## A mathematical model to describe the effect of temperature, reaction time and cell concentration on galactooligosaccharids production by *Bifidobacterium infantis*

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Introduction

*Bifidobacteria* have been used in the development of a great range of dairy products. Some strains of *B. infantis* produce galacto-oligosaccharides and this property is suitable for the production of symbiotic products that have a dual characteristic, a probiotic and prebiotic product (Gibson and Roberfroid, 1995). This strain has a high capacity of producing galactooliogosaccharides at high temperatures (Lamoureaux, 2000). These sugars has been studied for their physiological roles in the human intestinal tract like bifidogenic factors (Sako et al., 1999).

Processing of microbial systems for the production of functional foods, is dependant of variables like the concentration and composition of susbtrates, microorganism used, concentration of cells and environmental conditions like pH, temperature, ionic strengh or oxygen disolved. The study of each condition alone don't let to know the influence of one in another. The use of mathematical models its a way to optimize these variables and improve productiviy and to understand how one variable affect the others. The aim of this work was to define if the used conditions for the production of Galactooligosaccharides (GOS), temperature, reaction time and cell concentration are possible to improve.

## Material and Methods

An overnigth culture *of Bifidobacterium infantis* ATCC 17930 was used as the starting cells for GOS production experiments in Skimmed Milk 10% (w/w) that was autoclaved for 10 min at 110°C.

Table 1 show the cell concentrations, eaction time and temperature used. Batch fermentations to evaluate GOS production were performed in flasks of 250 ml, containing 100 ml skimmed milk. Samples were taken at the initial and final time. All the runs were performed a the same time in a rotatory growth chamber with an agitation of 100 rpm. Determination of cell concentration and cell survival in batch was done using Columbia agar base medium. Plates incubated at 37°C for 48 hr under anaerobic conditions. GOS, sugars and organic acids were determined by HPLC on a Waters chromatograph (Milford, Mass, USA) using a Waters 410 RI detector a UV detector and a Ion-300 column (Transgenomic, Omaha, USA) mainteined a 37°C. The mobile phase was  $0.02 \text{ N H}_2\text{SO}_4$  with a flow rate of 0.4 ml min<sup>-1</sup>.

## Experimental design and statistical analysis

Modeling of production was done by use of a response surface analysis. Table 1 show the modeling used. The values used for the response surface analysis taking into account temperature (T), reaction time (Rt) and  $\log_{10}$  cell concentration (C). In all cases the response was GOS% obtained after each batch. The experimental data were analysed by Statgraphics software 6.0 (Statistical Graphics Co.)

## Results

14 variables combinations and four central points were studied by duplicate. The statistical and ANOVA analysis shown that variables temperature and cell concentration were significants and not Rt. Neither of the interaction were significant and the production response curve for this experiment is given by the equation

 $Y=0.2071+T(50)+C(8.9^{-19})+T^{2}(-0.115891)+C^{2}(-0.1158)$ 

Resolving the equation, the theoretical production is 0.34% of Galacto-oligosaccharides and the production in batch was 0.37% (Fig 1)

Table 1. Modeling of the experimental design for GOS production					
	Code and level				
variable	- á	-1	0	1	+ á
T (°C)	36.9	40	50	60	63.1
Rt (h)	3.38	4	6	8	8.62
C ( $\log_{10}$ ufc ml <sup>-1</sup> )	6	7	8	9	10

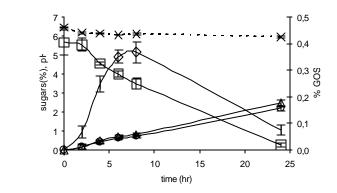


Fig 1. Growth and production kinetic of *B. infantis* in skimmed milk. ( )lactose, (Δ)glucose, (ο)galactose (.)GOS and (.)pH