

# Benefits of Using Yacon (Smallanthus sonchifolius) As a Prebiotic in Dairy Beverages Fermented by Lactic Acid Bacteria

#### Bruna Castro Alves Plank, Marcio de Barros, Karla Biguetti Guergoletto, Pedro Henrique Freitas Cardines, Thais de Souza Rocha<sup>\*</sup>

Department of Food Science and Technology, State University of Londrina, Celso Garcia Cid Road, Km 380, 86051-970 Londrina, Brazil

## ABSTRACT

The following text discusses fermented milk, such as yogurt, and their health benefits. These products contain probiotics, which are live microorganisms that offer health benefits when consumed in sufficient amounts, like improving gastrointestinal health and boosting the immune system. In addition to probiotics, fermented milk can also contain prebiotics, which are non-digestible food compounds that support the growth of beneficial bacteria in the intestine. Yacon, a plant native to the Andes region, is rich in prebiotics like inulin and Fructo-Oligosaccharides (FOS). Consuming yacon has been linked to several health benefits, including regulating blood glucose levels, controlling body weight, and positively affecting the intestinal microbiota. Adding yacon to fermented milk can enhance these products' health benefits, as the high concentration of prebiotics in yacon provides a favorable environment for probiotics in the intestine. Research has shown that this combination can improve digestibility, increase nutrient absorption, and promote a balanced intestinal microbiota, leading to overall better health. Therefore, fermented milk enriched with yacon presents a synergy between probiotics and prebiotics, creating a functional food with multiple benefits and displaying promises for future research and development in the realm of innovative food products.

Keywords: Inulin; Fructooligosaccharides; Probiotics; Microbiota; Health benefits

# INTRODUCTION

Throughout history, fermented milk has been consumed for its ability to preserve milk and its positive effects on health. They are rich in proteins, vitamins, and minerals and are also a great way to introduce probiotics into our diets [1]. Probiotics are live microorganisms that offer health benefits when consumed in adequate amounts. *Lactobacillus* spp., are commonly used probiotics, particularly in dairy products [2]. For probiotics to be effective, they need to survive the journey through our digestive system and reach the colon in sufficient numbers to colonize the intestinal environment. When consumed alongside prebiotics, probiotics are substances that the human gastrointestinal tract cannot digest, but they provide health benefits by supporting a

selective part of the intestinal microbiota. Fructooligosaccharides (FOS) are a group of prebiotics commonly found in foods.

Yacon, a plant native to the Andes region, is known for its sweet tasting roots, which are high in water and FOS. Studies have shown potential health benefits associated with its consumption, such as reducing the glycemic index, providing antioxidant activity, and promoting the growth of probiotic microorganisms through the fermentation of FOS [5].

Fermentation not only extends the shelf life of milk but also enhances its sensory qualities and leads to the production of metabolites with potential physiological activity [6]. For example, during fermentation and digestion, milk proteins are broken down into smaller structures known as bioactive peptides, which can have various health benefits, including immunomodulatory, antihypertensive, and antioxidant effects [7].

Correspondence to: Thais de Souza Rocha, Department of Food Science and Technology, State University of Londrina, Celso Garcia Cid Road, Km 380, 86051-970 Londrina, Brazil; E-mail: tsrocha@uel.br

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The human body produces free radicals through metabolic reactions, which leads to oxidative stress when excessive. Consuming foods with antioxidant compounds can help manage the balance between oxidizing and antioxidant molecules, potentially preventing diseases caused by oxidative stress [8].

Given the potential prebiotic effect of yacon root and the health benefits of milk fermented by probiotics, this work aims to explore the combined benefits of yacon and fermentation processes on health.

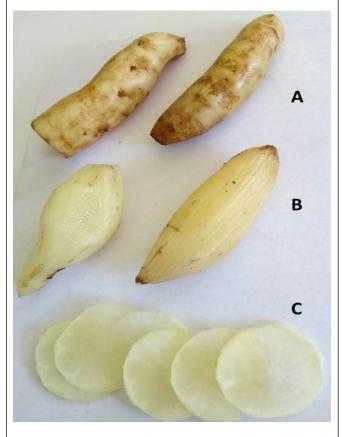
# LITERATURE REVIEW

#### Study design

This study consisted of a narrative bibliographical review of articles and books published in English. The review focused on the keywords fermented milk, yacon, fructooligosaccharides, probiotics, and prebiotics, both alone and in various combinations. Inclusion criteria comprised studies related to these components published between 2014 and 2024, while exclusion criteria included studies that did not meet the specified time frame or did not provide relevant information about the benefits and applications of these components in food. Some crucial information about definitions and primary knowledge prior to 2014 was maintained. The information was gathered from databases such as Scopus, Web of Science, Google Scholar, Science Direct, Lilacs, and Scielo.

#### Yacon

Yacon, scientifically known as *Smallanthus sonchifolius*, is a flowering plant originating in the Andean region and has garnered significant attention worldwide due to its diverse array of uses. Beyond its culinary appeal, yacon is esteemed for its remarkable nutritional properties, particularly its high concentration of Fructooligosaccharides (FOS), which imbue the root with prebiotic attributes. Although it is a tuberous root (Figure 1) akin to carrots and sweet potatoes, it is often categorized as a fruit owing to its delectably sweet flavor reminiscent of melon [9].



**Figure 1:** Yacon roots. A) Unpeeled yacon roots; B) Peeled yacon roots; C) Transversal cut of peeled yacon root.

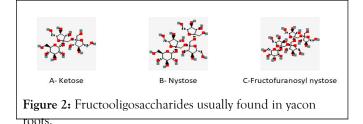
This versatile ingredient can be savored in its raw form, as well as fried or cooked, and serves as a key component in the production of various food items such as bread, syrups, juices, and fermented beverages (Table 1) [10].

Table 1: Average	yacon	composition	(g/	100	g).
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Components	Base seca (Dry base)
Carbohydrates	73.8
Fibers	4.81
Proteins	5.35
Lipids	0.55
Ashes	5.88
Potassium	1.79
Phosphorous	0.18

Calcium	0.06
Magnesium	0.05
Sodium	0.002
Iron	0.001
Vitamin B <sub>1</sub>	0.0004
Vitamin B <sub>2</sub>	0.002
Vitamin C	0.03
β-carotene	0.0007
Polyphenols	1.09

An average composition of yacon roots is presented in Table 1. Carbohydrates in yacon are primarily stored as Fructooligosaccharides (FOS), which remain stable even at temperatures of up to 140°C and are resistant to industrial thermal processing. Figure 2 displays the main FOS found in yacon roots, which are kestose, nistose, and 1-fructofuranosil nistose [4]. However, around 7 days post-harvest, FOS undergoes rapid hydrolysis, breaking into simple sugars [11].



In addition to FOS, yacon contains glucose, fructose, sucrose, traces of starch, and inulin [12]. Notably, yacon boasts a low caloric value and a high water content of approximately 85% of its weight, making it susceptible to rupturing and microbial proliferation. The carbohydrate composition varies based on factors such as the growing season and climate. FOS content can exceed 60% of the dry matter [13]. Furthermore, yacon harbors phenolic compounds endowed with antioxidant properties [14].

In order to prevent enzymatic browning, which is catalyzed by peroxidase and polyphenol oxidase enzymes, various techniques and antioxidants can be utilized. Dehydration and lowtemperature storage are effective methods for preventing enzymatic browning. Yacon flour production plays a vital role in deactivating enzymes without causing the degradation of Fructooligosaccharides (FOS) [4,14]. The use of heat allows the deactivation of enzymes without affecting FOS' integrity [15].

Driven by its health-promoting nutritional attributes, the consumption of yacon has surged, establishing it as a prominent prebiotic food. Research suggests that yacon enhances the viability of probiotics throughout digestion and fosters the proliferation of beneficial microorganisms [4,5].

In light of its identified nutritional and health-enhancing attributes, yacon root consumption has increased in recent decades, mostly for its potential as a prebiotic food [16].

# DISCUSSION

Studies conducted by Braschi, et al., [17] indicate that incorporating yacon into a probiotic product can contribute to maintaining cell viability during food processing and digestion. Their findings underscore the utilization of yacon's FOS by the Lactobacilli strains examined, thus positioning the root as a promising prebiotic source. Additionally, Leone, et al., have demonstrated the ability of yacon to shield bacterial cells during passage through the gastrointestinal tract, enhancing probiotic survival [3]. Another study has shown that adding yacon flour to milk fermented by *Lactobacillus plantarum* and *Lactobacillus casei* provides great stability to the strains and increases the rate of survival after simulated gastrointestinal digestion [4].

Furthermore, Bifidobacteria and selected Lactobacilli have exhibited the capacity to metabolize components present in yacon root, stimulating their growth and the synthesis of short chain fatty acids. Yacon flour, as illustrated in the work of Kahler [18], can function as a prebiotic by fueling the metabolism and expansion of probiotic microorganisms. This was also proved in the studies of Rolim, who developed prebiotic bread using yacon flour, and Gonzalez-Herrera, et al., who created a synbiotic beverage incorporating soy extract and yacon [19,20].

In addition to the prebiotic effect, studies suggest that yacon and its products, such as flour and syrup, can help increase satiety and control body weight [21-23]. In a study by Genta, 55 obese women participated in a double blind study for 120 days. They consumed the equivalent of 0.14 g of FOS from yacon syrup/kg/day. At the end of the experiment, a 16% reduction in body weight and a 10% reduction in abdominal fat were observed. Additionally, there were significant reductions in serum insulin and HOMA-IR parameters in comparison to those who consumed the placebo.

## Milk

Milk is a natural emulsion consisting of an aqueous liquid phase and suspended oily particles. The most commonly consumed milk worldwide is cow's milk, which comprises lactose (4.9%), lipids (3.5%), proteins (3.3%), minerals (0.7%), and vitamins. The protein content predominantly comprises approximately 80% casein and 20% whey proteins, including  $\alpha$ -lactalbumin and  $\beta$ -lactoglobulin. Furthermore, casein is made up of four main proteins:  $\alpha$ s1, $\alpha$ s2,  $\beta$ , and  $\kappa$ -casein [24].

In the context of milk processing, caseins are mainly found in the form of micelles, acting as colloidal particles offering essential technological characteristics such as thermal stability, coagulation potential using rennet, and contributing to the white color and texture of dairy products like cheese and yogurt [24].

Essentially, milk proteins are a crucial source of essential amino acids, influencing insulin secretion and aiding in mitigating postprandial glucose release, thereby reducing glycemic peaks and the risk of type 2 diabetes [25].

Milk boasts versatility as it serves as the foundation for several food items and provides essential nutrients, including carbohydrates, minerals, vitamins, and proteins [26]. Moreover, fermented milk, which has been consumed for centuries, not only helps preserve nutrients but also delays spoilage and offers numerous health benefits, particularly concerning gastrointestinal health, metabolism, and cardiovascular well-being [1].

#### Lactic acid bacteria

Lactic Acid Bacteria (LAB) are widely used in food fermentation due to their GRAS (Generally Recognized as Safe) status [27]. They are gram-positive bacteria and, according to their fermentation products, can be divided into two categories: heterofermentative bacteria that produce lactic acid, acetic acid, and  $CO_2$ , and homofermentative bacteria, which convert carbohydrates mainly into lactate [28]. Lactate is a short-chain fatty hydroxy acid that can be converted into short chain fatty acids such as acetate, propionate, and butyrate by some bacterial strains [29].

Bacteria of the genus *Lactobacillus* spp. are found in fruits, vegetables, naturally fermented products, and in the gastrointestinal tract of humans and animals [6]. Some probiotic Lactobacilli are capable of fermenting prebiotics due to the different enzymatic systems of the genus. Some probiotics have glycosidases capable of breaking down the prebiotic oligosaccharide molecules into monosaccharides that are then used by the microorganism. Some strains of *L. plantarum* also have specific transport systems for oligosaccharides, which allows their absorption and internal metabolization [30].

*Lactobacillus plantarum* is the most used species for the fermentation of fruits and vegetables [28]. In addition to metabolizing compounds with antioxidant activity, *L. plantarum* is associated with a reduction in inflammatory processes due to its influence on the large intestine and the immune system [31].

The wide use of *L. casei* in the food industry is due to its technological potential to improve the flavor and texture of fermented products and the health benefits that are conferred on products fermented by the species, such as the synthesis of metabolites with bioactivity and the maintenance of intestinal microbiota [6].

When LAB ferments foods, they improves their sensory attributes, increase food safety, and bring health benefits [27]. In fermentation, microorganisms convert carbohydrates present in the food into alcohols and organic acids [28], with milk being the primary raw material fermented by LAB. LAB carries out the hydrolysis of milk carbohydrates, which leads to the formation of organic acids, mainly lactic acid, which contributes to the development of the characteristic flavor and texture of fermented dairy products.

The metabolites formed by the fermentation of carbohydrates lead to acidification of the medium. When the pH of the milk approaches 4.7, the casein aggregation process occurs, which helps in the development of the texture of fermented milk [32].

A study examined the pH changes during refrigerated storage of milk fermented by *L. plantarum* and *L. casei*, with and without yacon flour. The study found that the pH decreased more in the beverages with yacon flour. This indicates that the *Lactobacillus* strains remained viable throughout the storage period, indicating metabolic activity and improved cell stability [4].

## Probiotics and prebiotics

The growing interest in functional foods has sparked an increase in the development of synbiotic products, driven by their associated physiological benefits, such as the production of bioactive compounds that help modulate physiological systems and improve intestinal function [29]. Probiotics are live microorganisms that, when consumed in adequate amounts, provide health benefits to the host. The most commonly used genera of probiotics include *Bifidobacterium* spp. and *Lactobacillus* spp. [33]. *Lactobacillus* spp., was recently reclassified into 23 new genera [34]. Ideal probiotic properties include nonpathogenicity, genetic stability, survivability through the gastrointestinal tract and processing, lactic acid production, antigenotoxic effects, rapid growth, and the ability to adhere to intestinal epithelial cells [2].

Consuming probiotics has been suggested to lower blood triglyceride and LDL cholesterol levels. One proposed mechanism for this reduction is that probiotics convert cholesterol into bile acids, increasing their excretion through feces. Research has also explored the clinical applicability of probiotics in maintaining health and preventing specific conditions like atherosclerosis and myocardial infarction [2].

Regular consumption of probiotics can help prevent diarrhea and constipation and promote a healthy intestinal microbiota composition. Probiotics can increase the production of short chain fatty acids, which help maintain a favorable pH in the intestinal environment and inhibit pathogen growth [35]. The primary short chain fatty acids produced by probiotics, including acetate, butyrate, and propionate, play pivotal roles in regulating the immune system, metabolism, and cell proliferation through cell signaling [36]. These short chain fatty acids are metabolized by certain microorganisms in the intestinal lumen during the fermentation of prebiotic substances [5]. Acetate and butyrate also play a role in managing mucus secretion, similar to the fibers found in prebiotic foods, which stimulate secretion mechanically. Consequently, a diet low in fiber intake can impact the protective barrier of the intestinal epithelium and the synthesis of short chain fatty acids.

In the 1980's, fermentable oligosaccharides garnered significant attention from researchers due to their ability to stimulate the growth of specific microorganisms. In 1995, these fermentable oligosaccharides were officially defined as prebiotics, indicating that they are substances capable of modulating the intestinal microbiota. The definition was further refined in 2010 to describe prebiotics as selectively fermented ingredients that bring about specific changes in the composition or activity of the gastrointestinal microbiota, hence providing health benefits to the host [33]. A subsequent update in 2016 by the International Scientific Association for Probiotics and Prebiotics [37] further refined the definition to describe prebiotics as "a substrate that is selectively utilized by microorganisms and confers health benefits on the host".

Common prebiotics such as Fructoseoligosaccharides (FOS) and inulin are often used in synbiotic foods [4]. Regular consumption of prebiotics has been linked to the regulation of metabolic diseases, modulation of the immune system, increased mineral absorption, and, primarily, the proliferation of beneficial bacteria in the colon [38]. Since prebiotics are mostly indigestible carbohydrates, they are classified as fibers, and their consumption is associated with enhancing the composition of the intestinal microbiota and mitigating inflammatory issues [37].

While fibers with prebiotic effects are naturally found in vegetables, fruits, and cereals, their consumption is generally low among the population. One strategy to increase intake is to incorporate fiber rich ingredients into daily foods. A diet low in fiber intake can potentially disrupt the balance of the intestinal microbiota, leading to inflammatory, chronic, allergic, and autoimmune conditions. Fibers alter the microbial balance in the colon, allowing for the expansion of microbial species capable of utilizing prebiotic substrates [36].

The ability of bacteria to derive benefits from prebiotic fibers depends on various factors, including the ability to break down the substrate using bacterial enzymes, adapt to environmental changes, and utilize secondary metabolites for their advantage [36]. Due to the variation in probiotics' ability to metabolize prebiotics based on the strain and substrate, it is essential to evaluate new combinations of probiotics and prebiotics for their efficacy in using the substrate [39].

Synbiotic foods, which contain both prebiotics and probiotics, can act synergistically to enhance the positive effects on consumers' health. This includes stimulating selective proliferation of the gastrointestinal microbiota, reducing pathogen adhesion to the intestine, and facilitating nutrient absorption [40]. With an increasing commercial interest in healthy and cost-effective foods, the prevalence of synbiotic foods in daily meals is expected to rise [41].

For probiotics to exert their effects, they need to survive passage through the gastrointestinal tract and reach the colon in sufficient quantities for colonization. This survivability is a crucial consideration in selecting probiotics [29]. Synbiotics can aid in the survival of probiotics under stressful conditions, increasing their chances of colonization [2].

#### Surviving the gastrointestinal tract

The human intestine is home to more than 400 species of microorganisms, most concentrated in the colon, reaching up to 1011 organisms per milliliter [42]. The composition of the intestinal microbiota can be influenced by diet, and one way to positively impact the microbiota is through the consumption of prebiotics, which is beneficial for the consumer's health and well-being [43].

Prebiotics such as Fructooligosaccharides (FOS) presented in yacon are fermented by particular microorganisms, such as Lactobacilli and Bifidobacteria, leading to the production of metabolites that inhibit the growth of harmful pathogens [44]. Furthermore, the viability of probiotics in the food is crucial for them to exert their effects and successfully colonize the colon. It is recommended that, at the time of ingestion, the viable cell count of the probiotic in the product is larger than 10<sup>9</sup> CFU per gram [45].

As probiotics may experience a decline in viability during their passage through the gastrointestinal tract, strategies to enhance their resistance and minimize this loss are being studied [46]. Prebiotics, since they are not digested throughout the gastrointestinal tract, have been found to increase the tolerance of bacterial cells in harsh conditions, thereby enhancing the viability of probiotics during storage and ingestion. While the exact mechanisms of prebiotic protection on bacterial cell viability are not fully understood, prebiotics are thought to act as a physical barrier, protecting probiotics from pH changes and gastric enzymes. Also, prebiotics serve as substrates for microorganisms during storage and in the epithelium, helping cellular maintenance [4,47].

In order to evaluate how resistant probiotics are to digestive enzymes and bile salts, simulated gastrointestinal digestions are used as an effective method to choose the best combination of microorganisms and food matrix to ensure that bacterial cells remain viable. This comprehensive assessment is vital in developing functional foods that contain probiotics and prebiotics to support gut health [48].

A study compared the effects of adding yacon flour to milk fermented by *L. plantarum* and *L. casei*. The study found that adding yacon flour improved the cell viability of the fermented milk after simulated gastrointestinal digestion. Additionally, it was observed that in the case of milk fermented by *L. casei*, the presence of more prebiotics in the formulation led to better protection of the cells during gastrointestinal passage. On the other hand, for milk fermented by *L. plantarum*, the protective effect of yacon flour was not dependent on its concentration [4].

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The study by Leone, et al., [3] confirmed the protective effect of probiotics when combined with FOS during their passage through the gastrointestinal tract. They included *L. casei* in dry yacon and assessed the survival rate after simulated gastrointestinal digestion in samples stored for 28 days. The study found that the percentage of viable cells was 82.81%.

#### **Bioactive** peptides

The processing of milk, such as fermentation or digestion, breaks caseins and whey proteins down into smaller molecules called peptides and amino acids. Some of these peptides have physiological effects, such as antimicrobial and antioxidant properties in the cardiovascular system. Because of their physiological effects, these molecules are referred to as bioactive peptides [7].

Maske, et al., [49] demonstrated that milk fermentation by Lactobacilli influences the release of amino acids and peptides during digestion by increasing the exposure of peptide sequences, making them more likely to be broken down by digestive enzymes. The LAB enzymatic system, composed of proteases and peptidases, can also result in the formation of bioactive peptides from milk proteins [27]. The ability of certain LABs, such as *L. casei*, *L. paracasei*, and *L. rhamnosus*, to produce peptides with physiological benefits is technologically interesting, as these microorganisms are already used as probiotics in dairy products. By synthesizing these peptides, they can contribute to the prevention of chronic diseases by directly impacting the pathophysiology or reducing oxidative stress [50].

Bioactive peptides produced from milk proteins can reduce the production of reactive oxygen species and protect cells from oxidative stress by capturing free radicals and acting as metal chelators [7]. Therefore, these fermented foods can also have antioxidant activity, helping to combat free radicals due to compounds released during probiotic metabolism, such as peptides released from the hydrolysis of milk proteins [50]. In the study by Plank, Guergoletto, and Rocha [4], the effect of antioxidant activity and the production of bioactive peptides on milk fermentation by the probiotics *L. plantarum* and *L. casei* in the presence of different concentrations of yacon flour was evaluated. The authors found that there was a significant increase in the antioxidant capacity of fermented milk after in vitro gastrointestinal simulation when compared to a control without yacon flour. These results imply that yacon has some influence during fermentation, leading to enhanced production of bioactive peptides with antioxidant properties, which could potentially benefit human health.

#### Atividade antioxidante

The nutritional benefits of yacon have caught the attention of consumers since there is an increase in demand for healthier foods [5]. Additionally, there is a growing trend in developing fermented dairy products with plant extracts [51]. These fermented foods may possess antioxidant properties, helping to combat free radicals. The antioxidant activity can be attributed to compounds released during probiotic metabolism, such as peptides from milk proteins [50].

Free radicals are produced in the body's biochemical reactions, where they act as defense and cell signaling molecules. However, excessive production of reactive oxygen species or decreased antioxidant defense leads to oxidative stress. This is defined as the inability of endogenous antioxidants to neutralize oxidative damage to biological targets and can trigger conditions such as cardiovascular disease or neurodegenerative disorders [52].

Antioxidants are substances that delay, prevent, or remove oxidative damage from a molecule by neutralizing free radicals. The reaction  $AH+FR \bullet \rightarrow A \bullet +FRH$  (where AH represents the antioxidant,  $FR \bullet$  the free radical, and  $A \bullet$  represents a less reactive secondary free radical) exemplifies the basic mechanism of action of antioxidants. In this reaction, the antioxidant donates a hydrogen atom to the free radical, stabilizing it into a less reactive molecule [53].

Antioxidants are not a cure for diseases related to oxidative stress. Oxidative stress is just one of many factors contributing to these diseases. However, antioxidants can help maintain the balance between pro-oxidants and antioxidants, preventing an excess of free radicals that can lead to the onset of these diseases. They act in prevention. Foods containing significant amounts of substances with antioxidant potential have been suggested as a way to strengthen the body's antioxidant defense and help in the prevention of chronic health conditions [54]. Eating foods rich in antioxidants is more beneficial and economical than consuming the substance alone. The combination of different types of antioxidants, such as fruits and fermented foods, is more effective in the long term [55].

The antioxidant defense system is complex, and no single in vitro method can fully replicate the body's mechanisms of action. Therefore, it is common to use multiple methodological principles to evaluate antioxidant capacity and gain different insights into the interaction of free radicals in the tested substances. Some widely used *in vitro* methods for determining antioxidant capacity include Ferric-Reducing Antioxidant Power (FRAP) and Oxygen Radical Absorbance Capacity (ORAC). These methods help in estimating the antioxidant capacity of substances and their interactions with free radicals in the evaluated matrix [56].

The inclusion of yacon flour as an ingredient in the making of fermented foods may promote greater consumption of probiotics and prebiotic fibers, thus contributing to improved health through dietary enhancement, as emphasized in the study by Plank, Guergoletto, and Rocha [4]. The study discovered that as the concentration of yacon flour in the milk fermented by L. casei increased, the antioxidant activity also increased, both before and after simulated gastrointestinal digestion. This revealed a direct relationship between the amount of yacon flour and the production of peptides with antioxidant properties in the fermented milk. Similarly, in the case of milk fermented by L. plantarum, there was a direct correlation between the amount of yacon flour and the antioxidant capacity during fermentation. However, after simulated gastrointestinal digestion, the production of peptides with antioxidant activity remained consistent regardless of the concentration of yacon flour added. When using the ABTS radical scavenging activity method, there was no observable influence of vacon flour addition on the results for milk fermented by both Lactobacilli. This suggests that the methodology used to assess antioxidant activity affects result interpretation. For evaluations aiming to determine health impacts, methods closer to those that can occur in the human body, such as FRAP and ORAC, should be applied [57].

## CONCLUSION

In this literature review, the use of yacon in fermented dairy products, like yogurt, demonstrates the promising combination of probiotics and prebiotics. This pairing not only improves the nutritional value of fermented milk but also expands its health benefits. The prebiotics found in yacon, such as inulin and fructooligosaccharides, promote the growth and activity of probiotics, leading to better gastrointestinal health, improved nutrient absorption, and a balanced intestinal microbiota. Additionally, yacon has been associated with regulating blood glucose levels and managing body weight, further highlighting its potential as a functional food ingredient. As ongoing research continues to confirm these benefits, the creation of innovative food products incorporating yacon-enriched fermented milk is expected to make a significant contribution to public health and nutrition.

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