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## Research Article

# Spray Drying Effect on the Viability of *Lactococcus lactis* in Presence of Yacon (*Smallanthus sonchifolius*) as a Prebiotic

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## Abstract

**Background and Objective:** Due to malnutrition and to the raise of non-communicable diseases, agrifood industry research has entered the development of functional foods; the “Yacon” is an Andean tuber with a prebiotic potential. The objective was to evaluate the effect of spray drying on the viability of *Lactococcus lactis* ATCC19435 in the presence of “Yacon” as prebiotic. **Materials and Methods:** Initially, the prebiotic effect of Yacon on *L. lactis* was studied by making a growth curve in Yacon’s nectar and by modelling data through the ComBase®; then *L. lactis* were spray dried in Yacon’s nectar analyzing the influence of the entrance temperature and the maltodextrin concentration. The micro-organism’s survival, water activity and moisture content were measured. Afterwards, the spray-drying effect over the bacteria stability in *in vitro* simulated gastrointestinal was tested. **Results:** The results show that *L. lactis* used fructo-oligosaccharides present in Yacon as a carbon source for its growth and metabolism with a viable population in a 92-98%, growth curve that fit Baranyi and Roberts model. Besides, it was possible to observe that the spray-drying technique used at 120°C-20% maltodextrin had a statistically significant effect over the bacteria stability in simulated gastric juice and bile fluids. **Conclusion:** Consequently, it is possible to infer that “Yacon” has a great potential to become a prebiotic product since a high fructo-oligosaccharides content was found both in the fresh root and in the powder; this is why it is cataloged as a promissory prebiotic food; beside, if probiotic bacteria such as *L. lactis* is added, it might be considered as a functional symbiotic powder food.

**Key words:** Lactic acid bacteria, stability, functional, probiotic, Yacon

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**Data Availability:** All relevant data are within the paper and its supporting information files.

## INTRODUCTION

Over the last few decades, the malnutrition problem has been more and more evident worldwide, impacting developing countries in an even more accentuated way<sup>1</sup>. Besides, the world's main causes of death are due to non-communicable diseases (NCD) (38 million dead people a year), out of which 75% occur in middle and low-income countries and 30% are attributed to food and behavioral factors<sup>2</sup>. The most common NCD: Cardiovascular ones, gastrointestinal ones, irritable bowel syndrome, arthritis, diabetes, hypercholesterolemia and cancer<sup>2,3</sup>.

Agroindustry research has led to the development of new tendencies in the last few years in order to diminish such problems. Thus, one of the world's main innovation axes is that of functional foods<sup>4</sup> and even though there is not a normalized definition of functional food yet, this might be considered as such if satisfactorily proven to give a beneficial action in one or more organism functions, beyond its nutritional effects, in such a way that it turns out to be relevant whether to improve the state of health and wellbeing or to reduce the risk of diseases<sup>5</sup>.

Prebiotics are among functional interest biocomponents. They're non-digestible food substances in a diet that produce beneficial effects selectively stimulating growth and/or activity of one or more types of beneficial bacteria in the colon, which at the same time, have the property of increasing the host's health potential<sup>6</sup>.

Some commonly known prebiotics are: fructo-oligosaccharides, inulin, galacto-oligosaccharides, lactulose, oligosaccharides from breast milk, among others. Fructo-oligosaccharides are obtained through synthetic processes from sucrose which undergoes transfructosylation with  $\beta$ -fructofuranosidase or from some food like wheat, onions, bananas, honey, garlic, leek, chicory root and Yacon<sup>7</sup>.

Yacon (*Smallanthus sonchifolius*) is an autochthonous root from the Andean region<sup>8</sup> presenting a high  $\beta$ -(2,1) fructo-oligosaccharides content which are soluble fibers with low-caloric content, known and used as prebiotics in food, stimulating non pathogenic gut flora growth as to strengthen immune response, enhance minerals absorption and lower sugar, cholesterol and triglycerides blood levels (metabolic diseases)<sup>9-11</sup>. Nonetheless, Yacon is very perishable despite all its benefits; it deteriorates quickly due to its high moisture content, microbial load and enzyme activity<sup>12</sup>.

Additionally, probiotics, described by the World Health Organization (WHO), are found within the wide range of functional foods as micro-organisms that provide the host with health benefits if taken in the right amount<sup>13</sup>.

According to the WHO, these probiotic foods must comply with some requirements such as containing at least  $10^6$  micro-organisms per gram or milliliter when consumed, supplying an antimicrobial effect, adhesion capacity, being non pathogenic and allowing the micro-organism's survival through the gastrointestinal tract, tolerating the stomach's low pH and the high bile salts conditions in the intestine<sup>13,14</sup>. This is why, encapsulation through spray-drying has become an alternative to potentially increase their stability prolonging the food's life cycle. This is due to the fact that it is a commonly used technique to protect the bioactive components against environmental adverse factors and to improve the delivery of molecules and living cells through entrapment in a coating material<sup>15,16</sup>. Spray-drying can give the cells protection through food processing and passing through the gastrointestinal tract<sup>17</sup>.

Therefore, the objective of this work was to test the microencapsulation effect through spray-drying on the viability of *L. lactis* ATCC19435 in presence of Yacon as prebiotic.

## MATERIALS AND METHODS

The study was conducted in the University San Buenaventura, Cali, Valle del Cauca and the University La Gran Colombia, Armenia, Quindío, during 9 months, starting from August, 2016 to May, 2017.

**Extraction and adaptation of raw material:** Yacon was bought in a local market in the municipality of Armenia, Quindío (Colombia). Its roots were cleaned with abundant water to remove any dirt and strange substances. Then, it was peeled and chopped in slices to be liquefied. Previously, citric acid at 0.2% had been added to these slices to avoid enzymatic browning as reported in Peruvian Technical Standards<sup>18</sup>. The resulting Yacon nectar was refrigerated for a maximum of 2 h for subsequent analysis.

### Testing Yacon's prebiotic effect on the micro-organism

**Biomass production:** Biomass production of the *L. lactis* ATCC 19435 lactic acid bacteria with probiotic potential<sup>19,20</sup> was carried out through progressive scaling, inoculating first in 10 mL of Brain Heart Infusion broth (BHI) (Scharlab, Spain) and it was brewed at 37°C for 48 h. The culture liquid was taken to 250 mL of Yacon's nectar enriched with peptone (Merck, Germany), sodium chloride (Merck, Germany) and disodium phosphate (Merck, Germany) and incubated at 37°C. A periodic process control was done (0, 2, 3, 5, 7, 9 and 10 h) during 10 h, evaluating population growth per culture in BHI

agar, determination of pH following AOAC 981.12 standard<sup>21</sup> using a pH meter (HANNA, Spain), total soluble solids by refractometry (Atago, Japón) under the AOAC 932.12 method<sup>21</sup> and lactic acid concentration by high-performance liquid chromatography (HPLC).

**Encapsulation effect over Yacon's features (moisture content, water activity) and probiotic's stability:** The encapsulation of the mixture of Yacon's nectar with the micro-organism was done through spray-drying in a Spray Dryer (Lab-Scale Spray Dryer, 7614YC015, Pilotech, China). Six treatments were completed where two variable factors were tested: three level drying temperature (110, 120 and 130°C) and coating material concentration (maltodextrin (Tecnas, Colombia)) with two levels (20 y 30%) and a set concentration of the micro-organism  $8.5 \pm 0.0723$  Log UFC mL<sup>-1</sup> cycles, 5 pressure bar and exit temperature of 70°C. Parameters were selected based on other studies and preliminary tests of Yacon's drying conditions.

Previous to the encapsulation process and after getting the powder, moisture content, water activity and lactic acid bacteria count were evaluated. Moisture content was determined through the gravimetric method following the AOAC<sup>21</sup> 934.01 method and water activity following the AOAC<sup>21</sup> 978.18 one using a water activity meter based on the dew-point method (Aqualab Lite, Decagon Devices Inc., USA).

Lactic acid bacteria recount in the powders was obtained by making a reconstitution of products in sterile peptone water (Merck, Germany) at 0.1%, for later serial dilutions and culture plate count in BHI agar with bromocresol purple at 0.05%, as a pH indicator in the medium, indicating lactic acid bacteria growth, that is *L. lactis* in our case. Culture plates were brewed at 37°C for 48 h in an incubator (Binder, Austria), then a UFC g<sup>-1</sup> count and a report were done. This, aiming to estimate under which process conditions it is possible to obtain less reduction of the microorganism's Log cycles.

**Stability of the symbiotic dried through spray-drying to *in vitro* gastrointestinal conditions:** An *in vitro* gastric model was created as the one reported by Dimitrellou *et al.*<sup>17</sup> in order to evaluate the spray-drying effect on the micro-organism's capacity for surviving the joint action of acid conditions and protease and bile salts the bacteria would tolerate after ingestion.

A simulated gastric solution containing sodium chloride (NaCl) (Merck, Germany) was made dissolving pepsin (Panreac Applichem, Spain) (3 g L<sup>-1</sup>) in the solution (5 g L<sup>-1</sup> NaCl) and the pH was adjusted to 2 with

HCl (Merck, Germany) 5 mM. After filtering gastric solutions through a syringe filter of 0.22 µm, samples of 0.1 g from the encapsulated and 0.1 mL from free cells suspension were separately added to 9.9 mL of the gastric solution and incubated at 37°C. After 0, 0.5, 1, 2 and 3 h of the process, aliquots were taken and spread over BHI agar to determine the micro-organism's survival<sup>17</sup>.

Tolerance to bile salts was carried out with the same above described process for low pH resistance of the gastric solution, but with 9.9 mL of salts solution having 10 g L<sup>-1</sup> bile salts. Samples at 0, 3 and 6 h of incubation were taken and the micro-organism's survival was evaluated both free and encapsulated after the BHI agar spreading<sup>17</sup>.

**Experimental design:** In order to determine if data are normal, a normality test of the results was carried out. Afterwards, a sequential experimental design was done. In the first phase (Yacon's prebiotic effect) a completely randomized design was applied. In the second phase (spray-drying effect) a factorial design with two factors: Process entrance temperature and maltodextrin concentration plus three response variables (moisture content, water activity and micro-organism's survival). Then in the third phase (stability in *in vitro* gastrointestinal conditions) a completely randomized design was applied with the micro-organism's survival as the only variable of response. The results were analyzed through variance analysis at a significance level of 95% and a variance homogeneity test was made to carry out the media comparison employing Tukey (homogeneous variances) or Dunnet (non homogenous variances) using the statistical software Minitab® version 16.

## RESULTS AND DISCUSSION

### Testing Yacon's prebiotic effect on the micro-organism:

Due to its high fructo-oligosaccharides content, Yacon has important prebiotic features. When resisting digestion in the upper gastrointestinal tract, these carbohydrates are hydrolyzed and fermented by intestinal bacteria such as the ones from the *Lactobacillus* and *Bifidobacterium* genus<sup>22</sup>. However, so far, it has not been possible to find studies proving the effect of this root on the increase of *L. lactis*, a lactic acid bacteria with high probiotic potential.

In order to test Yacon's prebiotic effect, the *L. lactis* growth kinetics in enriched Yacon's nectar was compared with the results from the BHI (control) broth, then data was modeled using the DmFit de ComBase® (www.combase.com) tool. It was found that the growth model to present a greater

adjustment ( $R^2 = 0.986$ ) was the one from Baranyi and Roberts<sup>23</sup> (No lag) with a maximum growth value of  $7.784 \pm 0.0826$  Log CFU mL<sup>-1</sup> and a growth rate ( $\mu_{max}$ ) of  $0.79 \pm 0.0826$  h<sup>-1</sup>, whereas, in the BHI broth, the full Baranyi and Roberts<sup>23</sup> model, that is, with an adaptation phase (Lag), showed better results with a  $R^2$  of 0.999, a maximum value of  $8.662 \pm 0.014$  Log CFU mL<sup>-1</sup> and a rate of  $0.422 \pm 0.0169$  h<sup>-1</sup>. It is to highlight that, although biomass production was higher in the controlled medium, *L. lactis* had an adaptation phase of  $1.799 \pm 0.107$  h, which is why the growth rate was 87% higher in Yacon's nectar. This indicates that nutrients from the root were metabolized by the LAB and FOS could be a more favorable carbon source for this bacteria than the glucose found in the commercial BHI broth.

Yacon's capacity to ferment fructo-oligosaccharides has been proven by different probiotic strains such as *Lactobacillus acidophilus* NRRL-1910, *Lactobacillus plantarum* NRRL B-4496 and *Bifidobacterium bifidum* ATCC 15696<sup>24</sup>. It was also proven that with Yacon's extract in *L. plantarum*, populations higher than  $10^9$  CFU mL<sup>-1</sup> were obtained, whereas, in MRS broth<sup>25</sup> they were  $10^8$  CFU mL<sup>-1</sup>.

Some other studies developed by Campos *et al.*<sup>26</sup> found that, when feeding guinea pigs with Yacon's flour, bifidobacterium and lactobacillus' concentrations were significantly higher ( $p < 0.05$ ) compared to the controlled medium.

Besides, Yacon's nectar was found not to be an appropriate medium for *Escherichia coli* growth getting viable cells' recounts below 2.4 Log CFU mL<sup>-1</sup> from the moment of inoculation until 10 h of the study. This behavior has been observed by some other authors such as Vegas *et al.*<sup>25</sup>, who found that the *E. coli* population was steady for 16 h and then started its death phase, which could have been due to this enterobacteria capacity to produce  $\beta$ -fructosidase and/or  $\beta$ -fructofuranosidase; enzymes in charge of hydrolyzing FOS<sup>27</sup>.

Based on the previous, it can be inferred that Yacon is a potential prebiotic source where *L. lactis* ferments FOS in this raw material producing lactic acid and other compounds like nisin. This is why the symbiotic combination of this root and LAB might inhibit pathogens strains' growth as a consequence of the pH reduction and the bacteriocins' antimicrobial effect.

Although results are positive, it is recommended to carry out a deeper research on the type of fructo-oligosaccharides that are consumed by *L. lactis* during its metabolism process since it has been studied depending on their degree of polymerization (DP), LAB consume them more efficiently or not. Yacon's FOS have a lower than 10 DP and main FOS are 1-kestose, nystose and 1-fructofuranosylnystose, each one with a different DP<sup>28</sup>, which is why Pedreschi *et al.*<sup>29</sup> found

that *L. plantarum* and *L. acidophilus* completely used 1-kestose molecules whereas, *B. bifidum* also used molecules with a higher Yacon degree of polymerization<sup>30</sup>. Just the same way, Caicedo<sup>27</sup> evaluated the effect of different FOS concentrations with several DP on the isolated growth of *Lactobacillus* and *Bifidobacterium* strains. Here, a greater preference for FOS consumption with lower DP such as 1-kestose and nystose (DP 2 and 3, respectively) was emphasized, followed by 1-fructofuranosylnystose (DP 4).

**Spray-drying effect:** The spray-drying effect on the *L. lactis* was tested and the resulting powder due to the fact that Yacon is an extremely perishable food and its fructo-oligosaccharides experience significant changes during the post-harvest and also because of the fact that LAB lose their viability and functionality during the storage of the product and its passage through the gastrointestinal tract.

**Moisture content and water activity:** A reduction in the moisture content of  $82.67 \pm 1.142\%$  in the product was obtained during the spray-drying process. As observed in Fig. 1, this content varied from 6.0 to 8.943% in the powder and was significantly affected by process entrance temperature and the coating material concentration, being 130°C and 20% maltodextrin the treatment with the lowest value. The results obtained showed that the higher the entrance temperature, the lower the moisture content is and vice-versa, which is due to the fact that as the temperature increases, there is a higher evaporation of the water in the product<sup>31</sup>. On the other hand, maltodextrin's concentration affected the moisture content (MC) in the powder, so that dried products with a 30% maltodextrin presented a higher MC than those with a 20%, which could have been due to the fact that increasing the material concentration, a faster wall formation is eased, restricting water steam diffusion from the inside of the liquid product to the surface during the process<sup>32</sup>.

Concerning water activity, a reduction of 0.74 was obtained through spray-drying finding values of  $0.24 \pm 0.03$  in the powder with no statistically significant differences ( $p > 0.05$ ) between the tested/evaluated treatments.

These results are similar to the ones reported by Dimitrellou *et al.*<sup>17</sup> ( $0.13 \pm 0.0025$ ), Liu *et al.*<sup>14</sup> ( $0.19 \pm 0.1$ ) and Barbosa *et al.*<sup>33</sup> ( $0.42 \pm 0.08$ ) who used the spray-drying method potentially probiotic bacteria in different matrix. The latter can be attributed to the high temperatures in the process entrance, thus leading to a low water activity which is necessary for commercially viable powders production with good management characteristics (low stickiness and agglomeration) and reduction of enzymatic reactions'

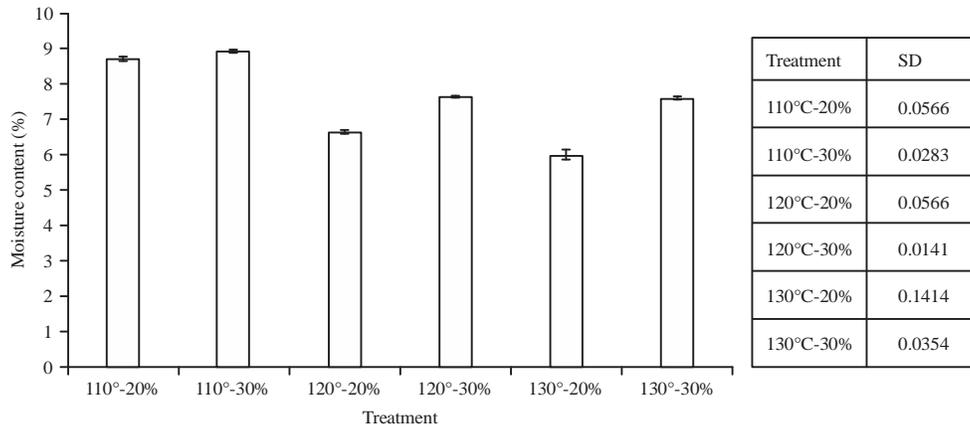


Fig. 1: Spray-dried products' moisture content in different temperature conditions and maltodextrin concentration

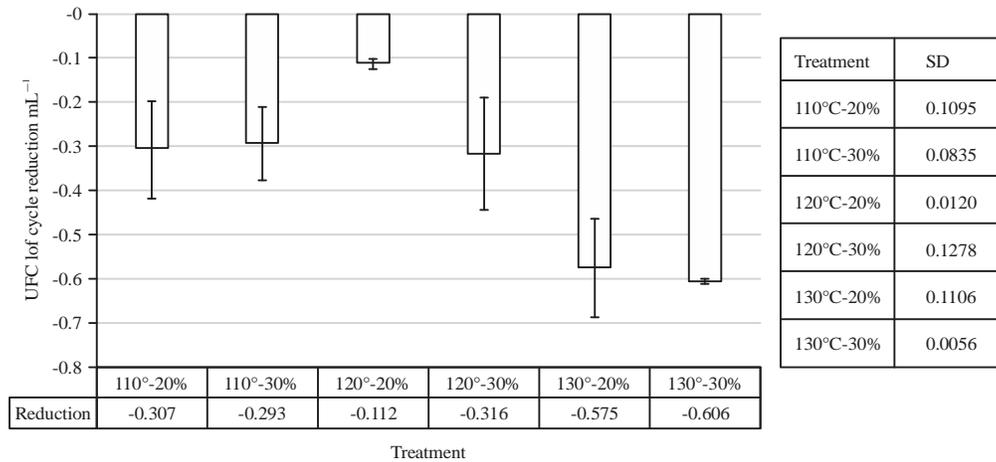


Fig. 2: Reduction of viable cells in the spray-drying process at different temperature and maltodextrin concentration conditions

degradation<sup>17</sup>. Besides, as mentioned by Teixeira *et al.*<sup>34</sup>, water activity between 0.11 and 0.23 can prevent cell death during storage, whereas high values are related to an accelerated mortality of probiotics<sup>14</sup>. This is why, in spray-drying processes at an industrial level, obtained values are around 0.2, improving the micro-organisms viability<sup>33</sup>.

**Lactococcus lactis survival:** Due to the fact that probiotics have viability problems, spray-drying could give sensitive cells protection during food processing and storage and ensure their delivery in the gastrointestinal tract. This is why the effect of process conditions on the *L. lactis* in an initial population of  $8.526 \pm 0.072$  Log CFU mL<sup>-1</sup> was tested. As shown in Fig. 2, although statistically significant differences were found ( $p < 0.05$ ), treatments 1, 2 and 4 were statistically the same, just like 5 and 6, being treatment 3 (120°C and 20% maltodextrin) the one to show less reduction in the viable cells.

The effect of temperature higher applied (130°C), showed the higher biomass reduction. This effect has been already reported by some other authors in *Lactobacillus reuteri* using 90-180°C temperatures<sup>35</sup>, which could have been due to the fact that cells experienced high temperature stress, dehydration and osmotic stress, leading to a metabolic loss and in some cases cell death because of the membrane damage, ribosomes and genetic material<sup>14</sup>.

Nonetheless, it is important to point out that through spray-drying under the tested conditions, 92-98% of the viable cell population could be maintained, which is similar to what Barbosa *et al.*<sup>33</sup> reported. As they also found that the number of *L. plantarum* and *P. acidilactici* cells didn't decrease during the spray-drying process and Rajam and Anandharamakrishna<sup>32</sup> kept the 70.77-72.82% of *L. plantarum* using FOS as coating material.

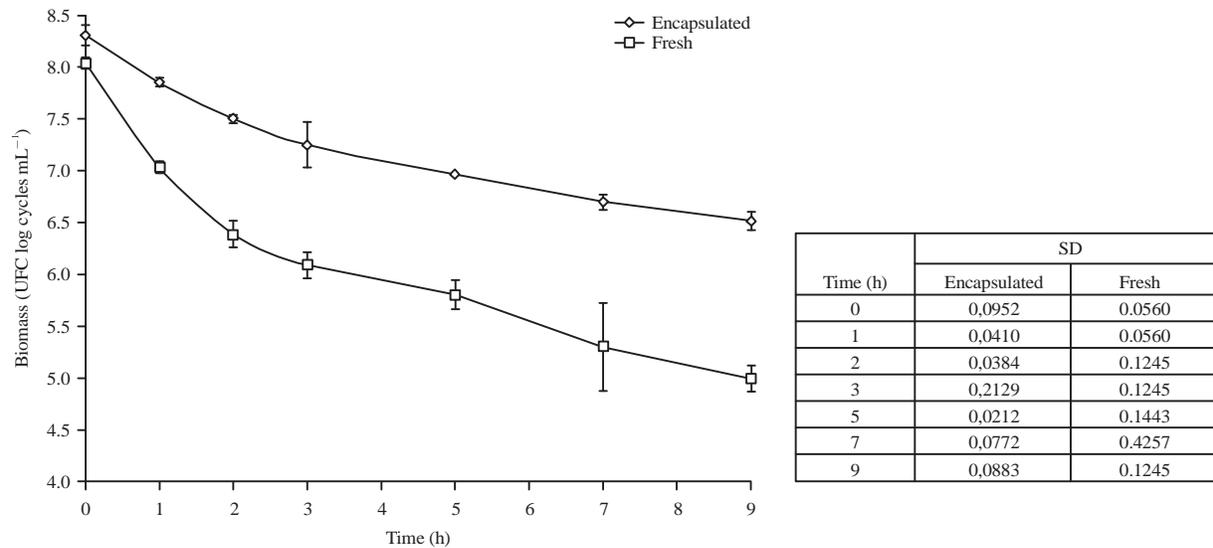


Fig. 3: *Lactococcus lactis* kinetics of biomass reduction under simulated gastrointestinal conditions. First 3 h, stomach phase; following 6 h, intestinal phase

The previous is possibly caused by the use of protectives and prebiotics, which is Yacon in our case<sup>14</sup>. Then, as mentioned by Kingwatee *et al.*<sup>31</sup>, by adding inulin to the encapsulating agent (maltodextrin or arabic gum) it was possible to find better results in cells' survival up until 2 Log CFU g<sup>-1</sup>. Another factor that might have caused this fact was that during atomization, drops are in touch with hot air which results in water evaporation and these get rapidly cold until they reach an inner temperature of 40-50°C, when the process out temperature is lower than 80°C, contributing to cell viability to be maintained<sup>32</sup>. Besides, low water activity found in this study, might have helped *L. lactis* survival just as it was previously stated.

Looking for a better cell survival rate, different methods have been developed and tested that have been proven to improve the spray-drying effect, among which the soft heat treatment before the process can be found. Previous studies carried out by Liu *et al.*<sup>35</sup> have proven that some strains like *L. reuteri* become thermotolerant after a moderate thermal shock, which causes thermal stress in the bacteria and leads to a higher survival in the drying process. Usually, the shock may be at a 10°C temperature above the optimal growth temperature, which can increase up until 16 times the rate of LAB survival during and after the spray-drying process<sup>34,36,37</sup> quoted by Anekella and Orsar<sup>38</sup>. Such a rise has been explained by other authors indicating that sub-lethal stress induces protective mechanisms as an alteration or reprogramming of the metabolic pathways to adjust to the

new environment. Besides, it improves survival in extreme drying conditions and increases viability during storage<sup>39-41</sup> quoted by Anekella and Orsar<sup>38</sup>.

**Stability to *in vitro* gastrointestinal conditions:** Aside the fact that probiotic cell's viability must be kept throughout the food's life cycle, after intake, these also need to survive passage through the gastrointestinal tract, tolerating acidity in the stomach, toxicity of bile salts and resisting enzymatic activity to be able to promote beneficial effects on health<sup>42</sup>.

Taking into account that the best results in *L. lactis* survival using the spray-drying process were obtained at 120°C entrance temperature and 20% coating material. The stability under simulated gastrointestinal conditions of the resulting powder under these parameters was tested. As shown in Fig. 3, in the stomach phase (first 3 h) at pH 2 adding pepsin (3 g L<sup>-1</sup>), a reduction of 2 Log cycles CFU mL<sup>-1</sup> in the free cells was observed, whereas in the dry product of only 1 Log cycle CFU mL<sup>-1</sup> proved spray-drying gives protection to *L. lactis* against the denaturation of membrane proteins because of the low pH and pepsin protease. These results coincide with previous ones carried out by Dimitrellou *et al.*<sup>17</sup>, where encapsulation generated a protective barrier going from a *L. casei* reduction of 4.03 to 1.63 Log cycles UFC mL<sup>-1</sup>; in the same way, De Souza Leone *et al.*<sup>42</sup> tested the percentage of surviving cells under simulated conditions of the human stomach, getting positive results for *L. casei* cells adhered to Yacon compared to the ones set free in the medium.

A similar behavior was observed in the intestinal phase with bile salts and trypsin. It was mainly found that the reduction in free cells was of 1 Log cycle CFU mL<sup>-1</sup>, whereas, in dry LAB it was only 0.75 Log cycles CFU mL<sup>-1</sup>. This effect has been reported by other authors such as De Souza Leone *et al.*<sup>42</sup>, who perceived that more than 80% of Yacon encapsulated cells survived the gut's conditions to be able to act as probiotics. Also, Dimitrellou *et al.*<sup>17</sup> found that spray-drying had a positively significant effect in cells since they only observed a reduction of 1.63 Log cycles CFU g<sup>-1</sup>, whereas, in the free cells it was of 3.05. Nevertheless, this cells' susceptibility to the intestinal phase may be due to the fact that bile has the ability to affect phospholipids and membrane proteins. Hence, interrupting cell homeostasis and altering the stability of the molecule<sup>43</sup>.

Bacteria survival may be attributed to the cells' strong adhesion to the encapsulating agent, in this case maltodextrin, that protects them from the acid and high biliary conditions. Besides, it's been reported that maltodextrin also serves as a moderate prebiotic together with Yacon's FOS acting as probiotic bacteria protectives<sup>38</sup>. The former has been evidenced in previous studies where the addition of inulin to maltodextrin allowed the *L. casei* survival in gastric juice and bile fluids, improving the impact of these conditions in the bacteria growth compared to the free cells<sup>31</sup>. Another factor that may have caused *L. lactis* survival was the thermal stress produced in the spray-drying process since several researches reveal that, when a microorganism is exposed to previous tension such as cold, heat or acid, surviving cells may tolerate better in a subsequent unfavorable environment like the adverse conditions of the gastrointestinal tract; this type of protection is known as cross protection stress response<sup>43</sup>.

Due to all of the above and taking into account how the WHO defines probiotic food, it can be said that the powder product obtained through spray-drying complies with the requirement of containing at least 10<sup>6</sup> microorganisms g<sup>-1</sup> or mL when it is consumed and survives the passage through the gastrointestinal tract under simulated *in vitro* conditions.

## CONCLUSION

Based on the obtained results, it may be inferred that Yacon has a great potential to become a profitable product for producing countries since a high content of fructooligosaccharides in its fresh root and in the powdered product can be found. This makes it possible to classify it as a promissory prebiotic food. Spray-drying Yacon adding potential probiotic bacteria such as *Lactococcus lactis* had a significant positive effect on the product's stability and the

bacteria's survival. This is why it might be considered a symbiotic powdered functional food with possible beneficial effects on health at the same time it makes it a perfect alternative to mitigate the nutritional predicament of today's world.

## SIGNIFICANCE STATEMENT

In the present investigation it was found that Yacon is a promising Andean resource, due to its prebiotic activity, thanks to its content of fructooligosaccharides. In addition, the spray drying process generated protection for the symbiotic, showing survival of lactic acid bacteria in simulated gastrointestinal conditions. The main findings of the study were the modeling the growth of lactic acid bacterium *L. lactis* both in a commercial medium and in Yacon nectar; the development of a powder symbiotic product between Yacon and *L. lactis*; and the generation of new processing alternatives for an underutilized Andean root.

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