Viability study of probiotic microorganisms in a symbiotic food made in an Dining Services

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Introduction

There are various investigations to assess the benefits of functional food on people. A study conducted in Sweden with 262 employees who consumed capsules daily with probiotics for three months, in amount of 10^8 CFU *Lactobacillus reuteri*. They showed significant decrease of work absences due to diseases such as intestinal infections and flu [1]. However, few studies have been conducted with functional foods specifically with the workers of a company in order to reduce absenteeism due to illness and increase the productivity of the company. Therefore it was decided to develop an inter-agency project funded by Secretaría de Economía y el Consejo Nacional de Ciencia y Tecnología (CONACyT) whose objective was to reduce labor workers of a company in the electronic industry absences due to the incidence of gastrointestinal illnesses, through the daily consumption of functional foods prepared by the Dining Services of the aforementioned company. The objective of this work was the development of symbiotic food and validation of the feasibility of probiotic microorganisms in food during storage.

Methodology

Physico-chemical and sensory characterization of food:
The parameters analyzed were acidity, pH, viscosity, osmotic pressure, soluble solids (° Brix), texture. Also characterized the inoculum of the symbiotic (Vector) by measuring pH and viscosity, to determine whether it would be necessary to adjust it to make it compatible with food. Symbiotic food measured the color, the gel strength ° Bloom, rupture and fragility as well as organoleptic aspects through a Duo-trio test.

Evaluation of the symbiotic food manufacturing process to ensure the survival of microorganisms during manufacture
During the preparation of gelatin, temperature and agitation speed were monitored to determine the values of these parameters to ensure the survival of microorganisms added through sampling and analysis of the process of elaboration in the Dining Services to replicate the process in pilot plant and non-Vector.

Evaluation of the growth of probiotics in the vector: b The objective of this stage was to find the initial conditions of pH and combination of substrates (glucose and type of fructan) most suitable for the production of biomass that would achieve a Vector that was reached when mixed with symbiotic food at the time of consumption final concentrations greater than 10^{-6-9} CFU/g (Kun Lee, 2009) [2]. Were tested initial pH value of 3.5, 4.5, 5.5 and 6.5.

Process of preparation of the food.
First proceeded to manufacture gelatin on a scale laboratory, with the modified process. I was then held at pilot scale to determine operating parameters, sequence and quality limits and finally climbed into the dining industry.

Stability of survival in the symbiotic food during storage
In this phase of the project is the study of kinetics of fermentation in the matrix of the functional food symbiotic, adding the *l. rhamnosus* HN001 and *B. animalis Bi07,* having as response variables the microorganisms count in plate, physicochemical analysis (pH, acidity, º Brix, viscosity), texture analysis and sensory.

Results and discussion:

Results of physico-chemical analysis
Table 1 shows the results obtained and as you can see, the acidity values below 0.38%,indicando this parameter was not affected by the presence of microorganisms inoculated. Also the degrees Brix of the jellies with vector showed significant differences compared with the control without inoculation, due to the composition of the used vectors. On the other hand, the pH of the inoculated gelatine showed an increase of 0.04 to the pH of the food without inoculum.

Table 1. Characterization of physical chemistry of the symbiotic food with and without inoculum.

Food	pH	° Brix	Color			Strength Gel		Rupture	Fragility
			L	to	b	4° C	10° C		
Without inoculum	3.6	18.79	30.65	4.35	1.5	99.961 +/-2.823	77.708 +/- 1256	455.600 +/- 7.778	12.117 +/-0.170
B. animalis/l. rhamnosus	3.53	20.39	29.9	4.7	1.1	92.078 +/-1,116	79.01 +/- 1.883	477.750 +/- 31.466	12.369 +/-0.057

n = 2

As for the color, it turned out to be a differentiation between the formulations with and without inoculum. However, in the intensity of the color, cannot be said that this parameter present a significant trend.

The measurement, at 4° C and 10° C, in gelatin gel strength control and the one with the combination of microorganisms *B. animalis/L. rhamnosus* (B-Rh), shows us that there is a decrease in strength (° Bloom) measured at 10 ° C in respect of 4 ° C. This was mainly due to the increase in entropy of the system, and thus to molecular mobility of the polymer chains of the gelatin, which decreased its stability and increased its fluidity [3]. In the results of rupture of gel, and fragility of the gel, observed the same pattern previously

analyzed in the gel strength. Therefore, addition of the mixture of probiotic used concentrations does not affect three parameters with and without inoculum.
Sensory tests, DUO-TRIO, with a panel of 12 judges demonstrates adding the combination *B. animalis/L. rhamnosus* product, does not affect significantly ($p < 0.05$) s the sensory characteristics of the gelatin

Design of production at industrial level, parameters of operation, sequence and quality limits.
The scaling of the incorporation of the vectors to the gelatin in the Dining Services was achieved without any difficulty, taking care that temperature before the dosage was at least at 40 ° C, the proper incorporation of the Vector to the food during its preparation, maintained viable microorganisms.

Evaluation of the growth of probiotics in the Vector:
Table 2 shows the factors time, pH and substrate proportions, were statistically significant ($P < 0.05$). The best production of biomass for both probiotic strains, was statistically higher ($P < 0.05$) at pH 6.5, regardless of the proportion of glucosa:fructanos syrup. For ECA of *Lactobacillus rhamnosus,* substrate ratios, increased production of biomass produced were 100:0 and 75:25, however, for *B. animalis*, best condition was only 75:25 ($P < 0.05$), therefore, was recommended to use the ratio 75:25 (glucosa:fructanos) to obtain the vectors. The time in which reaches the greater amount of biomass is less for *l. rhamnosus* (18 h) not so for *B. animalis* that was until 24 h

Table 2. Levels of the factors that influence the growth of probiotic strains in a significant way (P≤0. 05).

Probiotic strain	pH	Time (h)	Glucosa:Fructanos
B. animalis BI07	6.5	24	5
L. rhamnosus HN001	6.5	18	5 and 6

* Treatment refers to the proportions of the substrates 1 = 0: 0, 2 = 0:100, 3 = 25:75, 3 = 25:75, 4 = 50:50, 5 = 75:25 and 6 = 100:0

Stability of survival in the symbiotic food during storage:
The number of microorganisms in the vector for *Lactobacillus rhamnosus* HN001 and *Bifidobacterium animalis* Bi-07, was $9.0x10^{12}$ UFC. This integrated matrix of food concentration allowed a concentration of $5.5x10^{8}$ UFC/g. functional food. Table 3 shows the result of this count, 3 different times 0, 12 to 24 hours, and shown that *l. rhamnosus/B. animalis* had counts in the expected concentration. *L. rhamnosus* remained around 8 log CFU/g for storage, while *B. animalis* around 6 log CFU/g.

Table 3. micro-organisms account *B. animalis Bi-07* and *l. rhamnosus HN001* at time 0, 12 and 24 in water jelly .

Time (h)	*L rhamnosus HN001*	*B. animalis*
0	$1.5x10^{8}$ CFU/g	$1.0x10^{6}$ CFU/g
12	$1.3x10^{8}$ CFU/g	$2.0x10^{6}$ CFU/g
24	$2.0x10^{8}$ CFU/g	$2.0x10^{6}$ CFU/g

Shows that the *L. rhamnosus* was the organism that had a greater growth in this food, obtaining concentrations between 7-8 log CFU/g

Conclusions

Water jelly turned out to be an adequate array to maintain viable, as well as versatility of flavors, such as functional food for Diners of the dining room industrial, applied a probiotic blend probiotic micro-organisms of *l. rhamnosus* HN001 /*B. animalis* Bi07, in a vector with Prebiotics.

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