GROWTH RATE OF BIFIDOBACTERIA IN MILK SUPPLEMENTED WITH YEAST EXTRACT AND WHEAT GERMS

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Abstract

In this study was monitored the growth rate of bifidobacteria in milk supplemented with yeast extract or wheat germs, in order to use the results in the technology of probiotic dairy products . The contribution of these nutrients sources and growth factors to the improvement growth and metabolism activity of bifidobacteria in milk was determined by estimating the number of cells, titratable acidity and pH at different times.
In case of use yeast extract to stimulate the growth of bifidobacteria in milk, the growth rate of bacteria was greater than that of bifidobacteria in milk with wheat germs, but was higher than that of bifidobacteria from milk without ingredient. The population of bifidobacteria ($3.27x\ 10^9$ cfu/cm^3) in the milk enriched with 0.3% yeast extract was 2 times greater than that in the milk with 1.5% wheat germs.
In conclusion, the yeast extract and wheat germs contributed on one side to the enhancement of the growth rate of bifidobacteria and on the other side to the improvement of the sensory attributes of fermented milk containing bifidobacteria.

Key words: bifidobacteria, yeast extract, wheat germs

1. INTRODUCTION

The manufacture of fermented dairy products containing bifidobacteria known as probiotic products has been continuously increasing due to the benefic effects of these bacteria (Desjardins [3]).
Introduction of bifidobacteria into traditional acid dairy products was the best way to promote a positive image of probiotic products for many reasons:

- the fermented dairy products such as yogurt already have a record as being healthful;
- consumers are familiar with the fact that fermented products contain viable microorganisms;
- utilization of probiotic bacteria in the fermentation process makes a connection between them and traditional dairy bacteria;
- the image of yogurt like products as healthful foods facilitates recommendation of daily consumption of probiotics(Heller [6]).

Production fermented milks only by using bifidobacteria is a very difficult thing to do because they are not as easily assimilable as are nitrogen sources which come short in milk (Abu-Taraboush [1], Desjardins [3], Desjardins [4], Proulx [8], Shah [9]). Nitrogen sources such as enzymatic or acid casein hydrolysates, whey powder, whey protein concentrate were studied in order to stimulate the growth of bifidobacteria in milk (Dave [2], Etienne [5], Poupard [7], Shah [9]).
The yeast extract represents a valuable source of nutrients and growth factors, being used in the majority culture medium of bacteria. Through the valuable content of amino acids, the yeast extract may contribute to the growth stimulation of bifidobacteria and also to the improvement of these bacteria viability in dairy products. Also, the calcium pantothenate and biotin represent growth factors for bifidobacteria and are present in the yeast extract in a proportion of 30mg% and 0.25% (Sommer [11], Poupard [7]).
Wheat germs are remarkable for their valuable composition of amino acids, vitamins,

bioelements and polyunsaturated fatty acids with hypocholesterolemic effect (Segal [10]). From all the amino acids present in the wheat germs, cysteine is a growth factor of bifidobacteria which cannot be replaced by methionine, homocysteine or by other amino acids (Poupard [7]).
This study was performed to improve the growth of bifidobacteria used in singular cultures for the fermentation process of milk, by adding yeast extract and wheat germs with a view to turning to good account the results got at the end of the biotechnology of probiotic fermented milks.

2. MATERIALS AND METHODS

During our researches of cultivating bifidobacteria in a medium made from reconstituted milk with 12.4% nonfat dry matter (base medium). The base medium was supplemented with an addition of yeast extract and wheat germs in order to stimulate the multiplication process of cells and their fermentation activity.
In the first part of the experiment consisting of 4 different samples an addition of 0.1, 0.2, 0.3 and 0.4% yeast extract was performed. A base medium without yeast extract was used as a control. The samples were inoculated with 4% frozen-drying culture of bifidobacteria (Bif species 420, Danisco, Germany) used in the shape of suspension in a base medium. After the inoculating process the samples having a modified culture medium and the control sown similarly with bifidobacteria were thermostated at 37^0C. After 8 and 24 hours the multiplication process was evaluated by determining concentration of cells per millilitre.
The second part of the experiment focused on using wheat germs also to stimulate bifidobacterial growth. This nutritive factor was used in a range of 1, 1, 5, and 2%, especially nitrogen sources being easily assimilable on purpose to enlarge the base medium. After inoculating samples with bifidobacteria a thermostation at 37^0C followed.
Sowing those samples including the control 6 and 24 hours later (base medium without wheat germs), concentration of cells was determined using the direct counting method called Breed as well as in the previous variant which supervised the multiplication process of bifidobacteria in a medium containing yeast extract.
In both cases of adding the nutrients to the base medium used for bifidobacterial cultures, the multiplication process and the fermentation activity of cultures were both monitored at the some time by determining their titratable acidity.

3. RESULTS AND DISCUSSION

3.1. Influence of yeast extract on growth and fermentation activity of bifidobacteria

Analyzing our data one could notice that 8 hours after inoculating the samples, the number of the cells is proportional to the dose of yeast extract up to an addition of 0,3% inclusive. In comparison with the control sample that means an increasing number of cells from $2,49x10^8$ to $19,5x10^8$ cfu/ml which is 8 times greater than its own number. One could also notice that an addition of 0,4% yeast extract generates an increasing number of cells which is larger than in the control, but in a smaller proportion than in order used doses. During the incubation from 8 to 24 hours at 37^0C each culture was increased in a different way. This the cell number of the control becomes 5.2 times higher and the one the samples gets 14.6, 15.5 and 17.2 times higher by adding yeast extract in a range of 0.1, 0.2 and 0.3%. Adding a dose of 0.4% yeast extract at the same time, the cell number increases 1.6 times the cell number increases 1.6 times more and this is superior to the one of the control but inferior to other additions of yeast extract. Comparing the concentration of cells in the studied samples, after 24 hours, the highest number of cells was found in the sample using 0.3% yeast extract namely $32.7x\ 10^8$ cfu/cm^3 so it was 1.2 times higher than was in the control. The evolution of cells is also shown in figure 1.

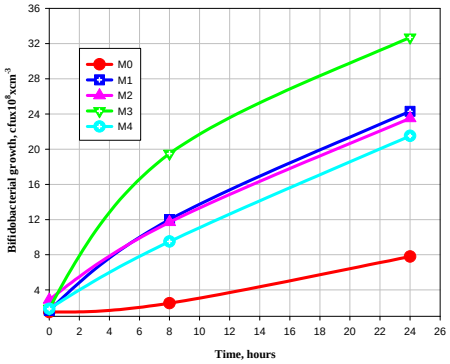

Figure 1. Dynamic of growth of bifidobacteria in milk samples supplemented with yeast extract
M_0=base medium (control sample); M1=Mo+0.1% yeast extract; M2=Mo+0.2% yeast extract; M3=Mo+0.3% yeast extract; M4=Mo+0.4% yeast extract

As for the fermentation activity of bifidobacteria in the studied samples with or without any addition of yeast extract, one could notice a continuous increase of activity during the supervised period but different for each used dose of ingredient. In the control sample a slow increase of activity occurs within 24 hours of incubation and the final reached point is 3.8 times higher than is the first one. Using samples with yeast extract in a range of 0.1, 0.2 and 0.3% the acidity becomes 1.24, 1.4 and 1.46 times higher than does in the control. In addition, the acidity made in 8 hours in sample with 0.3% yeast extract was appropriate with that a control sample made in 24 hours (figure 2).

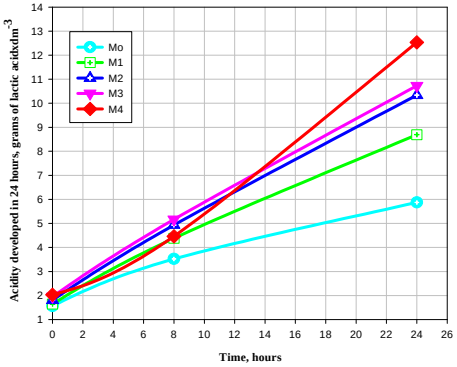

Figure 2. Evolution of acidity expressed in grams of lactic acid/ dm^3 in mediums with yeast extract during 24 hours of incubation

The acidity developed in the studied samples within 24 hours has a variation from 5.87 grams of lactic acid/dm3 in the control to 12.52 grams of lactic acid/ dm^3 in the sample with 0.4% yeast extract. The higher acidity in the sample with 0.4% yeast extract justifies the decreasing number of bifidobacteria in this environment as they are known for not being tolerant to acid. The level of acidity increases where as the level of pH decreases and has a variation from 4.81 in the control to 4.32 in the sample with 0.4% yeast extract (these data are not shown in the figures).

3.2. Influence of wheat germs on growth and fermentation activity of bifidobacteria

Our results showed that wheat germs added in a base medium in a range of 1, 1.5 and 2% had a positive effect on the multiplication process of bifidobacteria. The cell number was larger in the samples with addition of ingredient than in the control (figure 3).

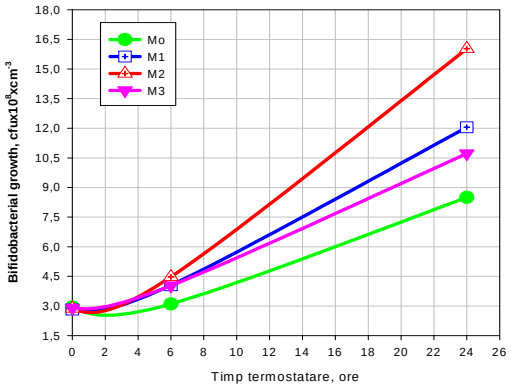

Figure 3. Dynamic of growth of bifidobacteria in mediums with wheat germs: Mo= base medium (control sample); M1=Mo+1% wheat germs; M2=Mo+1.5%wheat germs; M3=Mo+2%wheat germs

After 24 hours of incubation in optimum conditions, bifidobacteria reached a maximum number of cells $16.03x10^8$ cells/dm^3 in a medium with 1.5% wheat germs. Thus, an addition of 1.5% wheat germs in a base medium generates au increasing number of cells 6 hours after inoculation which is 1.52 times larger then in the control and 1.88 times

larger after 24 hours, satisfying results were also obtained in the medium with 1% wheat germs in which the number of bifidobacteria was 1.4 times higher than in the control after 24 hours of incubation.

Showing these data one can notice than the maximum number of bifidobacteria got after 24 hours in mediums with wheat germs is almost as increased as the one in mediums with yeast extract after 8 hours and that proves a more stimulating effect on bifidobacterial growth.

As for the acidity in mediums with wheat germs we mention that it was superior to the one in the control both 8 hours and 24 hours after inoculation. The acidity developed in mediums with 1, 1.5 and 2% wheat germs was higher than in the control and had an overrun of 41.1, 54.13 and 36.65% respectively (figure 4).

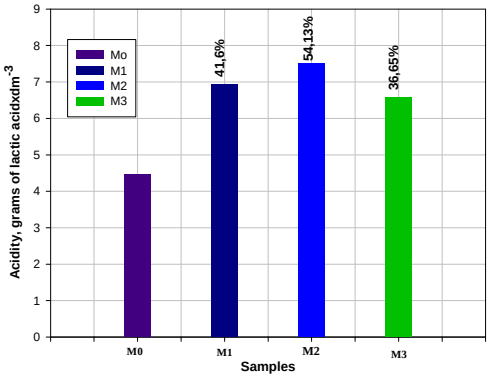

Figure 4. Evolution of acidity in mediums containing wheat germs during the supervised process

We also emphasize the sample containing 1.5% wheat germs after 8 hours or 24 hours of optimum incubation as in this situation the level of acidity is 1.36 and 1.54 higher than in the control. The acidity developed after 24 hours of optimum incubation in mediums containing wheat germs was lower than in samples containing yeast extract and the level of pH wasn't below a 4.5 score.

4. CONCLUSIONS

Yeast extract proves to be a good stimulating product for the growth rate of bifidobacteria. It provides a satisfying cell number ($3.27x10^9$cfu/cm^3) and a suitable acidity having no negative effect on the viability of cells when existing in a dose of 0.3% in a culture medium like milk.

Wheat germs can also be used for stimulating bifidobacteria culture, providing the best results when used in a dose 1.5% but not having a more stimulating effect than yeast extract. Furthermore, through the supplementation of milk with these ingredients (especially wheat germs) are obtained fermented milks with an appetizing taste of pound cake with yoghurt and a pleasant yellowish color.

5. REFERENCES

[1] Abu-Taraboush, M. H. and other (1998). Growth, viability and proteolytic activity of bifidobacteria in whole camel milk. *Journal of Dairy Science*, 81, 354-361.

[2] Dave, R. I. and Shah, N. P. (1998). Ingredient supplementation effects on viability of probiotic bacteria in yogurt. *Journal of Dairy Science*, 81, 2804-2816

[3] Desjardins, M. L. and Roy, D (1990). Growth of Bifidobacteria and Their Enzyme Profile. *Journal of Dairy Science*, 73, 299-307

[4] Desjardins, M. L. and other (1990). Uncoupling of growth and acids production in Bifidobacterium ssp. *Journal of Dairy Science*, 73, 1478-1484.

[5] Etienne, L. and other (1994). Growth promotion of Bifidobacterium animalis by bovine milk proteose-peptone. Lait, 74, 313-323.

[6] Heller, K. J. (2001). Probiotic bacteria in fermented foods: product characteristics and starter organisms. American Journal of Clinical Nutrition, 73(suppl), 374-379 .

[7] Poupard, J.A., Husain, I., Norris, R.F. (1973). Biology of Bifidobacteria. Bacteriological Reviews, 37(2), 136.

[8] Proulx, M. et al (1992). Utilisation d'hydrolysats enzymatiques de caséine pour la croissance des bifidobactéries. Lait, 72, 393-404

[9] Shah, N. P. (2000). Probiotic bacteria: selective enumeration and survival in dairy foods. *Journal of Dairy Science*, 83, 894-907.

[10] Segal, B. et al. (1991). Tehnologia produselor alimentare de protecție, Editura Ceres, Bucureşti, pp. 111-116

[11] Sommer, R. (1996). Yeast extract: Production, Properties and Components. 9th International Symposium of Yeast, Sydney