Prebiotics and Probiotics - Potential Benefits in Human Nutrition and Health

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Abstract

The growing interest of using probiotic bacteria into foods has lately increased due to its beneficial effects on intestinal microbiota. This fact has motivated researchers and the food industry to develop new functional products, such as probiotics. The probiotic fruit juices can be a good alternative for new foodstuff production mainly for people with galactosemia and lactose intolerance. Among the microencapsulation techniques of probiotics in juices, spray drying and freeze drying are two of the most used with 220 and 228 publications found in Science Direct, 8 and 0 Web Science, and 5 and 0 in SCOPUS, respectively. Several studies have reported the addition of probiotics in different fruit juices to produce functional beverages; however, there are no reports on the use of three or more probiotic microorganisms in mixed beverage. Our research group has been directing studies in this area in order to provide results of scientific interest and to food industry.

Keywords: new product, juice fruit, spray dryer

1. Introduction

Consumers’ growing interest in using food to improve their health and wellness has motivated researchers and the food industry to develop new functional products, such as probiotics [1]. Interest in healthy, nutritious, high-end food products has increased worldwide as probiotic-supplemented beverages beneficially affect one or more body functions and have the potential to promote health. Consumers are more aware of the relationship between good nutrition and health, so there is a growing demand for foods that, in addition to nourishing, provide health benefits.

Traditionally, probiotics are incorporated into dairy products. However, nondairy food matrices have been studied as potential carriers for these microorganisms because of the increasing number of individuals who are affected by lactose intolerance, milk protein allergy, galactosemia, and hypercholesterolemia. Seventy-five percent of the world’s population is suffering from lactose intolerance [2, 3].

Fruit juices, desserts, and cereal-based products featuring probiotics may be other suitable media for delivering probiotics. Most of the probiotic food products is categorized as functional foods. These products include fruit drinks. According to Corbo [4], functional beverages are nonalcoholic beverages containing...
nontraditional ingredients that offer health benefits. Within this context, fruit juices are indeed promising probiotic carriers due to their essential nutrient content along with their appeal to a niche of consumers who already care about healthier habits [5]. The great advantage of fruit juices as a probiotic food is that it is regularly consumed by high portion of the population, which would allow continuity of the beneficial effect from the probiotic microorganisms carried by this food.

However, the viability of the microorganisms is defined during processing, being a necessary application of methods that can maintain or improve their viability and functionality. With this, new technologies have been proposed, among them, the microencapsulation stands out as a promising technique. The microencapsulation can be defined as a process in which small solid particles, liquid droplets, or gases are evolved by a coating layer, or incorporated into a homogeneous or heterogeneous matrix, yielding small capsules with useful properties [6–9].

The microencapsulation of food ingredients in coating materials can be achieved by various methods. Some of them are spray drying, extrusion, freeze drying, fluidized bed, coacervation, and cocrystallization. Among these methods, atomization is the most used in the manufacture of foodstuffs [10, 11].

The spray drying consists of transforming a product in fluid state into solid state in the form of powder, a continuous operation, through a relatively short time [12–14], being also the most used method to encapsulate probiotic bacteria [15].

2. Nondairy food matrices as a carrier of microencapsulated probiotic microorganisms microencapsulated

The word probiotic comes from the Greek word “προ-βίος” that means “for life”; thus, probiotics are live microorganisms (mainly bacteria but also yeasts) that confer a beneficial effect on the host if administered in proper amounts [16].

The most common probiotic microorganisms used commercially in food are bacteria from the genera Lactobacillus and Bifidobacterium [17]. Lactobacillus species used as probiotics in food include L. acidophilus, L. crispatus, L. amylovorus, L. gallinarum, L. gasseri, L. johnsonii, L. helveticus, L. delbrueckii subsp. bulgaricus, L. salivarius subsp. salivarius, L. casei, L. paracasei subsp. paracasei, L. paracasei subsp. tolerans, L. plantarum, L. rhamnosus, L. fermentum, and L. reuteri [18].

Probiotics are widely used in commercial functional products of animal origin, mainly fermented milk, such as yogurt and cheeses; however, the use of milk-based products may be limited by allergies, cholesterol diseases, dyslipidemia, and vegetarianism, and therefore, several raw materials have been extensively investigated to determine if they are suitable substrates to produce novel nondairy functional foods [16, 19].

Nondairy probiotic products are found to a lesser extent in market and are usually restricted to traditional products based on cereals or soy [19]. The number of articles on the use of nondairy matrices for the transport of probiotic microorganisms increases each year (Table 1), which demonstrates the increasing interest they have received in the health sciences literature. From a scientific point of view, it is unquestionable that probiotics constitute an important field of investigation and study, with the digestive tract, more specifically the intestinal microbiota, as the main point [20, 21].

Fruit and vegetable juices can be considered as a new category of food matrices for probiotic bacteria [37] with developments reported in literature [38, 39]. Particularly, fruit juices have been reported as a novel and appropriate medium for probiotic for their content of essential nutrients. Moreover, they are usually referred as healthy foods, designed for young and old people [40].

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Fruit juices are indeed promising probiotic carriers due to their essential nutrient content along with their appeal to a niche of consumers who already care about healthier habits. Fruits are healthy and refreshing and have good taste and flavor profile and can be suitable for probiotics [41, 42].

The observations that plant compounds, like complex carbohydrates and phenolics, may act synergistically with probiotics [43, 44] in formulations for gut health were important for nondairy probiotic product developers. Every food category from cereals to soy, to fruits and vegetables, has been the subject of research for new product development.

The use of probiotics represents a promising, rapidly growing area for the development of functional foods. Probiotic crops are successfully applied to different food matrices [45]. However, the development of nondairy products represents a challenge for the industry, as each food matrix has unique characteristics, and it is necessary to optimize and standardize each type of product.

Probiotic foods should be safe and contain sufficient probiotic microorganisms during the shelf life of the product. Therefore, selected probiotic strains should be suitable for large-scale industrial production, with the ability to survive and retain

| Probiotic                        | Food matrices | Food product                  | References |
|----------------------------------|---------------|-------------------------------|------------|
| *L. rhamnosus*                   | Apple         | Apple cubes                   | [22]       |
| *L. casei* NRRL B-442            | Cashew        | Cashew juice                  | [23]       |
| *L. casei*                       | Melon         | Melon juice                   | [24]       |
| *L. casei* NRRL B-442            | Cashew        | Dehydrated cashew juice       | [25]       |
| *L. plantarum* 33                | Olive         | Olive paste                   | [26]       |
| *L. casei* 01                    | Lychee        | Dehydrated lychee juice       | [27]       |
| *L. casei* NRRL B-442            | Orange        | Dehydrated orange juice       | [20]       |
| *L. rhamnosus* HN001, *L. acidophilus* LA-5 e *L. plantarum* | Pineapple, banana, guava, apple, papaya, and mango | Fruit salad | [28] |
| *Bifidobacterium animalis* spp. lactis | Jussara       | Dehydrated jussara juice      | [29]       |
| *L. casei* NRRL B-442            | Orange        | Dehydrated orange juice       | [30]       |
| *L. paracasei*                   | Orange        | Orange juice                  | [31]       |
| *L. plantarum*                   | Apple         | Apple cubes                   | [32]       |
| *L. salivarius* spp. salivarius CECT 4063 | Mandarin     | Mandarin juice                | [33]       |
| *L. plantarum* LSS               | Lemon         | Lemon juice                   | [34]       |
| *L. plantarum* DW12              | Coconut water | Fermented drink of coconut    | [35]       |
| *L. casei* NRRL B-442            | Apple         | Dehydrated apple cubes        | [36]       |
| *L. plantarum* MTCC2621          | Lychee        | Dehydrated lychee juice       | [21]       |

Table 1. Studies on the use of nondairy matrices for the transport of probiotic microorganisms.
their functionalities during food processing and storage [17]. Several strains of *Lb. plantarum*, *Lb. acidophilus*, and *Lb. casei* can grow in fruit matrices due to their tolerance to acidic environments [46].

During food storage different factors may affect the viability of probiotic bacteria, such as probiotic strains used, pH, the presence of hydrogen peroxide and dissolved oxygen, buffering form, storage temperature, the nature of the ingredients added, and food matrices [23, 47, 48].

In order to exert beneficial effects on health, the number of viable cells of probiotic microorganisms should be located above 10⁶ CFU.g⁻¹ in the product for consumption, available over the entire shelf life. Therefore, the preservation of probiotic cultures in products during storage is of extreme importance [49].

In this context, microencapsulation of probiotic cells has been widely studied as a technique to improve the stability of these microorganisms by protecting them from unfavorable environments [17]. Microencapsulation also has a potential effect on reducing post-fermentation acidification and possible negative sensory effects of probiotic food products [50].

Among the microencapsulation technologies, spray drying and freeze drying are the most commonly used. However, spray drying is the most effective for large-scale industrial production because it is a continuous, rapid process and has relatively low cost and high reproducibility [51, 52]. It is suggested as a technique that improves the survival of probiotics during food processing and storage, as well as confers protection of probiotics against subsequent exposure to the harsh conditions of the gastrointestinal tract, as this process gives a coating to the cells, protecting them from the outside environment [53].

Among the advantages of atomization, a distinguished one is the ability to handle heat-sensitive materials with high surface area/droplets volume ratio, resulting in shorter time of exposition to drying temperature [54]. Besides protecting probiotic cells from adverse conditions, powders obtained through spray dryer have good reconstitution and low water activity and are suited for storage at ambient temperature, what it is desirable, especially in commercial applications, due to higher operational costs associated with cooled storage, transport, and distribution difficulties [55, 56].

The characteristics of the powder produced in driers depend mainly on the operational variables of the drier (air inlet and outlet temperatures), on the product composition, solid concentration, feed flow rate, and also on the type of encapsulating agent used in the formulation [57].

Several studies reported that microencapsulation by spray dryer might provide a more favorable anaerobic environment for sensitive probiotic bacteria, as well as a physical barrier from the harsh acidic conditions of fruit juice [58]. The addition of probiotics in different fruit juices to produce functional beverages microencapsulated by spray drying has also been reported [21, 25, 27, 29].

The most commonly used carrier materials for encapsulation are maltodextrin. Maltodextrin (MD) is a polysaccharide, which molecular weight and properties depend on the hydrolysis process employed to obtain it from starch. Maltodextrin is classified by its dextrose equivalent (DE) which measures the amount of reducing sugars present in a sugar product, relative to dextrose [59–61].

The wall material is one of the most important parameters in the food microencapsulation processes. Its chemical composition and structure can affect the quality of the powdered product and criteria, such as solubility, apparent density, absolute density, porosity, particle size distribution, morphology, hygroscopicity, cell viability, water activity, moisture content, and sensory evaluation [61, 62].

However, to date there are no reports on the use of various probiotic microorganisms incorporated into a mixed beverage. In view of this, aiming at
complementing nutrients, increasing nutritional characteristics, and developing new flavors, our research group has been directing studies in this area in order to provide results of scientific interest and to food industry.

2.1 Microencapsulated probiotic mixed fruit juice

In recent years our research group has been carrying out studies with microencapsulation of juices from various fruits, using maltodextrin as the main encapsulating agent. In a recent study [63], with mixed juice of tropical fruits, the process was optimized in order to obtain products with better sensorial and nutritional characteristics. Based on these results, in order to meet the growing demand for functional foods, probiotic microorganisms were added.

The study related to the process of addition of probiotics to mixed juice in powder is an innovation, consisting of a new food product. Thus, the patent of product and process category was registered, at Brazil’s National Institute of Industrial Property with Patent nature of Invention, under register number BR 102019 009006 5.

The objective was to develop a novel nondairy probiotic product, composed of mixed juice with three Lactobacillus microencapsulated by spray drying using maltodextrin DE 5.

Currently there is a growing market for juices composed of more than one fruit, and this tendency is most observed in products that use tropical fruits. Tropical fruits are widely accepted by consumers and are important sources of antioxidant compounds. For this reason the acerola and siriguela were selected.

Acerola (Malpighia emarginata D.C.) is a fruit native to Central America and Northern South America, with some of its largest plant area in Brazil, which has been increasingly produced, because of their high vitamin C contents from 700 to 1400 mg/100 g [64–66]. The siriguela (Spondias purpurea L.) is a fruit from Anacardiaceae family originally from Central America and widespread in all tropical countries, mainly in the northeast region of Brazil. It is a small yellow fruit, with a pleasant aroma and taste, being a source of carotenoids [67, 68]. Thus, acerola and siriguela juice is an interesting nondairy matrix for a probiotic beverage.

The viability of probiotic bacteria is the most important parameter in the spray drying process using microorganisms due to heat inactivation and exposure to dehydration, and maximum viability is there for the major goal for this type of product [25, 69]. Probiotic mixed powder juice with maltodextrin DE 5 presented viable cell counts above 6.0 log CFU.g\(^{-1}\) (Table 2), which is the minimum

| Analyses                         | Maltodextrin DE 5      |
|---------------------------------|------------------------|
| Microbial viability (log CFU.g\(^{-1}\)) | 8.50 ± 0.04            |
| Water activity                  | 0.12 ± 0.00            |
| Moisture content (%)            | 2.09 ± 0.06            |
| Hygroscopicity (g.100 g\(^{-1}\)) | 24.40 ± 0.70           |
| Apparent density (g/mL)         | 0.38 ± 0.01            |
| Absolute density (g/mL)         | 1.97 ± 0.01            |
| Porosity (%)                    | 31.13 ± 0.29           |
| Solubility (%)                  | 92.75 ± 2.03           |

Table 2. Physical properties and microbial viability of acerola and siriguela probiotic mixed juice microencapsulated by spray drying using air inlet temperature 140°C, feed flow rate 0.60 L/h, and 10% maltodextrin DE 5.
recommended level for probiotics in food products necessary to produce therapeutic benefit [25, 70].

Probiotics’ survival during spray drying can also be attributed to the strong adherence to the carrier, which protects cells from high acidic and bile conditions. Overall, maltodextrin is confirmed to serve as a good encapsulating matrix as well as a moderate prebiotic for high survival of probiotics. The sugars present in juice may also have contributed to the survival during spray drying since sugars act as thermoprotectants [71].

Moisture and water activity are important indexes for spray-dried powders due to their effects on the shelf life of the product; a large amount of retained water can accelerate degradation reactions and microorganism growth [72]. It is necessary that its equilibrium moisture content is less than 5 g water/100 g of dry solid and its water activity is in the range from 0.1 to 0.4, ensuring greater stability for dry food [73]. The moisture and water activity for probiotic mixed powder juice (Table 2) are in agreement with the study of [73]. Values similar were obtained by [20, 27, 30] studying orange, lychee, and orange juice probiotic microencapsulated by spray drying, respectively.

Hygroscopicity is the ability of a material to attract and hold moisture from the environment. It is generally calculated from the weight gain after storing the food powder in a high humid desiccator (relative humidity more than 60%) for a period (generally 1 week) [74, 75]. The hygroscopicity value (Table 2) was similar compared to other probiotic microencapsulated formulation [27] and much larger than [21]. The hygroscopicity can be easily controlled by using suitable packaging, maintaining the integrity of the product without significant changes.

Solubility, the ability of a powder to form solution or suspension in water, is defined as the most reliable criterion to evaluate the behavior of powder in an aqueous solution. This parameter shows the ability of the spray-dried powder to form solution or suspension in water [76]. Solubility of powders can be affected by many parameters such as initial composition of the raw material to be microencapsulated by spray-dried, compressed airflow rates, low feed rates, and the carrier agents [77]. In this study, high solubility values were obtained with maltodextrin DE 5 (Table 2). The characteristics of solubility normally contradict to apparent density; the powder showing high solubility should have low apparent density [78].

The absolute density of the particle matches the actual density of the solid and does not consider the spaces present between the particles, unlike the apparent density. A similar result with this study (Table 2) was reported by [75] using maltodextrin DE 10 for acai pulp drying.

The higher values of porosity indicate the presence of a larger number of spaces between the particles, containing oxygen available for degradation reactions [79]. In this study (Table 2), lower value of porosity was obtained. In this way, the maltodextrin DE 5 is suitable for the spray dryer process. The efficacy of low-DE maltodextrin as drying carriers is due to the encapsulating property and low moisture diffusivity [80].

Particle size distribution is an important physical property that directly affects the application of microcapsules into food formulations [81]. The microcapsules exhibited lower particle size distribution (7.07 μm ± 0.03). Small particles are preferred in food formulations for ensuring homogeneity and quality, since they have large surface area and enable interaction with microorganisms favoring the microbial reactivation [82] (Figure 1).

The morphological characteristics of the particles showed spherical shapes with various sizes that are features of spray-dried powders and surfaces with wrinkled predominance and few particles with smooth surface. The occurrence of roughness
on the surface of the microparticles may be associated with shrinkage of the wall material during initial stages of the process and temperature used in the drying chamber [83, 84]. With these results, a great potential for the use of such powders in the food industry is observed (Figure 2).

3. Conclusion

The development of new functional food products is very challenging, and it has to complete the consumer’s expectations for palatable and healthy products. Probiotic cells can be stabilized with microencapsulation to preserve them from detrimental processing and storage factors such as high acidity and low pH. Therefore, there is a potential market for nondairy probiotics such as vegetable-based products, fruit-based products, cereal- and legumes-based products, confectionary products, and breakfast cereals. The present investigation concludes that it is possible to obtain probiotic foods from several matrices, including fruit juice. The acerola and siriguela mixed juice is a suitable medium for the incorporation of *Lactobacillus* spp. with suitable counts (>6 log CFU g⁻¹), demonstrating to be an ideal substrate for the culture of probiotics, since it already contains beneficial nutrients such as minerals, vitamins, dietary fibers, and antioxidants. The microencapsulation by spray drying of probiotics in acerola and siriguela mixed juice is a viable alternative,
since the powder produced showed favorable physicochemical characteristics, suggesting their production for various applications in the food industry.

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**Conflict of interest**

The authors declare no conflict of interest.

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