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*Review*

# Probiotic Delivery through Fermentation: Dairy vs. Non-Dairy Beverages

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**Abstract:** Probiotics are live microorganisms that, when administered in adequate amounts, confer a health benefit on the host, mainly through the process of replacing or including beneficial bacteria in the gastrointestinal tract. Fermented dairy foods such as yogurt, fermented milk and cheese are the major vehicle in delivering probiotics, and probiotic delivery have been traditionally associated with these fermented dairy foods. Additionally, many other non-dairy probiotic products and non-food form such as capsules, pills and tablets are also available and some of these non-food forms are highly popular among the consumers. Certain non-dairy probiotic foods, especially beverages that are non-fermented products, can also play an important role in probiotic delivery. There is an increasing demand for non-dairy probiotic foods (both fermented and non-fermented) including fruit and vegetable juices, soy and certain cereal products due to vegetarianism, lactose intolerance and dairy allergies, as well as interest in low cholesterol foods. In this context, this review mainly focus on the different types of probiotic food products including beverages with special reference to their viability followed by a brief account on the applicability of using fermented and non-fermented beverage products in probiotic delivery.

**Keywords:** probiotics; fermentation; dairy; non-dairy; food matrices; fruit juice; vegetable juice; beverages

## 1. Probiotics: A Brief Overview

In recent years, probiotic foods have received a considerable attention among health-conscious consumers. According to the Food and Agriculture Organization of the United Nations and the World Health Organization [1], probiotics are defined as live microbial cultures of a single strain or mixture of different strains that beneficially affect the host animal, either directly or indirectly, by improving its intestinal microbial balance. Utilization of beneficial microorganisms in health promotion is not new, and in fact they have been consumed by humans, especially in the form of fermented dairy foods, for many years [2]. In the early 1990s, Noble Laureate, Elie Metchnikoff (1845–1916) observed exceptionally long healthy living among Bulgarians who regularly consumed soured/fermented dairy products, and then first documented the modern concept of probiotics in his book "*The Prolongation of Life*" [3]. Since then the use of probiotics in developing functional foods has gained a wide popularity in the world mainly due to the interest in gaining health benefits through consumption of probiotic fortified food products. The most common genera that have been used and possess probiotic characteristics are the lactic acid bacteria *Bifidobacterium* and *Lactobacillus*.

These genera are mostly given the generally-recognized-as-safe (GRAS) status, which indicates no or less health risks to the host upon consumption [2,4,5]. Few other microorganisms mainly bacteria and some yeast have also been utilized as probiotics (Table 1). However, there are some concerns regarding the safety of some probiotic genera such as *Enterococcus*, since they can be pathogenic, causing illness in the host.

**Table 1.** Microorganisms used as probiotic cultures.

| ***Lactobacillus* spp.** | ***Bifidobacterium* spp.** | **Other spp.** |
|---|---|---|
| *L. acidophilus*<br>*L. casei*<br>*L. crispatus*<br>*L. delbrueckii* subsp. *bulgaricus* [a]<br>*L. fermentum*<br>*L. gasseri*<br>*L. johnsonii*<br>*L. paracasei*<br>*L. plantarum*<br>*L. reuteri*<br>*L. rhamnosus*<br>*L. helveticus*<br>*L. lactis*<br>*L. sporogenes* | *B. bifidum*<br>*B. breve*<br>*B. infantis*<br>*B. longum*<br>*B. lactis*<br>*B. animalis*<br>*B. adolescentis*<br>*B. essensis*<br>*B. laterosporus* | *Escherichia coli* Nissle<br>*Saccharomyces boulardii*<br>*Saccharomyces cerevisiae*<br>*Kluyveromyces lactis*<br>*Streptococcus thermophilus* [a]<br>*S. cremoris*<br>*S. diacetylactis*<br>*S. intermedius*<br>*S. salivarius*<br>*Enterococcus francium* [b]<br>*Propionibacterium freudenreichii*<br>*P. freudenreichii* subsp. *shermanii*<br>*P. jensenii*<br>*Pediococcus*<br>*Leuconostoc lactis* subsp. *cremoris*<br>*L. lactis* subsp. *lactis Bacillus cereus*<br>*Clostridium butyricum* |

[a] There is still debate about the probiotic activity due to poor survival during gastrointestinal transit; [b] Safety concerns remain because of potential pathogenicity and vancomycin resistance. Adopted and modified from [6–10].

The human gastrointestinal tract contains trillions of microorganisms, consisting of up to 1000 or more different bacterial species, collectively known as the gut microbiota. The gut microbiota plays an important role in host health, influencing the maturation of the immune system and regulating energy metabolism [11]. In general, it is accepted that intake of probiotics contributes to the enhancement and maintenance of well-balanced intestinal microbiota. Many evidences support the use of these probiotics in prevention and treating diseases and health disorders such as high blood pressure & serum cholesterol, lactose intolerance [12] and many gastrointestinal disorders (irritable bowel syndrome, Crohn's disease, peptic ulcers, antibiotic associated diarrhea, etc.) [13–16]. Probiotics also possess anti-carcinogenic effects [17–19] and enhance the immune system [20,21]. Some examples of probiotic potential for therapeutic applications have been listed in Table 2.

**Table 2.** Examples of beneficial effects of therapeutic probiotic application in humans.

| Disorder | Probiotic Strain | Mode of Delivery | References |
|---|---|---|---|
| Antibiotic-associated diarrhoea in adults | Mixture of *L. casei*<br>*L. bulgaricus*<br>*S. thermophilus* | Drinking yogurt | [14] |
| Antibiotic-associated diarrhoea in children | *Lactobacillus reuteri* | Drops | [22] |
| Traveler's diarrhoea | Single strain of *Lactobacillus* GG | Powdered form dissolved in cold water | [23] |
| Irritable bowel syndrome symptoms | Mixture of *B. longum*, *B. infantis*, *B. breve*, *L. acidophilus*, *L. casei*, *L. delbrueckii*, *L. plantarum*, *S. salivarius* | Lyophilized powdered form | [13] |
| | Mixture of *L. rhamnosus*, *B. breve* & *P. freudenreichii* subsp. *shermanii* | Capsules | [24] |

**Table 2.** *Cont.*

| Disorder | Probiotic Strain | Mode of Delivery | References |
|---|---|---|---|
| | Mixture of *B. animalis*, *L. bulgaricus* & *S. thermophilus* | Fermented milk | [25] |
| | Mixture of *B. longum*, *B. infantis*, *B. breve*, *L. acidophilus*, *L. casei*, *L. delbrueckii*, *L. plantarum*, *S. salivarius* | Lyophilized powdered form | [26] |
| | Single strain of *L. plantarum* | Rose-hip drink with oat flour | [27] |
| | Single strain of *B. animalis* | Fermented semi skimmed-milk | [28] |
| Crohn's disease<br>Ulcerative colitis<br>Pouchitis | Mixture of *B. longum*, *B. infantis*, *B. breve*, *L. acidophilus*, *L. casei*, *L. bulgaricus*, *L. plantarum*, *S. thermophilus* | Lyophilized form | [29] |
| | Single strain of *E. coli* Nissle | Capsules | [30] |
| | Mixture of *L. acidophilus* La-5, *Bifidobacterium* Bb 12 | Fermented milk | [31] |
| Bacterial vaginosis | Mixture of *L. rhamnosus* & *L. reuteri* | Gelatin capsules | [32] |
| IgE associated eczema<br>Atopic dermatitis<br>Atopic dermatitis in infants | Single strain of *L. reuteri* | Freeze dried form in coconut or peanut oil droplets | [33] |
| | Single strain of *L. rhamnosus* | Skim milk based freeze-dried form | [34] |
| | Mixture of *L. rhamnosus* & *L. reuteri* | Lyophilized powdered form | [35] |
| | Mixture of *L. rhamnosus*, *B. animalis* subsp. *lactis Bb-12 (Bb-12) & L. acidophilus La-5* | Milk (maternal supplementation) | [36] |

Adapted and modified from [37].

For probiotic bacteria in foods to be beneficial in the host, they should be able to survive gastric transit and reach the small intestine in sufficient numbers to be effective. Hence, in order to provide health benefits to the host, probiotics should maintain minimum therapeutic level/minimum viability level ($10^6$–$10^7$ cfu/mL or g of carrier food product) at the time of consumption, possess the ability to tolerate harsh gastric and intestinal conditions (including acid, bile and enzymes) and be able to attach to the gut epithelium [38,39]. A potentially successful probiotic strain is expected to have several more desirable properties (Table 3) and these characteristics may influence its potential for the commercial applications.

**Table 3.** Key and desirable criteria for the selection of probiotics in food and nutraceutical applications.

| Criteria | Property/Characteristic | Target and Methods to Be Assessed |
|---|---|---|
| Safety | Origin<br>Pathogenicity and infectivity<br>Virulence factors-toxicity, metabolic activity and intrinsic properties, i.e., antibiotic resistance | Source or origin should be assessed: be isolated from the same species as its intended host is desirable due to higher efficacy in the same species. Probiotics of human origin may be desirable if they are intended for human use.<br>Pre-market clearance and post-market surveillance |
| Technological acceptability | High viability retention during manufacturing and storage of carrier foods<br>Acceptable organoleptic characteristics<br>Ability to produce at large-scale<br>Phage resistance | In vitro studies and food product development<br>Sensory testing of model and final products and consumer studies on product formulations |
| Functionality | Tolerance to gastric acid and juices including acidic conditions and enzymes<br>Bile tolerance<br>Adhesion to mucosal surface and colonization<br>Validated and documented health effects | Model systems for gastric and bile effects (e.g., in vitro, animal and human studies)<br>*In vitro* adhesion models (e.g., intestinal segments, mucus, cell culture), animal and human studies<br>Health effects confirmed by clinical studies |
| Desirable physiological criteria | Immunomodulation<br>Antagonistic activity towards gastrointestinal pathogens<br>Antimutagenic and anticarcinogenic properties | In vitro/In vivo animal and human studies.<br>Adhesion and competitive exclusion of pathogens in in vitro and in vivo model systems |

Adapted and modified from [37,38,40–43].

Probiotics can exert their effects by one or more actions, e.g., creation of a restrictive physiological environment for potentially pathogenic microorganisms. These effects are achieved by lowering the pH through production of organic acids, such as lactate and short chain volatile fatty acids, due to break-down of complex carbohydrates [44] or elaboration of antibiotic-like substances such as bacteriocin-like compounds [45]. Certain probiotic bacterial species, such as lactic acid bacteria, can adhere to the intestinal epithelium and thereby prevent invasion by pathogenic bacteria such as *Escherichia coli*, *Salmonella* and *Clostridium* spp. in the gut epithelium and this phenomenon is known as competitive exclusion. Feeding probiotics may also help to modulate the cellular and humoral immune system thereby enhancing the host's resistance to enteric pathogens [38,46].

## 2. Dairy vs. Non-Dairy Food Matrices

Dairy products such as yogurts, fermented sour milk and cheese remain at the forefront of probiotic food development at present. Although fermented dairy foods can be considered as one of the most common as well as the traditional modes of delivering of probiotics to humans, at present, many non-dairy as well as non-traditional and convenient probiotic products, such as capsules, have been developed and commercialized in many countries [47]. Soy products, cereal based products, fruit and vegetable juices, and fermented meat and fish products can be considered as main non-dairy probiotic foods available in the market at present. There are many different types and brands of non-dairy probiotic foods as well. The diversity of probiotic food products is summarized in Table 4. Many studies have clearly indicated that the type of carrier foods could affect not only the viability of probiotics during processing and storage, but also on their functional properties, such as susceptibility to adverse conditions in the gut (acidity, bile and various enzymes), capacity to adhere to gut epithelium and immunomodulation [39,48,49]. The incorporation of probiotics into dairy foods may aid in tolerating harsh gastro-intestinal condition better than that of non-dairy carrier foods, as the buffering action of milk as well as milk fat, might protect probiotics in such conditions by reducing their direct exposure to harsh conditions [2]. Dairy foods rich in milk fat, such as ice cream, were found to be more effective in enhancing the survivability and bile acid tolerance of probiotics [50]. However, the physical structure of non-dairy probiotic carrier foods such as vegetables (for example artichokes and olives) might provide protective environment for probiotics and reduce their exposure to harsh gastrointestinal conditions as well [51]. Sausage matrix and microstructure have also shown a potential in retaining the viability of probiotics through gastrointestinal transit [52,53].

**Table 4.** The diversity of probiotic food products and the viability of each probiotic in different products at the end of appropriate storage conditions (either freeze or cold storage).

| Product Type | Product | Probiotic Strain | Viability at the End of Storage | Total Storage Time | References |
|---|---|---|---|---|---|
| Dairy based | Fermented cow's milks | *L. acidophilus*<br>*L. rhamnosus* | $10^7$ cfu/g | 7 days | [54] |
| | Fermented goat's milk | *L. acidophilus*<br>*Bifidobacterium* BB-12 | <$10^6$ cfu/g<br>$10^6$–$10^7$ cfu/g | 21 days | [55] |
| | Fermented dairy drink from goat's milk | *L. acidophilus*<br>*B. animalsi* ssp. *lactis* | $10^7$ cfu/mL | 21 days | [56] |
| | Fermented skim milk (cow's milk) | *L. acidophilus*<br>*B. animalsi* ssp. *lactis* | $10^6$ cfu/mL | 21 days | [57] |
| | Cow's milk yogurt | *L. acidophilus*<br>*B. longum*<br>*B. psedolongum*<br>*B. infantis*<br>*B. bifidum* | >$10^6$ cfu/g | 42 days | [58] |
| | | *P. jensenii* | $10^5$ cfu/g | 15 days | [59] |
| | Cow's milk fruit yogurt | *L. acidophilus*<br>*B. animalsi* ssp. *lactis* | $10^6$–$10^7$ cfu/g | 35 days | [60] |

**Table 4.** *Cont.*

| Product Type | Product | Probiotic Strain | Viability at the End of Storage | Total Storage Time | References |
| --- | --- | --- | --- | --- | --- |
| | Rice incorporated cow's milk yogurt | *B. animalis* subsp. *lactis BB-12* | $10^8$ cfu/g | 21 days | [61] |
| | Low fat set yogurts (cow's milk) | *B. infantis*, *B. longum* subsp. *infantis* | $10^7$ cfu/g | 28 days | [62] |
| | Goat's milk yogurt | *L. acidophilus*<br>*B. bifidum*<br>*L. paracasei* subsp. *casei* | $10^7$ cfu/g | 14 days | [63,64] |
| | Ewe's milk yogurt | *L. acidophilus*<br>*B. bifidum*<br>*L. casei* | $10^7$ cfu/g | 14 days | [65] |
| | Ice cream | *L. johnsonii*<br>*L. acidophilus*<br>*B. lactis* | $10^7$ cfu/g<br>$10^5$–$10^6$ cfu/g | 8 months<br>90 days | [66]<br>[67] |
| | Ice cream (vanilla flavoured) | *L. acidophilus* | $10^6$ cfu/mL | 60 days | [68] |
| | Goat's milk ice cream (chocolate flavoured) | *L. acidophilus*, *B. animalis* subsp. *Lactis*, *Propionibacterium jensenii* | $10^7$–$10^8$ cfu/g | 52 weeks | [50] |
| | Cheddar cheese | *L. paracasei*<br>*Lactococcus lactis* subsp. *cremoris*,<br>*L. lactis* subsp. *lactis*,<br>*Lactobacillus helvetics*,<br>*S. thermophiles*,<br>*Lactobacillus rhamnosus* | $10^7$ cfu/g<br>$10^8$ cfu/g | 90 days<br>4 weeks | [69]<br>[70] |
| | Fresh Minas cheese | *L. paracasei* | $10^8$ cfu/g | 21 days | [71] |
| | White Turkish cheese | *L. acidophilus* | $10^7$ cfu/g | 90 days | [72] |
| | Semi hard Argentinian cheese | *L. paracasei*<br>*L. acidophilus* | $10^8$ cfu/g | 60 days | [73] |
| | Argentinian Fresco cheese | *B. bifidum*<br>*B. longum*<br>*L. acidophilus*<br>*L. casei* | $10^6$ cfu/g | 60 days | [74] |
| | Requeijao-cheese (Portuguese-whey cheese) | *L. animalis*<br>*L. acidophilus*<br>*L. paracasei*<br>*L. brevis* | $10^7$ cfu/g | 28 days | [75] |
| | Semi hard goat's cheese | *L. acidophilus*<br>*B. lactis* | $10^6$ cfu/g | 70 days | [76] |
| | Crescenza cheese (soft Italian cheese) | *B. bifidum*<br>*B. infantis*<br>*B. longum* | $10^5$ cfu/g | 14 days | [77] |
| Soya based | Soya frozen dessert | *L. acidophilus*<br>*L. paracasei*<br>*B. lactis*<br>*L. rhamnosus*<br>*S. boulardii* | $10^7$ cfu/g<br>~$10^5$ cfu/g | 28 weeks | [78] |
| | Soy milk | *B. breve* | $10^9$ cfu/mL | 20 days | [79] |
| Cereal based | Oat bars | *B. lactis* | $10^9$ cfu/25 g bar | 7–14 days | [80] |
| | Milk based maize/rice pudding | *B. animalis*<br>*L. acidophilus*<br>*L. rhamnosus* | $10^8$–$10^9$ cfu/g | 21 days | [81] |
| | Oat meal gruel mixed with fruit drinks (i.e., rose hip, strawberry) | *L. plantarum* | $10^{10}$ cfu/mL | 30 days | [82] |

**Table 4.** *Cont.*

| Product Type | Product | Probiotic Strain | Viability at the End of Storage | Total Storage Time | References |
| --- | --- | --- | --- | --- | --- |
| Fruit and fruit juice | Blackcurrant | *L. plantarum* | Not reported | | [83] |
| | Dried apple fruits | *L. casei* | $10^6$ cfu/g | | [84] |
| | Apple juice | *L. acidophilus*<br>*L. rhamnosus*<br>*L. salivarius*<br>*L. plantarum*<br>*L. paracasei*<br>*B. longum*<br>*B. lactis* type Bi-04<br>*B. lactis* type Bi-07 | $10^6$ cfu/mL | 6 weeks | [85] |
| | Pineapple juice | *L. casei* | $10^6$ cfu/mL | 42 days | [86] |
| | | *Pediococcus pentosaceus*, *Lactobacillus rhamnosus*, *Pediococcus pentosaceus* | $10^9$ cfu/mL | 4 weeks | [87] |
| | Cashew apple juice powder (spray dried) | *L. casei* NRRL B-442 | ~$10^6$ cfu/g | 35 days | [88] |
| | Orange juice after Spouted bed drying | *L. casei* | ~$10^6$ cfu/g | 5 weeks | [89] |
| | Orange juice powder (spray and freeze dried) | *L. plantarum* 299v | $10^6$ cfu/g | 180 days | [90] |
| | Orange | *Lactobacillus* GG | Not reported | | [91] |
| Vegetable based | Table olives | *L. rhamnosus*<br>*L. paracasei*<br>*B. bifidum*<br>*B. longum* | $10^6$–$10^8$ cfu/g | 90 days | [92] |
| | Carrot blended with orange juice | *L. plantarum* CECT 220 | $10^8$–$10^9$ cfu/mL | 30 days | [93] |
| | Tomato juice | *L. plantarum*<br>*L. acidophilus*<br>*L. casei*<br>*L. delbrueckii* | $10^4$–$10^8$ cfu/g | 30 days | [51] |
| | Beet juice | *L. plantarum*<br>*L. acidophilus*<br>*L. casei*<br>*L. delbrueckii* | $10^6$–$10^8$ cfu/mL | 4 weeks | [94] |
| | Cabbage juice | *L. plantarum*<br>*L. delbrueckii* | $10^7$ cfu/mL<br>$10^5$ cfu/mL | 4 weeks | [95] |
| | Artichokes | *L. plantarum*<br>*L. paracasei* | $10^7$–$10^8$ cfu/g | 90 days | [96] |
| | Fermented Kale juice (*Brassica oleraceae*) | *L. plantarum*<br>*L. casei*<br>*L. acidophilus*<br>*L. brevis* | $10^8$ cfu/mL | 4 weeks | [97] |
| | Fermented vegetable soybean beverage | *L. acidophilus* La-5, *B. animalis* Bb-12 | ~$10^6$ cfu/mL<br>$10^8$ cfu/mL | 28 days | [98] |
| | Vegetable pickle products | *L. casei LA284* | $10^4$–$10^8$cfu/g | 70 days | [99] |
| Meat and fish products | Fermented sausage | *L. plantarum* | - | - | [53] |
| | Dry sausages-beef + pork | *L. rhamnosus* | $10^8$ cfu/g | 28 days | [100] |
| | Fermented fish sausage | *Lactobacillus* spp. | satisfactory | 7 days | [101] |
| Miscellaneous | Encapsulated and spray dried milk powder | *L. acidophilus*, *B. animalis* subsp. *Lactis*, *Propionibacterium jensenii* | $10^5$–$10^7$ cfu/g | 24 weeks | [102] |
| | Chocolate | *L. acidophilus LA3*, *B. animalis* subsp. *lactis* BLC1 | $10^7$–$10^8$ cfu/g | 120 days | [103] |
| | African beverages made from maize and milk | *B. lactis* | $10^7$ cfu/mL | 21 days | [104] |

Adapted and modified from [37,51].

Nevertheless, both dairy and non-dairy products may contain various other ingredients such as prebiotics (ingredients which ferment in the latter part of the gastrointestinal tract and stimulate the growth and activity of beneficial gut microbes) that could interact with probiotics to alter their functional properties [105]. Presence of these substances could be specific to certain carrier foods. For example, naturally, milk does not contain inulin (plant derived polysaccharide with prebiotic properties), yet certain root vegetables/rhizomes, fruits and cereals (artichoke, oat, bananas, garlic, onions, leeks) contain high level of inulin. Nevertheless, there is also a possibility of production of probiotic dairy products by incorporating various prebiotics such as inulin or its breakdown products (fructooligosaccharides and oligofructans). Prebiotic oligosaccharides are essentially obtained by one of three processes: direct extraction of natural oligosaccharides from plants, controlled hydrolysis of natural plant polysaccharides, and enzymatic synthesis, using hydrolases and/or glycosyltransferases [106]. Apart from the direct prebiotic activities, many plant and microbial derived oligosaccharides help to deliver the probiotic organisms to the target sites. The encapsulation of probiotic organism with such compounds prevents the gastrointestinal digestion of the probiotic organism enabling them to be present at large intestine which is the target site of probiotics [107]. Prebiotics from plant sources such as Arrowroot (*Maranta arundinacea*) carbohydrates and Raftilose® (commercially available inulin) can be used to enhance the survivability of *Lactobacillus* sp. and lactic acid bacteria in bio-yoghurt during refrigerated storage [108]. These prebiotic substances may aid probiotics to survive through the gastrointestinal transit and colonize in the large intestinal epithelium. Another study [46] revealed that supplementation of broiler chicken feed with specific prebiotic compounds supported the growth of specific probiotic *Lactobacillus* spp. such as *L. johnsonii* in their ileum and caeca. Therefore, careful selection of probiotics, prebiotics and carrier food matrices (both dairy and non-dairy) when produce probiotic foods is essential in maximizing the functional efficacy of probiotics during manufacturing, storage and upon ingestion. It seems likely that in most cases the carrier food matrices possess synergistic effect on probiotic microorganisms during processing as well as in the gastrointestinal environment [2]. However, questions about such synergistic effects may arise when non-food probiotic carriers such as capsules are used.

Many probiotics with potential health benefits have be isolated from the gastrointestinal tract of healthy humans (human origin). There are some non-human sources as well. Raw cow's milk is considered as an excellent source of probiotic bacteria [109]. Usually heat treatments such as pasteurization and sterilization destroy these beneficial microorganisms in raw milk, however better synergistic effect may be expected through re-introduction of such probiotics into milk when produce dairy products mainly due to their dairy origin. Efficacy of dairy origin probiotics when incorporated into non-dairy foods could be significantly affected and in line with this, more research is needed to discover the effect of functional properties of dairy origin probiotics when incorporate into non-dairy food matrices including beverages. However, the development of probiotic containing dairy products is not always easy and requires the overcoming of certain technological intrinsic requirements related to their processing stages. For example, selection of probiotic strains withstanding freezing is essential in production of probiotic ice cream despite its origin [50].

Although there are many benefits of having probiotics with dairy foods, non-dairy probiotic foods also play a significant role in human health. For instance, there are major drawbacks related to dairy foods such as allergy, lactose intolerance and cholesterol content, hence non-dairy probiotic foods are beneficial for the people having such health disorders. Furthermore, cultural (strict vegans) as well as specific religious believes among certain communities may also limit the consumption of dairy foods. In such situations, non-dairy probiotic carrier foods and convenient mode of deliveries such as tablets could be the only way of providing probiotics. Each probiotic strain is unique in many aspects such as optimum growth conditions and growth medium. Consequently, the selection of probiotics which perform better or equally in non-dairy foods compared with dairy foods may be useful in developing non-dairy probiotic food [110–113]. In general, most non-dairy probiotics are beverages. In this context, it could be argued that non-dairy probiotic carriers including beverages may be equally importance as dairy related carriers in terms of human health and nutrition. Some commercially available non-dairy probiotic beverages are shown in Table 5.

**Table 5.** Commercially available non-dairy probiotic beverages.

| Product Label | Manufacturer | Major Characteristics | Probiotic Strain/s |
|---|---|---|---|
| Golden Circle Healthy Life Probiotic Juice | Golden Circle, Australia | Mixture of apple juice and mango puree or orange, apple, pineapple, passionfruit with banana puree | *L. paracasei* 8700:2 and *L. plantarum* HEAL 9 |
| PERKii Probiotic Water | PERKii, Australia | Fruit juice mixtures such as raspberry and pomegranate, lime and coconut, mango and passionfruit and strawberry and watermelon | *Lactobacillus casai* Lc431 |
| Bravo Friscus | Probi AB, Swdeen | Orange apple and tropical fruit juices | *L. plantarum* HEAL9 and *L. paracasei* 8700:2 |
| ProViva | EMEA Probi AB, Sweden | Fruit juice (orange, strawberry or blackcurrant) | *L. plantarum* 299v |
| Bio-Live Gold & Dark | Bio-Live/Microbz Ltd., UK | Mixtures of fruit juices such as acai berry, cherry, goji, noni, pomegranate, lemon, and various herbs | *Mixture of 13 strains including L. acidophilus, L bulgaricus, L. casei, L. fermentum, L. plantarum, Lactococcus lactis, Bacillus subtilis, B. bifidum, B. Infantis B. longum; Streptococcus thermophilus, Comobcillus and Saccharomyces cerevisiae* |
| Biola | TINE, Norway | Mixture of apple, grapes and passion fruit or orange and tangerine | *L. rhamnosus* GG |
| Malee Probiotics | Malee Enterprise Compny Ltd., Thailand | Fruit juices such as prune, grape and orange | *L. paracasei* |
| GoodBelly® Carrot Ginger Flavor Probiotics Juice Drink. | Goodbelly, USA | Carrot Juice, ginger extract and cane sugar contains 2% or less of gluten-free oat flour, | *L. Plantarum* 299v |
| KEVITA | KEVITA, USA | Various fruit based mixtures such as strawberry and coconut, lime, mint and coconut, mango and coconut, pineapple and coconut | *Bacillus coagulans* GBI-30 6086, *L. rhamnosus*, *L. plantarum*, *L. paracasei* |
| Tropicana probiotics | Tropicana, USA | Fruit juice mixtures such as strawberry and banana, pineapple and mango and peach passion fruit | *B. lactis* |
| Probiotic Naked Juice | Naked® Juice, USA | Mixture of apple, orange, pineapple juices and mango and banana puree with fructooligosacccharides | *Bifidobacterium* |

Adapted and modified from [114].

## 3. Fermented vs. Non-Fermented Beverages

Fermentation technology is one of the oldest food technology applications and fermented products are the result of the metabolic activity of a complex microbiota, consisting of the naturally occurring indigenous microorganisms, and/or selected microorganisms such as bacteria and yeasts which inoculated as starter cultures. Fermentation of food products helps their preservation due to the organic acid production as well as imparting them pleasant sensory properties and additional nutritional values [52]. In terms of probiotic beverage production, fermentation process is not compulsory (Figure 1). There are many types of probiotic fermented milk in the global market produced under various brand names. Major physicochemical properties of probiotic fermented milk products vary basically on the type of probiotic microorganism, type of milk and use of other starter cultures in the product. In addition, fermented probiotic dairy products vary in their textures ranging from liquid drinks such as acidophilus milk and kefir to semi-solid/ropy or firm products such as drinking yogurt and villi [115]. Microorganisms used in starter cultures are of great industrial significance since they play a vital role in flavour and textural development of fermented food products. These starter cultures may not necessarily possess probiotic properties. For example, the term "probiotics" may not be suitable for yogurt starter cultures (*Streptococcus thermophilus* and *Lactobacillus delbrueckii* spp. *bulgaricus*) due to their poor survival in the digestive tract [6]. However, some beneficial health promoting effects of yogurt starter cultures including improved lactose utilization and enhancement of immune system have also been reported [116,117]. In addition to the starter culture microorganisms, various probiotics can be added during the production of fermented food products including beverages to achieve the therapeutic benefits. Having starter cultures in probiotic products may provide benefits as well as some disadvantages. For example, starter cultures may create a suitable environment for probiotic growth during yogurt manufacturing. Yogurt starter culture bacteria, in particular *S. thermophiles*, are also identified as oxygen scavengers and thus may be beneficial in improving the growth and viability of anaerobic probiotics. These starter cultures were previously demonstrated to complete the fermentation of milk within 5–10 h and utilised most of the oxygen in milk [118]. In contrast, variations in the starter cultures and probiotic combinations may also influence the probiotic viability in the final product due to antagonistic or symbiotic relationships [37]. However, other options have been showing interesting, as the addition of enzymes [119] and should be considered for dairy and non-dairy processors to guarantee the viability of the probiotic strain during commercial shelf-life of the products.

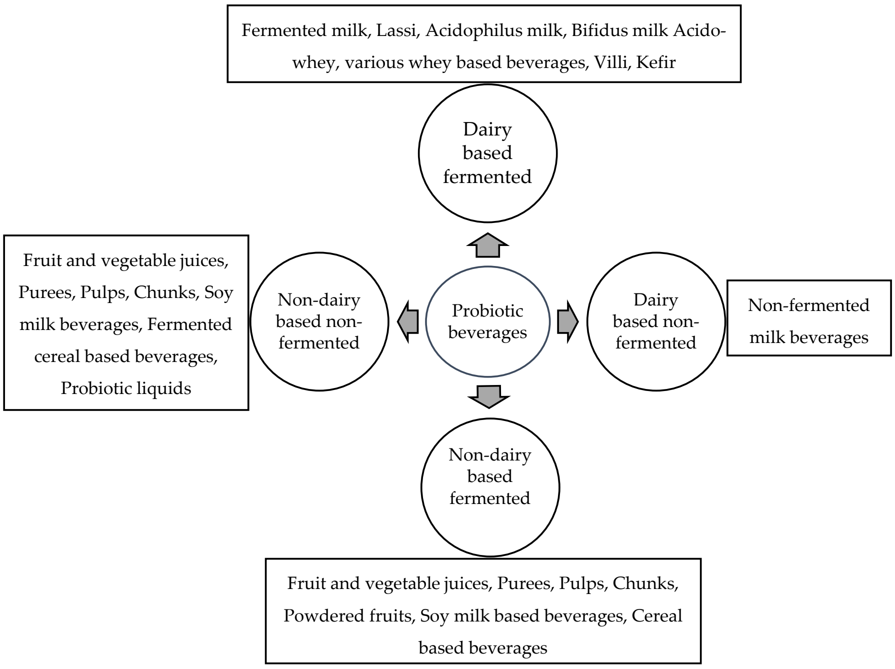


**Figure 1.** Classification and types of probiotic beverages. Adapted and modified from [115,120].

The majority of the probiotic dairy beverages in the present-day market can be categorized into fermented products (Figure 1). Based on the type of microorganisms involved in the fermentation, these dairy beverages can be classified into different categories (Table 6). Many non-dairy probiotic carriers such as fruit and vegetable juices are however produced mainly without fermentation since fermentation may cause undesirable sensory properties in such products [10] due to various factors such as acidity development, changes in viscosity, texture and colour. Production of fermented and non-fermented fruit and vegetable juices can be very similar with only one additional step of fermentation when manufacture fermented juices (Figure 2). In terms of achieving desirable food characteristics, fermentation is a complex process and selecting appropriate conditions such as optimum temperature during fermentation is a critical parameter that must be considered to prevent lethal or sub-lethal damages to the probiotic cells during the processing and subsequent storage. These conditions affect the biomass yield as well [121]. Duration of fermentation (full or partial fermentation) may also affect the quality of the final product. Partial fermentation of dairy foods could results drinking dairy beverages and recently there has been high demand for such products [56]. Further, probiotics in fermented foods may demonstrate better stability in the product as fermentation time can provide them an opportunity to grow and stabilize well [50]. However, despite the variations in the production process and possible disparities in product's physico-chemical, sensory, nutritional and therapeutic properties, modern health conscious consumers have a strong demand for both fermented and non-fermented probiotic food products, especially for probiotic beverages [10,122] and this fact can be an advantage for increase the sales of these beverage products.

**Table 6.** Types of fermentation used to produce different fermented dairy beverages.

| Fermentation Type | Type of Microorganism Involved | Beverage Products |
|---|---|---|
| Lactic Fermentation | Mesophilic type<br>Thermophilic type<br>Therapeutic | Cultured buttermilk<br>Bulgarian buttermilk, Drinking yogurt<br>Acidophilus milk, Yakult |
| Yeast-lactic Fermentation | Yeast and lactic acid bacteria | Kefir, Acidophilus yeast milk |
| Mould-lactic Fermentation | Mould and lactic acid bacteria | Villi |

Adapted and modified from [115,123].

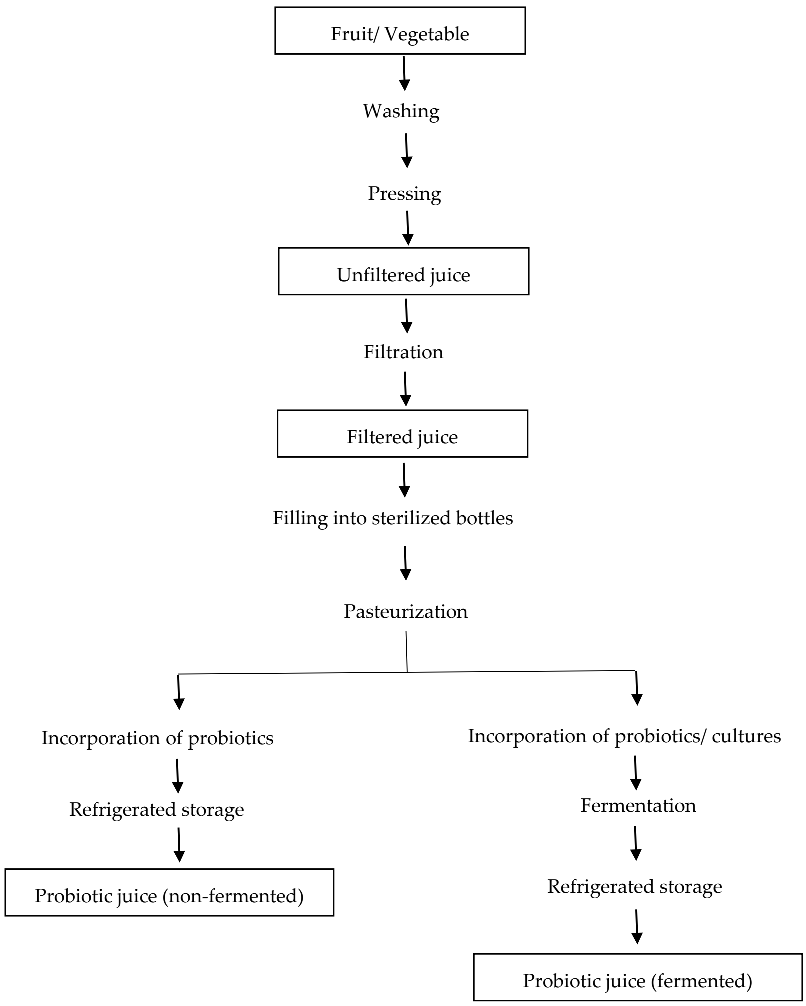


**Figure 2.** Flow chart for the production of fermented and non-fermented fruit/vegetable juices enriched with probiotics.

## 4. Conclusions

Fermented dairy products remain at the forefront of probiotic delivery. However, the variety of non-dairy foods both fermented and non-fermented nature is available and these products also play a significant role in delivering probiotics to humans. Generally, most probiotic beverages, in particular those with fruit and vegetable origin are non-fermented formulations. The efficacy of probiotics when delivered through fermented vs. non-fermented status of a particular carrier food matrix with special reference to beverages has not been studied thoroughly and in order to reap the maximum benefits of probiotics, further research in this aspect is needed. Simultaneously, sensory tests should be applied to evaluate the consumer's acceptance of these beverage products, providing more optimized formulations.

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