GROWTH KINETICS OF SEVERAL LACTIC BACTERIA USEFUL AS STARTER FOR EWE'S CHEESE PRODUCTION

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SUMMARY

The growth kinetics in batch culture of 6 strains, potentially useful as starter cultures for ewe's cheese-making, has been analysed. Under the experimental conditions used a high conversion of carbohydrates into lactic acid was achieved. The production of lactic acid can be used as an indirect biomass measurement. Two mathematical models (Logistic and Gompertz's) have been used to describe the growth of these strains in batch culture. Both models fit well to the experimental results, however the Gompertz's model describes slightly better the growth kinetics.

INTRODUCTION

The use of starter cultures in different food manufacturing processes requires the optimization of their production at the industrial level. The assessment of the growth kinetics of microorganisms used as starter is a required tool in order to optimize their industrial production.

To date most of the reported kinetic studies were done in continuous culture (Ishizaki et al., 1990). It is not so difficult to identify the mathematical parameters for the steady state of the continuous culture, however, these kind of studies in batch cultures (Rogers et al. 1978, Zwietering et al., 1990, Ishizaki et al., 1990, 1991) are less frequent although this technique is still the most common practice for industrial applications.

In this paper, the growth kinetics in batch culture of 6 different strains of lactic bacteria is reported. These strains can be used as starter cultures for the production of pasteurized milk manufactured ewe's cheese. Two sigmoidal mathematical models have been used: Logistic (Jason, 1983) and Gompertz (Bratchell et al., 1989) modified by Zwietering et al. (1990). These models describe the biomass evolution without taking into account the substrate consumption rate, since it is assumed that the carbohydrate concentration remains high enough to achieve the maximum number of microorganisms during the culture time.
MATERIALS AND METHODS

Bacterial strains. Bacteria belonging to the Lactococcus, Lactobacillus and Leuconostoc genera were used. These microorganisms were isolated during the different manufacturing steps of the industrial production of ewe's cheese. The strains were identified as follows: Lactococcus lactis subsp. cremoris (GB-1), Lactococcus lactis subsp. lactis (GB-2), Lactococcus lactis subsp. diacetylactis (GB-3), Lactococcus lactis subsp. diacetylactis (GB-4), Leuconostoc lactis (GB-5) and Lactobacillus casei subsp. casei (GB-6).

All bacteria were grown at the same culture conditions in a Bioflo III laboratory fermenter (New Brunswick Scientific, Edison NJ, USA) using a working volume of 1.5 l. The culture media were MRS (de Man et al., 1960) for Lactobacilli and Leuconostocs and MI7 (Terzaghi and Sandine, 1975) for Lactococci. Inocula consisted of 5% (v/v) of cultures grown for 18 h. Not all the strains had the same density at that time of the culture. In all cases, temperature (30°C), pH (6.5) and agitation speed (50 rpm) were controlled automatically.

Growth assessment. Cell growth was estimated by measuring the absorbance of cultures at 560 nm in a Hitachi U-2000 (Tokyo, Japan) spectrophotometer. Previously, calibration equations of optical density versus cell number were obtained.

Analytical methods. Total carbohydrates in the culture media was estimated by the anthrone method (Clegg, 1956). Lactic acid production was estimated from the volume of 5 N NaOH (97 %) to maintain constant pH of the culture media at 6.5.

Data processing. The yield (Y) and productivity (P) of each strain were determined using the following expressions:

\[ Y = \frac{100 \times \text{[Lactic acid]}_{\text{measured}}}{\text{[Lactic acid]}_{\text{theoretical}}} \]
\[ P = \frac{\text{mol Lactic acid}}{\text{(mol subs. \times exp.)}} \]

Logistic and Gompertz models were used to estimate the fermentation kinetics of the starter strains. Their mathematical expressions, respectively were:

\[ y = A/[1 + \exp(b - cx)] \]
\[ y = A \exp\{-\exp[b - cx]\} \]

and their modified equations respectively were:

\[ y = A/[1 + \exp[4\mu_m/(\lambda - t/A + 2)] \]
\[ y = A \exp\{-\exp[\mu_m(\lambda - t)/A + 1]\}. \]

RESULTS AND DISCUSSION

Tables 1 and 2, summarize the fermentation data obtained for each strain. Lactococci strains showed high values of yield and productivity in conversion of glucose into lactic acid. Similar yields, or even higher, were obtained by Thomas and Turner (1981) using lactic streptococci. Lactococcus lactis subsp. lactis var. diacetylactis (GB-4) appeared as the strain showing the highest acidifying capacity as measured by the total NaOH. The acidifying capacity is very important in the first steps of the cheese manufacturing process, when a fast acidification of milk permits its coagulation and inhibits the growth of the contaminant flora (Law and Sharpe, 1977). Among the strains grown on MRS medium, Lactobacillus casei subsp. casei (GB-6) showed the best acidifying capacity. The yield and productivity values in both media were not comparable, because of the differences in their composition. The productivity levels in biomass terms are shown in Fig. 1.