EVALUATION OF THE ACTIVITY OF LIQUID STARTER CULTURE (M.varians) USING DIFFERENT CARBOHYDRATES

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INTRODUCTION

Micrococci are dominant microflora of typical Bulgarian raw dried sausages and Pasterma (salted and dried noncomminuted meat) during the ripe
For their production are used starter cultures including pure cultures of micrococci. It is known that starters of micrococci and non-pathogenic sta
lococci influence on the quality of fermented products (KNAUF, 1995, CORETTI, 1977, BACUS, 1986). Many factors affect the activity of st
cultures. One of them is the higher concentration of carbohydrates. For production of different kinds raw dried sausages and especially for Pasterma
used higher concentrations of carbohydrates. There is no clear evidence how such concentration influence on the activity of starters of micrococci
objectives of this study were: 1) to examine the activity of liquid starter culture of M.varians K7 at 1,5% concentration of different carbohydrates
2) to determine whether there is the relationship between viable count of the starter culture and its optical density.

MATERIAL AND METHODS

Starter culture: M.varians K7 was isolated from raw dried non-smoking type salami. The strain was maintained in lyophylised form and in medi
(pH 6,5) that contained 10g meat extract powder, 10g protease peptone, 5g NaCl, 10g agar per liter of destilled water.

Experimental design: Basal liquid medium used in all experiments contained 10g meat extract powder, 10g protease peptone, 5g NaCl and 2
K_2HPO_4 per liter of destilled water. Two carbohydrates - glucose and saccharose at final 1,5% (w/v) concentration were added separately for each e
periment. After adjusting the pH at 5,8±0,1 the medium was dispense in 50 ml amounts in flasks.

Preperation of preculture ("mother culture"): The medium with different carbohydrates was inoculated with M.varians K7 using 5ml bacterial s
pension from slant agar culture. The preculture was incubated for 24h at 30°C.

Preparation of liquid culture: 5ml of precultures were transferred into the flasks with medium containing the carbohydrates and incubated for 24
and 72h at 30°C.

Analysis: Viable cells count and pH determination were used to evaluate the activity of precultures and liquid cultures of M.varians K7 under exp
mental conditions. Plate count agar (pH 6,5) was used for M.varians K7 count. Plates were incubated for 48h at 30°C.

The pH was determined by using pH-meter.

The optical density (OD) of the cultures was measured spectrophotometrically at 530nm length wave.

The results for bacterial count, pH and OD were analyzed by Statistical Analysis System. Analysis of variance based on means was used.

RESULTS AND DISCUSSION

The growth of liquid culture of M.varians K7 at 1,5% concentration of glucose and saccharose is shown in Fig.1A. Regarding the effect of the
sugars it can be seen that the count of 24 hours culture with saccharose increased of about ten fold than this of the culture with glucose, although
initual cells number of the both precultures was close ($1,7.10^6$ and $2,9.10^6$ viable cells/cm^3 for the precultures with glucose and saccharose respectively

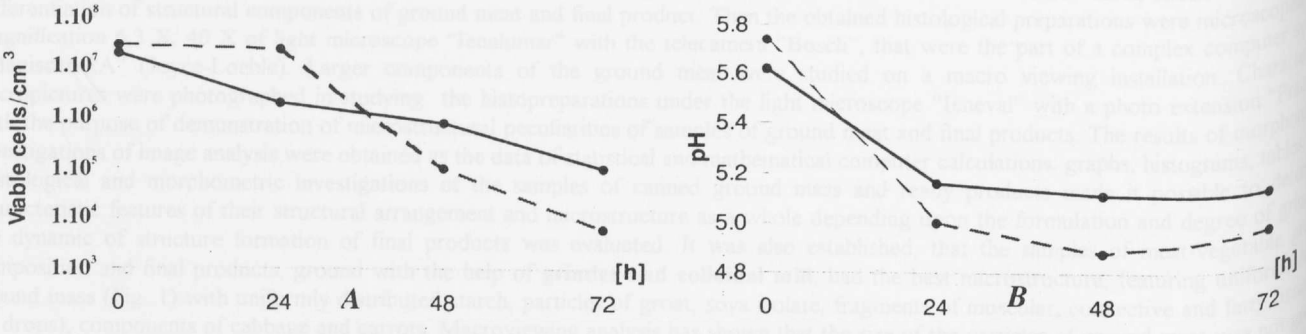

Fig.1. Viable cells count (A) and pH-changes (B) of liquid culture of M.varians K7 at 1,5% concentration of glucose (——) and saccharose (- - -) during 72 hours incubation

Viable number of M.varians K7 after 48 and 72 hours incubation tended to decrease in the both cultures. In the cultures with saccharose the co
declined more and finally reaching a level of $7,1.10^3$ viable cells/cm^3. This fact is probably connected with the increasing of the quantity of metabol
which inhibited the growth of M.varians K7.

The used carbohydrates influenced on the pH values of the liquid culture of M.varians K7 (Fig.1B). Data showed that pH values of the cultures
glucose and saccharose decreased significantly after 24 hours incubation. Although the initial pH values of the both cultures were almost equal (5,6
5,7 for glucose and saccharose respectively), the reduction of pH of 24 hours culture with saccharose was greater than for the culture with gluc
Along the incubation period, the pH values of the culture with glucose remained unchanged, whereas the culture with saccharose showed a slight
creasing after 48 hours and slight increasing after 72 hours incubation (Fig.1B). The results indicated that the pH-changes of 24 hours cultures ca

associated with increasing of the bacterial growth, but after this period the variation of the pH values not dependent on either the cell number of the culture. Our results are close to the findings reported by KUUSELA et al., 1978.

Concerning to the relationship between the viable cells number and optical density of precultures and 24 hours liquid cultures with 1,5% glucose and saccharose was determined non linear correlation.For the precultures of M.varians K7 the following equations were determined:

$$y = \left(1 - 0{,}52\,\frac{x}{\ln x}\right).10^8$$ -for the preculture with glucose

$$y = \left(-0{,}31 - \frac{1{,}26}{\ln x}\right).10^8$$ -for the preculture with saccharose

where y - viable cells count/cm^3;
x - OD value at 530nm length wave.

The relationship between viable cells count and OD of the 24 hours liquid cultures of M.varians K7 with tested sugars is shown in Fig.2 and Fig.3

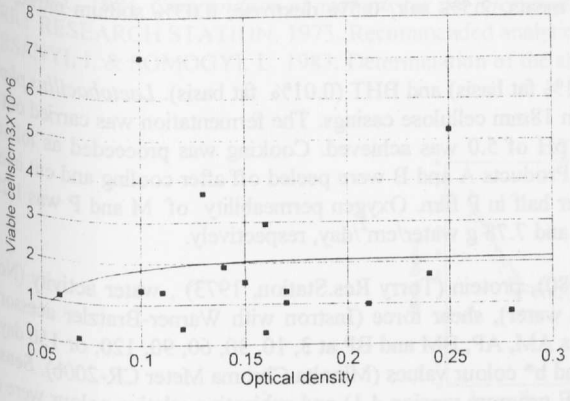

Fig.2. Relationship between viable cells count and OD of 24 hours culture of M.varians K7 with 1,5% glucose

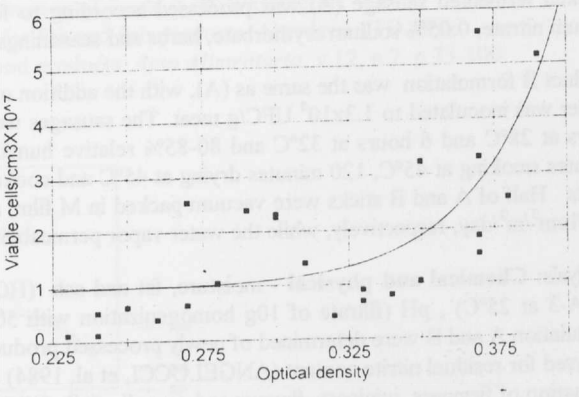

Fig.3. Relationship between viable cells count and OD of 24 hours culture of M.varians K7 with 1,5% saccharose

The following equations were determined:

$$y = \left(2{,}13 + 0{,}001\,\frac{\ln x}{x^2}\right).10^6$$ -for 24 hours culture with glucose

$$y = \left(0{,}95 + 3{,}25.10^6.e^{\frac{x}{0{,}03}}\right).10^7$$ - for 24 hours culture with saccharose

where y - viable cells count/cm^3;
x - OD value at 530nm length wave.

CONCLUSIONS

24 hours liquid starter culture of M.varians K7 grow better and cause greater decrease of pH values when 1,5% concentration of saccharose was used. The obtained relationship between viable cells count and OD values gives rapid evaluation of the activity of M.varians K7 liquid culture for practical use.

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