



## **A REVIEW ON FRUIT JUICE PROBIOTIFICATION MAJOR CHALLENGES AND TECHNOLOGICAL CONSTRAINTS**

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### **Abstract:**

Fruit juices are now integral part of dietary regime and are widely accepted by consumer as part of daily healthy diet plan. In light of the research based on the food fortification the juices are mainly studied for the formulation of probiotic juices. Many juices till date have proven as good source for the delivery of probiotic in human intestine. More research still needs to encourage for development of quality probiotic juices from variety of fruits. The technological constrains needs to resolve for better commercialization. To gain the wide acceptability of probiotic juices the sensory acceptability should also be reviewed critically. Nowadays, there is huge demand for concentrated juices and probiotic juices. Juice composition, type of probiotic strain and processing conditions are the major challenges in the development of ideal probiotic fruit juice, these needs to be focused during the formulation of probiotic juices .Because of variations in the nutrient composition and bioactive profile of fruits and even reported suitability of specific probiotic stains for particular juice, the formulation of quality probiotic juice of different fruits is presently a great scope of research.

**Key Words:** Fruit Juice, Probiotic, Sensory Property & Bioactive Components

### **Introduction:**

Extensive research has been carried out on the health benefits of fruit juices which have indicated the vast health attributes associated with all fruits. Even health conscious driven food acceptability now made the fruit juices as a part of a balanced diet which have proven profound disease risk reduction properties. Fruit juices are at high demand as alternative medicine for different kinds of illnesses and their moderate consumption has shown effect on chronic inflammation, diabetes, high blood pressure, muscle ache and pains, headaches, heart disease, cancers, mental depression, poor digestion, blood vessel problem drug addiction. In juices minerals are present in the form of electrolytes so they are easily absorbable by the human body. They are also rich in diverse sources of vital nutrients which include vitamins such as vitamin A vitamin B, vitamin C folate and minerals like iron, copper, potassium, iodine, zinc, selenium, sulfur, manganese, boron, molybdenum and magnesium, dietary fiber and antioxidants. Amino acids and bioactive compounds are the main phytonutrients of juices. They are mainly composed of water (70-93%) and other hundreds of compounds, like carbohydrates (glucose, fructose, starch, cellulose, and hemicellulose), amino acids, proteins, lipids, vitamins, fibers, acids, phenolic compounds, carotenoids, organic acids and minerals. These compounds contribute to taste, aroma, color, and texture of the fruit juice (D. M. Barrett *et al.*, 2010).

Being a readily digestible juices have no toxic effect on the body and exert cleansing effects on the blood and digestive tract, they also facilitate absorption of certain nutrients. Fruit juice in daily diet has been strongly associated with reduced risk of some forms of cancer, cardiovascular disease, positive effect on the bone health and skin related problem, allergies, gastrointestinal problem, insulin resistance, oxidative stress, inflammation, dental health, brain health, ageing, and other chronic diseases. Fruit juices also prevent the formation of kidney and gall stones, due to their high amount of potassium salt. Fruits were directly consumed by health conscious people but in today's era of changing lifestyle, modified eating habits and in view of seasonal availability, people have shifted towards nutritious ready to eat or ready to serve products like fruit juices (R. L. Bhardwaj *et al.*, 2014).

Research pertaining to development of juices as functional foods, lots of study is facilitated in the area of "Probiotic juices". Generally probiotics are inclusive in dairy products, dairy fermented products have been considered as best carrier for probiotics, but these products are not always a good choice for incorporation of probiotic. Recently up to 70% world population affected by lactose tolerance. So now a day fruit juices have been analyzed as a novel and appropriate carrier for probiotic for their content of essential nutrient. Usually they are considered as healthy foods, design for young and old people. The best way to increase probiotic stability in fruit juice would be the fortification of juice with some prebiotics or some ingredients which are able to exert a protective effect (M. Perricone *et al.*, 2015).

Minimally steps of vegetables peeling and cutting promote the release of the cellular content rich in minerals, sugar, vitamins and other nutrients creates the ideal condition for microbial growth (Oliveira *et al.*, 2011) but this characteristic allows the use of fruit and vegetable food products as a probiotic carriers (C.Roble *et al.*, 2010). Studies on the development of various probiotic juices also reported the negative sensory attributes

in terms of flavor and aroma that can be mask by blending the juice with different aroma dominant juices or extracts mango or pineapple juices can be the choice for same (T. Luckow et al., 2006).

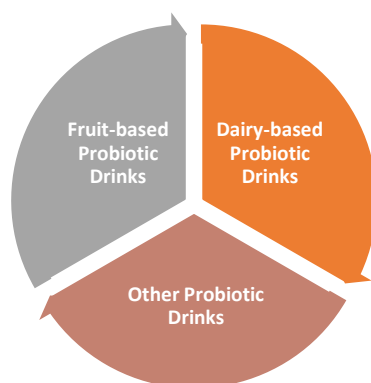
Considering the major limitations associated with the formulation of probiotic juices and in order to satisfy the aspirations of consumers more research needs to be facilitated to brought the variety of probiotic juices in market.

#### **Need of Probiotic Juices:**

The probiotic juice market is driven by the increasing health-conscious among the consumers. Probiotic drinks are the functional beverages, which improves the health conditions of the gut. Development of non-dairy probiotic products, like fruits, vegetables and cereals has been utilized as one of the best choices and increase the demand for nondairy probiotic (Yoon et al., 2006; Prado et al., 2008; Granato et al., 2010 a,b). The structural characteristic and composition (nutrients like minerals vitamins, dietary fibers and antioxidant) of fruits and vegetables and cereals are suitable substrate for probiotic microbes (Reddy et al., 2015). Development of fruit juice based probiotic demand is increasing nowadays as the consumers are more attracted because of taste, nutrient and the general acceptance as being healthy and refreshing food (Nulkaekul et al., 2011).

As per available data, dairy based products approximately 43% of the functional beverage market, and it is mostly comprised of fermented products (Ozer and kirmaci, 2010). Fermented milk like yogurt –style products, are the most popular functional probiotic beverage with kefir in Western Europe and North America. Traditional dairy fermented products have been consider as the best vehicle for the probiotics to carry it due to their easy processing. Consumers are widely accepting milk, yogurt, cheese, milk proteins, and milk related desserts for the probiotification (Boza- Mendez.et al., 2012.) Dairy products being a high in nutritional content especially proteins and high amount of lipid offers the protection against the adverse conditions of digestive tract. Because of the same reason many probiotic milk formulations include milk proteins as a carriers for probiotics. (Ritter *et al.*, 2009)Increasing lactose intolerant (around 70% in Asia), milk protein allergic population and being high in cholesterol content made the consumer to demand the non-dairy probiotic products. Even the Consumer vegetarianism in the developed countries are one of the reason for increasing demand of vegetarian probiotic products. (Heenan et al., 2004; Yoon et al., 2006). However because of provision of various combination of the antioxidants, dietary fiber, minerals and vitamins. The nondairy functional /probiotic foods market have annual growth rate of 15% between 2013 and 2018 (Markets and Markets, 2013). U.S. functional food market is developing in a various fashion from that seen in Europe, the functional food sector has defined as nutraceutical and consumer attraction tending to lie more with botanical dietary supplement rather than fortified of food.

The global probiotic drinks market is projected to grow at a CAGR 7.5% during the forecast period. (<https://www.mordorintelligence.com/industry-reports/probiotic-drinks-market>) Nowadays dairy-based probiotic drinks segment and fruit-based probiotic drinks segment is attracting market. As there is growing demand for functional food and beverages many fruit and beverages companies have started manufacturing functional beverages which mainly includes probiotic drinks. Asia-Pacific is the largest and fastest growing market for functional juices. In the Asia-Pacific region, majority of probiotics are consumed in the form of yogurts and fermented products (from soy to vegetables and even eggs). Drinking yogurt is in demand as a source of probiotics from the region.



Global Probiotic Drinks Market is segmented by Product Type

#### **4. Probiotic Juices Research Insight:**

Fruit and vegetable juice based beverages have been proposed as new product. Recently studies have shown fruit and vegetable juices could be suitable carrier vehicle for probiotic bacteria (U. A. Shaikh et al., 2018).Till date studies have reported various fruit and vegetable juices, like tomato, mango, orange, apple, grape, peach, pomegranate, watermelon, carrot, beet root and cabbage juices are good raw material for the production of probiotic juices. The Cornelian cherry (*Cornus mas L.*) juice has been studied as a medium for the production of probiotic beverages (Mantzourani et al., 2018). Study reported antibacterial and radio protectives

properties of cornelian cherry juice against most of the pathogens and also exhibits antihistamine, cytotoxic, anti-malarial, and anti-inflammatory effects (F.H. Jaghdani et al., 2017). Viability of probiotics in fruit juices is critical factor (probiotic food product should contain about  $10^6$ – $10^7$  cfu/mL probiotic cells at the time of consumption). Cabbage juice is proven well medium at 37°C for growth of probiotic bacteria. The *L.plantrum* and *L.delbruekii* were proven as suitable probiotic bacteria for the production of healthy beverages from the cabbage (K.Y.Yoon et al., 2004). Studies on cashew apple probiotic juice using *L.casei* shows good viability till 42 days. In this study *L. casei* was able to overcome the natural micro biota of cashew apple juice dismissing thermal treatment. Being a good source of ascorbic acid and phenolic content cashew apple is considered a good source of antioxidant compounds also (A.L.F. Pereiraa et al., 2017).

S.No	Probiotic	Juice	Reference
1	<i>B.longum, L.acidophilus, B.lactis</i>	Apple	W.K.Ding et al.,(2008)
2	<i>Lactobacillus casei, Lactobacillus debrueckii, Lactobacillus plantrum</i>	Cabbage	K.Y.Yoon et al.,(2005)
3	<i>B.longum, L.acidophilus, B.lactis</i>	Orange	W.K. Ding et al.,(2008)
4	<i>Lactobacillus delbrueckii, Lactobacillus plantrum</i>	Sweet Orange	H. W. Deshpande et al., (2017)
5	<i>Lactobacillus casei, Lactobacillus paracasei Lactobacillus rhamnosus</i>	Pineapple	M. perricone et al., (2015)
6	<i>Lactobacillus acidophilus, Lactobacillus casei, Lactobacillus plantrum</i>	Tomato	M. Perricone et al., (2015)

#### Commercial Juices:

S.No	Company name	Product	Type	Microorganisms
1	Golden Circle	Healthy Life Probiotics	Fruit Juice	<i>Lactobacillus paracasei 8700:2</i>
2	Tine BA	Biola	Fruit Juice	<i>Lactobacillus rhamnosus GG</i>
3	Copper Moon	Colombia Probiotics And Frenchvanilla	Coffee	<i>Bacillus. Coagulans GBI 30 6086</i>
4	Tropicana Essentials Probiotics	Juice Strawberry Banana	Fruit Juice	<i>Bifidobacterium lactis</i>
5	Farmhouse Culture	Mango Guava	Fruit Juice	<i>Bacillus coagulans GBI-30</i>
6	Perkii	Perkii Probiotic Water	Flavoured Water	<i>Lactobacillus casei [Lc431]</i>
7	Biogaia	Rela	Fruit Juice	<i>Lactobacillus reuteri MM53</i>
8	Farmhouse Culture	Jalaperio Gut Shot	Vegetable Juice	<i>Bacillus coagulans GBI-30</i>
9	Suja	Suja	Flavoured Water	<i>Bacillus coagulans GBI-30 6086</i>
10	Next Foods	Good Belly	Fruit Juice	<i>Lactobacillus plantarum 229v</i>
11	Skane Dairy , Sweden	Proviva	Fruit Juice	<i>Lactobacillus plantarum 299v</i>
12	Naked Juice USA	Probiotic Naked Juices	Fruit Juice	<i>Bifidobacterium sp.</i>
13	H-E-B, USA	Kevita	Fruit Juice	<i>Bacillus coagulans GBI-306086, Lactobacillus paracasei, Lactobacillus plantarum,</i>
14	Skane majerier, Sweden	Bravo Friscus	Fruit Juice	<i>Lactobacillus plantarum HEAL9 and Lactobacillus paracasei 8700:2</i>

#### Challenges in Probiotic Juice Formulation:

Survival of probiotic is important aspect in the formulation of probiotic food products and juices. The survival of probiotic is governed by number of factors as given below

- Food parameters: pH, titratable acidity, molecular oxygen, water activity, presence of salt, sugar and chemicals, like hydrogen peroxide, bacteriocins, artificial flavoring and coloring agents;
- Processing parameters: heat treatment, incubation temperature, cooling rate, packaging materials and storage methods, oxygen levels, volume;
- Microbiological parameters: strains of probiotics, rate and proportion of inoculation (Marianne Perricone et. al., 2015)

Survival of probiotics in juices is quite challenging because of low pH and antimicrobial effects of acids. Among the probiotics *Lactobacillus* spp. is one of the resistant strain as compare to *Bifidobacteria* spp (Tripathi, M.K. et al., 2014). Nualkaekul *et al.*, investigated the factors that affected the survival of studies on *B. longum* in various fruit juices like orange, grapefruit, blackcurrant, pineapple, pomegranate and strawberry study reported 0.8 log CFU/mL cells after six week of storage at 4 °C, in orange, grapefruit, blackcurrant, and pineapple juices while quite highest cell count found in orange and pineapple juice. On the other hand, the probiotic was below the detection limit after one week in pomegranate and four weeks in strawberry juice. Moreover, in grapefruit was only 0.5 log CFU/mL (despite the low pH (3.21) and the high concentration of citric acid (15.3g/L). These results suggest that the survival was the result of synergistic and antagonistic action of some parameters, and in that phenolic compounds could play a significant role in determining probiotic viability. Generally, pH exerts a detrimental effect, but protein and dietary fiber could protect cells from acidic stress; the role of citric and malic acids is controversial, as they seemed to protect probiotics, whereas phenols could cause a strong viability loss. Probiotic survival in juices make them more resistant to acidic pH also and develop resistance for low pH and could remain survive in vivo conditions of gastrointestinal tract (Ranadheera *et al.*, 2014). Probiotification of food remains challenging because of all these reason and to protect the probiotic cells in food products microencapsulation strategy has been adopted by so many researchers.

### **Technological Feasibility for Fruit Juice Probiotification:**

Desirable properties required by probiotic strain to survive in the fruit based products includes type of strain, type of fruit juice appropriate physicochemical properties, optimal growth conditions ( water activity, processing and storage temperature, oxygen content and mechanical stress). The major parameters needs to be monitor includes measurement of pH, acidity, optical density, detection of organic acids, biogenic amines, nitrates and nitrites which are important indices of probiotic survival in juice. The selected probiotics strains should resistance to storage temperature and oxygen toxicity. *Lactobacillus* species highly resistance for low temperature and pH while bifidobacteria shows less survival in adverse condition of pH and temperature. The selected probiotics strains should be stable and viable in food products for long period of time and it should retain till reaches to gastrointestinal tract. Nowadays no. of researchers have performed the studies on fruit juice fermentation using probiotic strains reports from their research highlighted the suitability of fruit juices as a medium for growth of probiotic bacteria. The main reason pointed out by the scientific community stated the following reasons.

- Acidic pH of fruit juices is a favorable condition for probiotic growth. In fact, the pH of juices lies between 2.5 to 3.7 makes the strains more competent to resist the acidic pH of stomach.
- High acidity of juice is also one of the reasons which supports the survival of probiotic strains.
- Being a rich in saccharides juices supports probiotic bacteria.
- The phenolic content of fruit juice inhibit the pathogenic microorganisms in fruit juices and in turn minimizes of competition of pathogenic probiotic bacteria in juice.
- Despite this benefit of fruit juices as a medium for probiotic growth the only one constrains reported or noted is reduction in sensory characteristics.

However this can be nullified by use of advancement of food technology. Number of researchers have recommended use of non-thermal pasteurization of fruit juices before probiotification and even the technologies like high hydrostatic pressure, high intensity electric field pulses at super critical carbon dioxide have proven as successful in improving the quality of not only fermentation process but also improve quality of probiotic products. The active research in the arena of probiotic foods mainly coiling around in improving nutrition content, appearance, sensory and shelf life of fruit based probiotic products. Also reported with microencapsulation can be anticipated technology to protect the probiotics in the probiotics fortified food products. In micro encapsulation of probiotic bacteria microbial cell are encapsulated the forms of emulsion, extrusion and spray drying and polymeric materials such as Alginate, Gellans Gum, K-Carrageenan starch and Gelatin are commonly used as encapsulation material. Studies have been performed on development of Chitosan coated Alginate beads of *Lactobacillus plantarum* in pomegranate juice. The research limits with the assessment of physical properties probiotic beads includes size & hardness which effects on organoleptic sensory of probiotic fortified pomegranate juice. The said research also highlighted on the crucial factor of uniform size of micro and nano capsules for probiotification is one of the crucial factor in commercialization.

Presently, as per the references one of the method have proven as successful technology for commercialization. In light of the research on the probiotic juices though it is challenging probiotic fortified juices have potential to impart into new industrial option. More studies are required in line with the optimization of conditions required for probiotic growth (growth conditions varies from strains to strains and varies from physico chemical properties of different fruit juices). Considering the choice of consumer towards health beneficial food products. Fruit based probiotic foods as a category of best functional food needs extensive scientific work.

Research performed by Pereira and his colleagues proven the successful cultivation of *L.casei* in cashew apple juice. Studies on pineapple and cantaloupe melon juice have proven as a better medium for the cultivation of *L. casei*. However their findings indicate fruit juice sonication is a prerequisite for the cultivation *L.casei* (Costa MGM et al., 2013, Fonteles et al., 2012).

In studies performed by Naulkaekul and Charalampopoulos revealed the growth of *L.Plantarum* in orange, pineapple and black current juices. According to their findings citric acid concentration and pH are the crucial factors in determining the growth of probiotic bacteria in foods.

However the reason for decrease or lowering the growth of *L. Plantarum* in pomegranate and cranberry juice is correlated with the phenolic compound of pomegranate juice and pH of cranberry juice pH 2.5. Widely applied probiotic strains in dairy products includes *L.casei* DN- 114001, *L. rhamnosus* GG and *L.paracasei* (Sheehan V.M. et al., 2007). Same strains used successfully in orange, pineapple juice s and also proven good shelflife with these juices. *L.casei*, *L.acidophilus* and *L.rhamnosus* as well as *B.lactis* are the most applied strains in probiotification of fruit juices (Espirito- Santo A.P. 2011).

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