milk products

Probiotics have been researched extensively for their health benefits. The Food and Agriculture Organization and the World

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Health Organization define probiotics as “live microorganisms which when ingested in enough amounts improve the health of the host (Hill et al., 2014; Shori et al., 2020). Efficacious probiotics must-have essential properties as follows: strong stability under storage and distribution conditions, must be non-pathogenic, non-toxic, sustainable in the host body, effective adhesion, resistant to low pH and bile salts in the gut, and should have good activity with beneficial effects on the host (e.g., improved immunity; Shori, 2021; Zendeboodi et al., 2020).

A previous study has indicated that fermented almond milk by lactic acid bacteria possesses antioxidative properties due to phenolic contents which may minimize oxidative stress-related diseases such as atherosclerosis, coronary heart disease, and cancer (Wansutha et al., 2018). Similar results have shown that the fermentation of soy milk increased antioxidative, ACE inhibitory, antibacterial, and anti-inflammatory effects as compared to unfermented soy milk (Shori, 2013b; Singh et al., 2020; Sadeghi et al., 2020). In addition, Miraghajani et al., (2019) showed a strong relationship between intake of fortified soy milk with *L. plantarum* and improvement of renal function for type 2 diabetic kidney disease, where consumption of probiotic soymilk led to a substantial decrease in the levels of renal function biomarkers cysteine C (Cys-C) and inflammatory adipokine progranulin (PGRN) levels compared with the soy milk without probiotics. Fermentation of soy milk by the riboflavin-producing strain *Lactobacillus plantarum* CRL 2130 has prevents experimental colitis induced by trinitrobenzene sulfonic in a murine model (Levit et al., 2017). In addition, the fermented brown rice by *Aspergillus oryzae* has been reported to suppress inflammation and has the ability to prohibit the chemical carcinogenesis of the colon (Tasdemir & Sanlier, 2020).

3 Type of plant-based milk

Healthy food supplies are becoming an ever-present problem for both consumers and companies alike due to the concern of feeding an increasing number of people each year. Therefore, plant-based dairy alternatives have been used for decades, and day by day the sensory attractiveness increases the acceptability of plant-based dairy (Jeske et al., 2018). In addition, plant-based dairy sales in the U.S. have grown 61% from 2012 to 2017 (Mintel Press Team, 2018) achieving \$1.9 billion by Good Food Institute (2019). Non-conventional dairy substitutes of plant origin have been developed including soybean, almond, oat, coconut, rice, hemp, peanut, and cashew (Clay et al., 2020).

3.1 Soy milk

Soy milk provides proteins of great quality with low-cost production. In addition, it has a good level of kcal (52/100 g), proteins (3.9 g/100 g), fats (2.4 g/100 g), saturated fatty acids (14%/100 g), mono-unsaturated fatty-acids (21.6%/100 g), and poly-unsaturated fatty-acids (63.5%/100 g) as compared to cow milk (Mazumder & Begum, 2016). Previous studies have investigated that protein-rich soy products could reduce total cholesterol, low-density lipoproteins (LDLs), and triglycerides (Weiße et al., 2010). In addition, fermented soy products have been found to be effective in reducing the symptoms of diabetes

mellitus, blood pressure, heart attacks, and cancer-related complications (Jayachandran & Xu, 2019).

3.2 Coconut milk

Coconut milk has been used as a non-dairy substitute since 2010. It contains nutritious properties (i.e. carbohydrate, lipid, proteins, and potassium) and therapeutic values such as antioxidant activity (Daramola et al., 2016). In addition, coconut milk has an average of 70 kcal /100 mL compared to cow milk that has up to 150 kcal /100 mL (Katz, 2018). The calcium content in coconut milk is about 4% of daily calcium needs. In addition, it is enriched with vitamins and minerals such as vitamin C, E, B1, B3, B5, B6, iron, and phosphorus. Coconut milk contains 17% of saturated fats. However, these fats are easily metabolized by the body and almost 87% of these fats are saturated with lauric acid (44%), followed by caprylic and capric acids (13%). The main saturated fat with lauric acid is also found in mother's milk and has been shown to promote brain development and bone health (D'Amato et al., 2012; Paul et al., 2020).

3.3 Almond milk

The almond contains many nutrients, phytochemicals, and fatty acids which tremendously reduce cardiovascular risk (Manzoor, 2017). Almond milk was discovered in the early '90s and has no lactose, cholesterol, or saturated fat. Almond milk is suitable for people suffering from lactose intolerance and contains low calorie ranges from 30-60 kcals/ 100 mL. Moreover, it is cheaper than cow milk and contents 3.0% ash, 3.4% fats, 1.7% proteins, and 4.5% carbohydrates (Manzoor, 2017). Almond milk supplies about 20% of vitamin E, 4% of riboflavin, and 2% of iron. In addition, other minerals such as calcium, magnesium, phosphorus, zinc, and potassium have been found in almond milk (Vanga & Raghavan, 2018). It also acts as antioxidants, anti-inflammatory, antihyperlipidemic, antitumor, reduces platelet aggregation or change for blood clots, and enhances the immune system (Barreira et al., 2008; National Institutes of Health, 2008).

3.4 Rice-milk

Health-wise, there are no added benefits of consuming rice-milk, it is, however, considered the most hypoallergenic when compared to all the other plant-based milk. Rice-milk is extracted from boiled brown rice and brown rice starch, it has a thin consistency and no fiber (Wongthawewatana et al., 2021). Despite rice-milk content less protein than cow's milk (only 1 g per 8 oz. cup) and a small amount of natural calcium, the rice-milk products available in the market are fortified with calcium and vitamins (i.e., A, D, and B12; Bridges, 2018).

Un-malted brown rice flour contains 7.10% protein, 1.26% fat, 1.05% ash, 1.17% fiber, and 89.42% carbohydrate (Abou-Dobara et al., 2016). However, the average chemical composition of rice-milk was as follows protein (0.6%), carbohydrates (10.6%), sugar (4.0%), fat (1.0%), and sodium (0.051%). Abou-Dobara et al., (2016) reported that rice-milk promoted immunity and possessed antimicrobial activity due to the presence of selenium and magnesium.

3.5 Peanut milk

Peanut milk is a yellow liquid with almost no fat and high protein content. It is prepared by grinding raw peanuts with water (1:6) for 30 min and the pH is adjusted to 9.0 before remove the fat using a cream separator (Arya et al., 2016). Peanut milk can be fermented by lactic acid bacteria to produce a beverage (Kadam & Salunkhe, 1989). Although peanut milk is not popular, it is used extensively in low-income countries to overcome severe malnourishment and physical health. It has high mineral content, proteins, non-soluble fibers, and fatty acids such as linoleic, oleic acid, and phytic acids. In addition, peanuts are found to be enriched with p-coumaric acid which is known to be beneficial as antioxidants (Bansal et al., 2016). Phenolic compounds such as resveratrol, phenolic acid, flavonoid, and phytosterol are present in peanuts and reported to prevent the absorption of cholesterol from the food (Arya et al., 2016).

3.6 Hemp milk

Hemp milk is a natural beverage with high nutritional value. It is made by mixing hemp seeds with hot water then milled until the required consistency is reached and thereafter cooled down and this technique is carried out to extend the product shelf life (Curl et al., 2020). Hemp seeds contain protein such as arginine with several sulfur-rich proteins (20-25%), carbohydrates (20-30%), insoluble fiber (10-15%), oil (25-35%), vitamins such as vitamin A, and minerals, particularly phosphorous, potassium, magnesium, sulfur, calcium, iron, and zinc (Ustun-Argon, 2019). Hemp seeds have been reported to possess diverse biological activity such as antioxidant activity and lowering blood pressure (Aluko, 2017). In addition, Szparaga et al. (2019) have concluded that hemp milk could be a good carrier for probiotic bacteria such as *Lactobacillus casei* subsp. *rhamnosus*.

3.7 Lupin milk

Lupin is widely used in the preparation of plant-based milk (Afolabi et al., 2018). Lupin milk is prepared by grinding lupin seeds with hot water for 5 min at high speed to get homogenized lupin milk and filter the slurry through a cheesecloth before cool-down (Elsamani et al., 2014). It has protein content (30%), dietary fiber content (16%), and fat content (6%). In addition, lupin seeds have 33% and 18% of the recommended daily intake of thiamine and riboflavin, respectively (Oliveira et al., 2014). They have also large concentrations of polyphenol, carotenoid, phytosterol, tocopherol, and peptide with antioxidants, antimicrobials, anti-cancer, and anti-inflammatory activities (Khan et al., 2015). There is an increase in the demand for lupin-rich food products due to low glycemic index and higher protein content, and thus are beneficial for obese and diabetic patients with hypertension (Al-Saedi et al., 2020). In addition, the lactic fermentation of lupin milk to produce yogurt and ice cream has been previously reported (Jiménez-Martínez et al., 2003; Elsamani, 2016).

3.8 Cashew milk

Cashew milk is prepared by milling cashew nuts with water 1:3 (w/v) ratio and the resultant slurry is filtered by using

cheesecloth (Manzoor et al., 2017). The cashew nut milk products can serve as a vegetable protein source (23%) with the potential of reducing the incidence of protein-energy malnutrition (Nair, 2010; Bruno et al., 2019; Oyeyinka et al., 2019). It is known as a rich source of essential fatty acids (44%) and unsaturated fats (82%). Cashew nuts are increasingly being consumed for functional benefits beyond their nutrition (Shori et al., 2022). The health benefits of cashew nuts are lowering the cholesterol level in the blood, controlling diabetes and coronary heart disease risk, maintaining healthy bones, and preventing high blood pressure (Tola & Mazengia, 2019).

4 The growth and viability of probiotics in plant-based milk products

4.1 Soymilk

Recently, soy products have gained much attention as probiotic carriers. The nutritional value of fermented soymilk products differs depending on the kind of probiotic strain such as lactic acid bacteria (LAB). In addition, the presence of oligosaccharides, amino acids, and peptides in soymilk could enhance the growth and viability of probiotics (Farnworth et al., 2007; Šertović et al., 2020). The previous studies have documented the effectiveness of soy milk as a good substrate for the growth of probiotic and LAB strains (Table 1). Myagmardorj et al., (2018) have reported that soymilk increased the viable cell count of *Lactobacillus fermentum* BM-325 to 12.6 log cfu/mL during 20 hours of fermentation. Seven strains of *Lactobacillus* spp (*L. helveticus* V3, *L. rhamnosus* NS6, *L. rhamnosus* NS4, *L. bulgaricus* NCDC 09, *L. acidophilus* NCDC 15, *L. acidophilus* NCDC 298, *L. helveticus* NCDC 292) in fermented soymilk have been studied by Hati et al., (2018). The viable cell counts of all the seven strains ranged between 6-9 log cfu/mL for 24 hours at 37 °C. In addition, the viability of *L. casei* PLA5 was significantly ($p < 0.05$) increased by 1.24 and 1.09 log cfu/mL in fermented soymilk supplemented with maltodextrin and fructooligosaccharides; respectively (Kumari et al., 2018). This is possibly attributable to its ability to produce various glycosyl hydrolases that hydrolyses maltodextrin and fructooligosaccharides to glucose for growth (Liong & Shah, 2006; Silva et al., 2018). Similar results were observed by (Yeo & Liong, 2010).

Soymilk kefir-based functional beverage significantly increased the viability of *L. bulgaricus* and *Streptococcus thermophiles* during 28 days of storage (Silva et al., 2018; Table 1). Similar results have been demonstrated in fermented soymilk and black bean milk with kefir grains (Liu & Lin, 2000; Lim et al., 2019). Fermented soymilk with FOS in the presence of *L. plantarum*, *L. acidophilus*, or *L. rhamnosus* was investigated by Mishra & Mishra, (2013). The viability of *L. plantarum* and *L. acidophilus* was significantly enhanced ranging between 14 – 12 log cfu/mL during 28 days at 4 °C whereas the VCC of *L. rhamnosus* was reduced to 9.51 log cfu/mL. However, the viability of *L. rhamnosus* in binary and/or mixture with *L. plantarum* and *L. acidophilus* was improved in fermented soymilk with FOS. This finding is consistent with Horáčková et al. (2015) and Yi et al. (2020) who have indicated that mixed strains tend to be more effective than pure culture. The growth and viability of probiotics in fermented soymilk

Table 1. Viable cell counts (VCC) of probiotics in soymilk during fermentation and refrigerated storage.

Type of products	Probiotic bacteria	PH	Titratable acidity (TA)	VCC (Log cfu/mL)	Temperature	Time	P- value	References
Fermented soy milk (control)	<i>Lactobacillus casei</i> PLA5	6.79 - 4.13 4.1 - 3.81	0.01 - 0.09%	4.21 - 9.42 8.11 - 7.6	30 °C 4 °C	0 - 48 h 4 - 14 d	p < 0.05	(Kumari et al., 2018)
Fermented soymilk with maltodextrin	<i>Lactobacillus casei</i> strain Shirota	6.80 - 4.64 4.6 - 4.12	0.01 - 0.07%	4.13 - 9.19 7.83 - 7.2	30 °C	24 h		
Fermented soymilk with fructooligosaccharides	<i>Lactobacillus casei</i> PLA5	6.75 - 4.23 6.71 - 4.32	0.01 - 0.12% 0.01 - 0.09%	5.15 - 10.25 4.91 - 10.1	30 °C			
Fermented soymilk + Fructooligosaccharides	<i>Lactobacillus acidophilus</i> <i>Lactobacillus plantarum</i> <i>Lactobacillus rhamnosus</i> Mixed culture <i>Lactobacillus acidophilus</i> and <i>Lactobacillus plantarum</i>	4.48 - 4.1 4.49 - 4.09 4.52 - 4.1 4.42 - 4.0	N.D	13.56 - 11.63 13.64 - 12.08 13.8 - 9.51 13.21 - 11.53	4 °C	1 - 28 d	P < 0.05	(Mishra & Mishra, 2013)
Fermented soymilk drinks without any additives (control)	Mixed culture <i>Lactobacillus acidophilus</i> and <i>Lactobacillus rhamnosus</i> Mixed culture <i>Lactobacillus plantarum</i> and <i>Lactobacillus rhamnosus</i> Mixed culture <i>Lactobacillus acidophilus</i> <i>Lactobacillus plantarum</i> and <i>Lactobacillus rhamnosus</i> Yogurt culture <i>Streptococcus thermophilus</i> and <i>Lactobacillus bulgaricus</i>	4.41 - 3.9 4.4 - 4.2 4.4 - 4.0 4.51 - 4.23	0.43 - 0.77%	13.41 - 11.34 13.08 - 10.01 13.34 - 11.02 12.89 - 10.14 13.15 - 11.43 13.31 - 11.12 12.89 - 10.21 12.74 - 10.76 12.69 - 11.23				
Fermented soymilk drinks with 4% sucrose without the fruit pulp	<i>Streptococcus salivarius</i> spp. <i>Thermophilus</i>	4.92 - 4.63	0.53 - 0.93%	7.56 - 6.58 7.34 - 5.89 7.34 - 6.14 6.61 - 5.72				
Fermented soymilk drinks with banana and 4% sucrose	<i>Lactobacillus delbrueckii</i> spp. <i>Bulgaricus</i>	4.81 - 4.33	0.8 - 1.24%	8.23 - 7.88 8.02 - 7.1 8.12 - 7.58 8.14 - 7.62				
Fermented soymilk drinks with guava pulp and 4% sucrose	<i>Lactobacillus acidophilus</i> LA-5	4.82 - 4.41	0.75 - 1.15%	8.16 - 7.71 8.02 - 6.86 8.15 - 7.36 8.11 - 7.51	5 °C	Fresh - 21 d	p ≤ 0.05	(Ismaiel et al., 2018)
Fermented soymilk drinks with mango pulp and 4% sucrose	<i>Bifidobacterium</i> BB-12	4.84 - 4.46	0.72 - 1.05%	8.04 - 7.46 7.93 - 6.57 8.02 - 7.23 8.07 - 7.32				
Soymilk (Control)	<i>Lactobacillus plantarum</i> 70810	6.5 - 4.5 6.5 - 4.2	N.D	8.1 - 9.2 8.02 - 9.3	37 °C	0 - 8 h	p < 0.05	(Xindong et al., 2016)
Soymilk kefir-based functional beverage with peach-flavor	<i>Lactobacillus bulgaricus</i> and <i>Streptococcus thermophilus</i>	4.5 - 4.3	N.D	7.3 - 7.0	5 °C	0 - 28 d		
Soymilk kefir-based functional beverages with soymilk kefir		4.5 - 4.6	0.5 - 0.6%	7.5 - 8.2	37 °C	16 h	P < 0.05	(Silva et al., 2018)
Fermented soymilk without kefir grains		6.70 6.64	-	N.D N.D				

N.D. = not detected.

Table 1. Continued...

Type of products	Probiotic bacteria	PH	Titratable acidity (TA)	VCC (Log cfu/mL)	Temperature	Time	P- value	References
Fermented soymilk with kefir grains	<i>Lactobacilli</i>	6.18 5.97 6.79 5.77 6.15 5.68	-	5.83 5.88 N.D N.D 5.00 5.91	15 °C, 20 °C	24 h	P < 0.05	(Lim et al., 2019)
Fermented black bean milk without kefir grains		6.90 - 5.35 6.04 - 5.05 6.92 - 5.74	0.13 - 0.24% 0.18 - 0.35% 0.10 - 0.23%	7.54 - 9.34 8.02 - 9.47 7.00 - 9.17				
Fermented black bean milk with kefir grains	<i>Lactobacillus acidophilus</i> <i>Lactobacillus parabases</i> <i>Bifidobacterium lactis</i>							
Chocolate mousse using soy milk		6.94 - 4.48 6.67 - 3.98 6.85 - 5.07	0.07 - 0.89% 0.13 - 1.27% 0.08 - 0.43%	7.17 - 9.36 8.10 - 9.62 7.62 - 8.69	4 °C	1 - 21 d	p ≤ 0.05	(Taghizadeh et al., 2018)
Chocolate mousse using milk and soy milk		5.51 5.50 5.49 5.50	0.27% 0.27% 0.36% 0.27%	7.78 - 6.75 7.55 - 5.26 7.84 - 5.38 8.20 - 6.26				
Frozen fermented desserts 100% soymilk 75% soymilk + 25% bovine milk 50% soymilk + 50% bovine milk 25% soymilk + 75% bovine milk	<i>Lactobacillus acidophilus</i> (La-05)							
Fermented soymilk	<i>Bifidobacterium bifidum</i> (Bb-12)	5.51 5.50 5.49 5.50	0.27% 0.27% 0.27% 0.27%	8.21 - 7.76 8.19 - 7.43 8.46 - 7.63 8.04 - 7.16	-20 °C	After 90 days	P < 0.05	(Aboulfazi et al., 2016a)
Fermented soymilk	LAB strain <i>Lactobacillus fermentum</i> BM-325	6.5 - 3.9	N. D	6.2 - 12.6	37 °C	20 h	p < 0.05	(Myagmardorj et al., 2018)
Fermented soymilk	<i>Lactobacillus bulgaricus</i>	7.18 - 4.87	N.D. - 1.57%	N.D. - 8.29				
Fermented soymilk	<i>Lactobacillus plantarum</i>	7.18 - 4.73	N.D. - 1.64%	N.D. - 8.33	37 °C	0 - 12 h	P < 0.05	(Yi et al., 2020)
Fermented soymilk	Mixed <i>Lactobacillus bulgaricus</i> and <i>Lactobacillus plantarum</i>	7.18 - 4.70	N.D. - 1.79%	N.D. - 8.36				
Fermented soymilk	<i>Streptococcus thermophilus</i> MD2 <i>Lactobacillus helveticus</i> V3 <i>Lactobacillus rhamnosus</i> NS6 <i>Lactobacillus bulgaricus</i> NS4 NCDC 09 <i>Lactobacillus acidophilus</i> NCDC 15 <i>Lactobacillus acidophilus</i> NCDC 298 <i>Lactobacillus helveticus</i> NCDC 292	6.4 - 4.43 6.8 - 4.8 6.9 - 4.5 6.7 - 4.43 6.93 - 4.6 6.93 - 5.2 6.8 - 5 6.93 - 4.9	0.17 - 0.34% 0.13 - 0.47% 0.09 - 0.35% 0.1 - 0.33% 0.12 - 0.36% 0.16 - 0.33% 0.09 - 0.33% 0.1 - 0.28%	7.40 - 9.1 6.7 - 9.25 7 - 8.8 7.1 - 8.9 7.2 - 8.8 6.25 - 7.70 7.1 - 8.5 6.4 - 9.1	37 °C	0 - 24 h	P < 0.05	(Hati et al., 2018)

N.D. = not detected.

Table 1. Continued...

Type of products	Probiotic bacteria	PH	Titrateable acidity (TA)	VCC (Log cfu/mL)	Temperature	Time	P- value	References
Fermented soymilk by solid-state	<i>Pediacoccus pentosaceus</i> KTU05-9	5.31 - 5.27	N.D	6.77	35 °C 32 °C 30 °C	0 - 24 h	p ≤ 0.05	(Slapkauskaitė et al., 2019)
		5.57 - 5.34		8.34				
		5.09 - 4.93		8.34				
Fermented soymilk by submerged state	<i>Lactobacillus sakei</i> KTU05-6	6.19 - 5.10	N.D	7.04	N.M.	0 - 28 d	N.M.	(Niamah et al., 2017)
		5.78 - 5.28		9.41				
		5.94 - 4.62		9.3				
Fermented soymilk	<i>Streptococcus thermophilus</i> <i>Lactobacillus acidophilus</i> <i>Bifidobacterium</i> sp.	4.95 - 4.6	0.61 - 0.81%	8.94 - 7.97.3 - 4.1 7.53 - 6.5	N.M.	0 - 28 d	N.M.	(Niamah et al., 2017)
		5.72 - 4.47	0.17 - 0.51%	5.5 - 13.9				
Fermented soymilk	<i>Bifidobacterium longum</i>	5.72 - 5.64	0.17 - 0.21%	5.5 - 10.0	28 ± 2 °C 4 ± 1 °C	0 - 3 w	P < 0.05	(Ebhadaghe et al., 2012)
Fermented soymilk	<i>Bifidobacterium. bifidum</i> <i>Bifidobacterium. animalis</i> subsp. <i>lactis</i>	1.21	17.5 mmol H+ kg	7.08				(Horáčková et al., 2015)
		1.52	22.8 mmol H+ kg	7.08				
Fermented soymilk by yogurt culture	<i>Streptococcus thermophilus</i> <i>Lactobacillus delbrueckii</i> subsp. <i>bulgaricus</i>	2.20	39.0 mmol H+ kg	8.08 6.46	37 °C	16 h		
Fermented soymilk	<i>Streptococcus thermophilus</i> <i>Lactobacillus delbrueckii</i> subsp. <i>bulgaricus</i>	2.27	46.3 mmol H+ kg	8.15 6.73				
Fermented soymilk	<i>Bifidobacterium. bifidum</i> <i>Streptococcus thermophilus</i> <i>Lactobacillus delbrueckii</i> subsp. <i>bulgaricus</i>	2.25	46.8 mmol H+ kg	7.00 8.15 6.38				
Soymilk yogurt inoculated with a concentrations of 2×10 ⁶ cfu/mL	<i>Lactobacillus helveticus</i> isolate H9	4.47 - 4.11 4.41 - 4.07 4.42 - 3.98 4.54 - 3.91	N.D	8.49 - 7.93 7.72 - 7.23 7.74 - 7.59 7.78 - 7.92	4 °C	0 - 28 d	p < 0.05	(Wang et al., 2015)
5 × 10 ⁶ cfu/mL	<i>Lactobacillus acidophilus</i>							
1 × 10 ⁷ cfu/mL								
2 × 10 ⁸ cfu/mL								
Soymilk yogurt	<i>Bifidobacterium lactis</i>	N.D	1.31 - 1.32%	5.4 - 7.1 5.1 - 6.3	10 °C	0 - 1 w	p < 0.05	(Joel et al., 2019)
Fermented white rose flavored with soy yoghurt	Mixed <i>Lactobacillus rhamnosus</i> K4E (KX950834) and <i>Lactobacillus helveticus</i> K14 (KU644578)		0.33 - 0.51%	6.81 - 8.69	6 - 8 °C	1 - 10 d	p < 0.05	(Mishra et al., 2019)
		5.65 - 4.20						

N.D. = not detected.

Table 1. Continued...

Type of products	Probiotic bacteria	PH	Titratable acidity (TA)	VCC (Log cfu/mL)	Temperature	Time	P- value	References
Yogurt 75% cow's milk + 25% soymilk		4.49	0.67%		43 °C	Until pH reached a value of 4.6		
Yogurt 50% cow's milk + 50% soymilk		4.47	0.58%	N. D				(Šertović et al., 2020)
Yogurt 25% cow's milk + 75% soymilk		4.57	0.53%				P < 0.05	
Yogurt 100% soymilk		4.59	0.47%					
	<i>Lactobacillus acidophilus</i> (La5)	4.62 - 4.3	0.81 - 0.89%	7.64 - 6.53				
		4.7 - 4.3	0.55 - 0.82%	8.41 - 7.11	4 °C	0 - 21 d		
		4.65 - 4.39	0.41 - 0.7%	8.03 - N.D				
		4.63 - 4.51	0.43 - 0.54%	7.97 - 8.49				
Soy milk ice cream	<i>Lactobacillus acidophilus</i> (La-05) <i>Bifidobacterium bifidum</i> (Bb-12)	7.14 7.15	0.072% 0.072%	7.857 - 7.847 7.858 - 7.767	-20 °C	after 30 days	P < 0.05	(Aboulfazli et al., 2014)
Fermented ice cream 100% soy milk	<i>Lactobacillus acidophilus</i> (La-05)	5.51 5.50 5.49 5.50	0.27% 0.27% 0.27% 0.36%	8.40 8.33 8.04 8.13		until pH reduced to 5.5		
Fermented ice cream 75% soymilk								
25% cow milk Fermented ice cream 50% soy milk 50% cow milk Fermented ice cream 25% soymilk 75% cow milk	<i>Bifidobacterium bifidum</i> (Bb-12)	5.51 5.50 5.49 5.50	0.27% 0.27% 0.27% 0.27%	8.76 8.32 8.19 8.05	42 °C		P < 0.05	(Aboulfazli et al., 2016b)
Fermented ice cream with 100% soy milk Fermented ice cream with 75% Soy 25% cow milk Fermented ice cream with 50% Soy 50% cow milk Fermented ice cream with 25% Soy 75% cow milk	<i>Lactobacillus acidophilus</i> (La-05) <i>Bifidobacterium bifidum</i> (Bb-12)	-	-	70% 68% 66% 63%				
Fermented soymilk ice cream	<i>Lactobacillus casei ssp. casei</i> CRL-431	5.81	N. D	6.49 - 6	-24C	180 d	p < 0.05	(Aboulfazli et al., 2015) (Homayouni & Norouzi, 2016)

N. D. = not detected.

products are likely to be related to an increase in TA and a drop in pH. This is attributed to the production of acids (mainly lactic acid) and organic acids during fermentation (Kumari et al., 2018; Šertović et al., 2020). In addition, post-acidification phenomena can be illustrated according to each microorganism's metabolic behavior (Mishra & Mishra, 2013).

4.2 Coconut milk

The development of new non-dairy products from fermented coconut milk by probiotic bacteria was well studied (Yuliana & Rangga, 2010; Amirah et al., 2020). Several studies have indicated

that the fermentation process and time, temperature, and type of probiotic could be the main factors that affected the viability of probiotics in coconut milk during fermentation and storage (Mauro & Garcia, 2019; Amirah et al., 2020). The incorporation of probiotics in coconut milk as a food matrix and their growth and viability during fermentation and storage is shown in Table 2. Aboufrazli et al. (2015, 2016a) demonstrated that coconut milk ice cream can be a good carrier for *Bifidobacterium bifidum* (Bb-12) and *L. acidophilus* (La-05) during storage at -20 °C for up to 90 days of storage with viability ranged between 7-8 log cfu/g. However, the viable cell counts of *B. bifidum* in coconut

Table 2. Viable cell counts (VCC) of probiotics in coconut milk during fermentation and refrigerated storage.

Type of products	Probiotic bacteria	PH	Titrateable acidity (TA)	VCC (Log cfu/mL)	Temperature	Time	P- value	References
Coconut milk yogurt (CY) with raisin puree	Mixed culture	4.33		6.04				
CY 0%	<i>Lactobacillus acidophilus</i> <i>Lactobacillus salivarius</i> <i>Bifidobacterium bifidum</i> and <i>Streptococcus thermophiles</i>		N. D	6.11				
CY17%		4.24		6.23	4 °C	15 h	P < 0.05	(Amirah et al., 2020)
CY 23%				6.38				
CY 29%		4.19						
		4.20						
Fermented coconut milk drink	<i>Lactobacillus acidophilus</i>	6.1 - 4	0.1 - 0.6%	4.32 - 9.89	37 °C	0 - 20 h	P < 0.05	(Yuliana and Rangga, 2010)
		3.79 - 3.58	0.524 - 0.742%	8.347 - 10.201	5 °C.	0 - 16 d		
Coconut milk	<i>Lactobacillus casei subsp. rhamnosus</i>	6.12 - 5.66		11.72 - 13.26	37 °C	6 h	p ≤ 0.05	(Szparaga et al., 2019)
		5.61 - 4.81	N. D	13.26 - 9.41	4 °C	1 - 21 d	p < 0.05	
Cow milk +10% - 50% coconut milk		4.1 - 3.8	0.9 - 1%	9.07 - 9.30	30 °C	14 h		
100% Coconut milk	<i>Lactococcus lactis</i> MTCC 3041	4.6	1.1%	9.20			p < 0.05	
Coconut milk with cow milk supplemented	<i>Lactobacillus lactis</i> MTCC 3041 and <i>Lactobacillus plantarum</i> MTCC 5422	6.2 - 4.8	0.9 - 1.2%	10 - 9.11				(Sridhar et al., 2015)
Dahi (curd)	<i>Lactobacillus lactis</i> MTCC 3041 and <i>Leuconostoc sp.</i> MTCC 10508	6.3 - 4.6	0.5 - 1.3%	10 - 9.61	37 °C	0 - 16 h	p < 0.05	
		6.36 - 4.28	0.14 - 0.33 g/100 mL	6.21 - 8.2	37 °C	0 - 48 h	p < 0.05	
Coconut milk beverage	<i>Lactobacillus reuteri</i> LR 92	6.45 - 3.32	0.15 - 0.32 g/100 mL	6.24 - 9.3	34 °C			(Mauro & Garcia, 2019)
	<i>Lactobacillus reuteri</i> DSM 17938	4.59 - 4.33	0.13 - 0.16 g/100 mL	8.04 - 7.55	4 °C	0 - 30 d		
		4.53 - 3.60	0.15 - 0.46 g/100 mL	8.64 - 8.57				
		5.50	0.27%	8.07 - 7.3				
Frozen fermented dessert	<i>Lactobacillus acidophilus</i> (La-05)	5.50	0.27%	7.61 - 6.74				
		5.51	0.27%	8.02 - 6.32	-20 °C	after 90 days	P < 0.05	(Aboufrazli et al., 2016a)
100% coconut milk		5.50	0.27%	8.42 - 6.87				
75% Soymilk + 25% Coconut milk								
50% Soymilk + 50% Coconut milk								
25% Soymilk + 75% Coconut milk		5.50	0.27%	8.42 - 7.91				

N.D. = not detected.

Table 2. Continued...

Type of products	Probiotic bacteria	PH	Titrateable acidity (TA)	VCC (Log cfu/mL)	Temperature	Time	P- value	References
Fermented ice cream with 100% coconut milk	<i>Bifidobacterium bifidum</i> (Bb-12)	5.52	0.27%	8.51 - 7.82	42 °C	until pH reduced to 5.5	p < 0.05	(Aboulfazli et al., 2016b)
		5.50	0.27%	8.56 - 7.94				
	5.51	0.27%	8.59 - 8.04					
	5.50	0.27%	8.30					
	5.51	0.27%	8.18					
Fermented ice cream with 75% soy milk and 25% coconut milk	<i>Lactobacillus acidophilus</i> (La-05)	5.50	0.27%	8.76				
		5.51	0.27%	8.18				
Fermented ice cream with 50% soymilk and 50% coconut milk	<i>Bifidobacterium bifidum</i> (Bb-12)	5.5	0.27%	7.73	-20 °C	after 90 days	p < 0.05	(Aboulfazli et al., 2015)
		5.50	0.27%	8.70				
		5.52	0.27%	8.57				
		5.50	0.27%	8.57				
		5.51	0.27%	8.59				
Fermented ice cream with 100% coconut milk	<i>Lactobacillus acidophilus</i> (La-05)	N. D	N. D	64%				
				69%				
Fermented ice cream with 75% soy milk and 25% coconut milk	<i>Lactobacillus acidophilus</i> (La-05)			67%				
				64%				
Fermented ice cream with 50% soymilk and 50% coconut milk	<i>Bifidobacterium bifidum</i> (Bb-12)	N. D	N. D	86%				
				89%				
				82%				
				81%				
				81%				
Coconut milk ice cream	<i>Lactobacillus acidophilus</i> (La - 05)	6.71	0.117%	7.121 - 6.870				
		6.72	0.126%	7.745 - 7.371				
	<i>Bifidobacterium bifidum</i> (Bb-12)				-20 °C	after 30 days	p < 0.05	(Aboulfazli et al., 2014)

N.D. = not detected.

milk ice cream were significantly higher (86%; $p < 0.05$) than *L. acidophilus* (64%) after 3 months of storage at -20 °C. This indicated that the viability of probiotics in coconut milk ice cream is strongly influenced by the strain used. Yuliana & Rangga (2010) have studied the coconut milk fermented with *L. bulgaricus*, *S. thermophilus*, and *L. acidophilus*. The authors found that the growth of *L. acidophilus* in coconut milk was increased ($p < 0.05$) from 4.32 to 9.89 log cfu/mL after 20 hours of fermentation at 37 °C. In addition, the viability of *L. acidophilus* was remained constant during two weeks of refrigerated storage at 5 °C. Similarly, *L. rhamnosus* and *L. reuteri* showed a significant growth and survival rate in fermented coconut milk during fermentation and storage (Table 2). Furthermore, the growth and viability of probiotics strains in fermented coconut milk might be enhanced with the food additives such as raisin puree and fermented milk "Dahi" (Sridhar et al., 2015; Amirah et al., 2020).

4.3 Other plant-based milk

Several researchers have studied the growth and viability of probiotics in fermented plant-based milk products during fermentation and storage (Table 3). Previous studies have confirmed that almond- and cashew- milk were an excellent carrier for probiotics (Bernat et al., 2015; Wansutha et al., 2018; Bruno et al., 2019). Kabier et al., (2014) have investigated the growth of *B. pseudoatenuatum* G4 in peanut milk during fermentation at 37 °C. They found that *B. pseudoatenuatum* G4 increased from 4.74 to 7.12 log cfu/mL after 24 hours of fermentation. In addition, fructooligosaccharides supplementation enhanced the growth of *B. pseudoatenuatum* G4 in peanut milk during fermentation. On the other hand, *B. longum* BB536 showed a significant reduction (~2 log cfu/mL; $p < 0.05$) in fermented peanut milk after a week of refrigerated storage at 4 °C (Table 3). Fermented

Table 3. Viable cell counts (VCC) of probiotics in other plant-based milk during fermentation and refrigerated storage.

Type of products	Probiotic bacteria	PH	Titrateable acidity (TA)	VCC (Log cfu/ml)	Temperature	Time	P- value	References
Fermented almond milk	Lactic acid bacteria strain (F3)	7 - 5.6	N. D	6.2 - 8.97				
	Lactic acid bacteria strain (M47)	7 - 4.7		6 - 9.12	37 °C	0 - 24 h	N. D	(Wansutha et al., 2018)
	Lactic acid bacteria strain (A62)	7 - 4.7		6.1 - 8.97				
Almond milk yogurt	<i>Lactobacillus reuteri</i>	4.657 - 4.650	N. D	7.9 - 7.3	4 °C	0 - 28 d	P < 0.0	(Bernat et al., 2015)
	<i>Streptococcus thermophilus</i>			7.9 - 6.6				
Rice milk in glass bottles	<i>Lactobacillus casei</i>			9.66 - 9.24	4 °C	0 - 21 d	N. D.	(Padma et al., 2019)
	<i>Bifidobacterium longum</i>	↑ 4	↓ 1	9.75 - 9.12				
	<i>Lactobacillus bulgaricus</i>			8.77 - 8.12				
	<i>Streptococcus thermophilus</i>			7.71 - 4.23 9.77 - 7.3 9.66 - 8.01				
Rice milk in HDPE bottles	<i>Lactobacillus acidophilus</i>			9.75 - 7.54				
				8.77 - 7.01				
				7.71 - 8.45				
				9.77 - 6.5 9.66 - 6.78				
Rice milk in LDPE bottles				9.75 - 6.5				
				8.77 - 6.53				
				7.71 - 9.02				
				9.77 - 6.3				
Peanut milk	<i>Bifidobacterium pseudoatenulatum</i> G4	6.51 - 4.97	0 - 5.275 mmol/ml	4.74 - 7.12	37 °C	0 - 24 h	-	(Kabier et al., 2014)
Peanut milk+ fructooligosaccharides		6.53 - 4.80	0 - 7.765 mmol/ml	4.97 - 8.31				
Peanut milk + fructooligosaccharides + yeast extract		6.44 - 4.64	0 - 6.377 mmol/ml	4.66 - 8.04				
Peanut milk 85% Peanut milk and 15% millet thin porridge.	<i>Bifidobacterium longum</i> BB536	6.24 - 5.83	0.25 - 0.29%	8.83 - 6.75				(Ibraheem et al., 2015)
		6.20 - 5.89	0.26 - 0.28%	7.94 - 6.51				
	70% Peanut milk and 15% millet thin porridge.	6.17 - 5.98	0.24 - 0.28%	7.60 - 5.90	4 °C	0 - 1 week	p < 0.05	
55% Peanut milk and 15% millet thin porridge.		6.11 - 5.87	0.24 - 0.36%	7.63 - 4.79				
Ice creams made using lupine milk	<i>Lactobacillus acidophilus</i>	7.11	0.72%	8.87 - 7.25	-20 °C	0 - 30 d	P < 0.01	(Elsamani, 2016)
	<i>Bifidobacterium bifidum</i>	7.15	0.70%	8.59 - 7.27				
Ice creams made using peanut milk	<i>Lactobacillus acidophilus</i>	6.71	0.12%	9.58 - 7.53				
	<i>Bifidobacterium bifidum</i>	6.74	0.11%	9.87 - 7.65				
Hemp milk	<i>Lactobacillus casei</i> subsp. <i>rhamnosus</i>	6.79 - 6.39	N. D	8.41 - 10.92	37 °C	6 h	p ≤ 0.05	(Szparaga et al., 2019)
		6.47 - 5.78		10.92 - 7.35	4 °C	1 - 21 d	p < 0.05	
	<i>Bifidobacterium animalis</i> BB-12			8.72 - 8.49				
Cashew nut milk	<i>Lactobacillus acidophilus</i> (Howaru® Dophilus)	6.45 - 5.65	N. D	8.17 - 8.89	4 °C	0 - 30 d	P < 0.05	(Bruno et al., 2019)
	<i>Lactobacillus plantarum</i> Lyofast SP-1			8.04 - 8.38				

N.D. = not detected.

rice milk prepared from broken rice was found to be suitable substrates to support high cell viability of probiotic strains such as *L. casei*, *B. longum*, *L. bulgaricus*, *S. thermophilus*, and *L. acidophilus* during 21 days of refrigerated storage (Padma et al.,

2019; Table 3). In addition, glass bottles were the best packaging material for fermented rice milk to maintained high viable cells count of probiotics (Padma et al., 2019). A recent study revealed that the viability of *L. rhamnosus* in hemp milk was significantly

($p < 0.05$) increased from 8 to 11 log CFU/mL during 6 hours of fermentation. However, this value was reduced to 7.35 log CFU/mL after 21 days of storage at 4 °C (Szparaga et al., 2019).

5 Conclusion

In the last few years, there is an increase in the need to develop dairy substitutes for the sake of physical health. Based on evidence-based reviews of research findings on the fermentation of plant-based milk alternatives, it can be concluded that plant-based milk could be a good alternative as a probiotics carrier. Therefore, further studies are required for plant-based dairy products to improve the fermentation process, nutritional values, growth, and viability of probiotics, extend the shelf life of the final product, and improve the functional properties.

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