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EFFECTS OF MILK FERMENTED WITH LACTOBACILLUS ACIDOPHILLUS AND BIFIDOBACTERIUM ON YOGHURT QUALITY AND GLUCOSE CONTENT

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*The aim of this research are comparing the yoghurt quality and the glucose content, for the effect to the consumers. The samples are the yoghurt fermented with *Lactobacillus bulgaricus* and *Streptococcus thermophilus* compared to the yoghurt fermented with *Lactobacillus acidophilus* and *Bifidobacterium*. Results indicated that the quality of both yoghurt has no significancy, but the yoghurt fermented with *Lactobacillus acidophilus* and *Bifidobacterium*, has lower glucose content; but still in the normal range than the *Lactobacillus bulgaricus* and *Streptococcus thermophilus* fermented yoghurt.*

Key words: yoghurt, *Bifidobacterium*, *Lactobacillus acidophilus*, *Lactobacillus bulgaricus*, *Streptococcus thermophilus*.

Industrial microbiologists have extended the term fermentation to describe any process for the production of product by the mass culture of a micro-organism.

Contamination during milking and milk handling will result in a large variety of micro-organisms starting to develop after milk has been drawn. But in most cases lactic acid bacteria (LAB) will become predominant by overgrowing the other bacteria. The acid produced is generally not considered unpleasant by the consumer and because it also suppresses pathogenic as well as proteolytic and lipolytic bacteria it actually helps to preserve milk. From a comparison of the various fermented liquid milk products produced in the world, all products can be classified in a few groups. The principal differences between these groups depend on: the type of milk used, the type of fermenting flora, and the way the milk is processed either before or after fermentation. The flora that is found in indigenous fermented milk products largely depends on the ambient temperature and the character of the contaminating bacteria. Some LAB are mesophilic, others are thermophilic. The LAB can be characterized by the way they ferment lactose in milk after this disaccharide has been transformed into two monosaccharides, viz. glucose and galactose (Stanbury and Whitaker, 1984).

Lactic acid bacteria produce various compounds such as organic acids, diacetyl, hydrogen peroxide, and bacteriocins or bactericidal proteins during lactic acid fermentations (Oyetayo et al, 2003). In many fermented products, the presence of specific micro-organisms other than LAB may determine the character of the product. The *Streptococcus sp.* that homofermentative will produce diacetyl, but the *Lactobacillus acidophilus*, also homofermentative does not producing diacetyl (van den Berg, 1988).

The aim of this work is comparing the effect of using the *Bifidobacterium* and *Lactobacillus acidophilus* as cultures on the yoghurt quality and glucose content in mice blood.

MATERIALS AND METHODS

The milk. Raw milk (cow milk) from farm animals at the faculty.

The bacteria. The bacteria are pure cultivated *Bifidobacterium*, *Lactobacillus acidophilus*, *Lactobacillus bulgaricus* and *Streptococcus thermophilus*. The identification of the cultures was based on the characteristics of the lactobacilli and streptococci as described in Bergey's Manual of Determinative Bacteriology (Holt, et al, 1994).

Yoghurt quality. was tested by preference test and for the glucose content we used mice as animal test. The parameter for yoghurt quality are preference test and pH.

Blood glucose content in mice after three weeks treatment with 1.25% and 2.00 % dosage.

RESULTS AND DISCUSSION

The effect of the treatment on pH yoghurt.

The pH or the hydrogen ion concentration of yoghurt indicates its real acidity. In Table 1, presents the results of the yoghurt pH for each yoghurt (*Lactobacillus bulgaricus* and *Streptococcus thermophilus*, and *Bifidobacterium* and *Lactobacillus acidophilus*).

Table 1. pH yoghurt

No.	Yoghurt starters	pH
1	<i>Lactobacillus bulgaricus</i> and <i>Streptococcus thermophilus</i>	4.01
2	<i>Bifidobacterium</i> and <i>Lactobacillus acidophilus</i>	4.64

Streptococcus thermophilus is homofermentative and is especially found in thermophilic starters and cultures, that are particularly suitable for products culture such as yoghurt, and producing diacetyl. *Lactobacillus bulgaricus* which, together with *Streptococcus thermophilus* is known for its capacity to produce the flavour component acetaldehyde in addition to diacetyl. Greater acid production is obtained when the two organisms are growing together than if grown separately. This is due to the proteolytic action of *Lactobacillus bulgaricus*, whereby valin, which stimulates the growth and acid production of the streptococci, is released. *Streptococcus thermophilus*, however, also produces a stimulant for the growth of lactobacilli; this agent has been proved to be formic acid.

Lactobacillus acidophilus is a slow acid producer in cultures used for the production of yoghurt that has a plain acid flavour, and may be combined with other bacteria to obtain a more palatable product. *Lactobacillus acidophilus* grows very slowly in milk and therefore it is important to protect the milk against contamination. The milk has no real aromatic flavour and is frequently recognized as too sour, so it is sometimes mixed with other bacteria. The reason for using *Bifidobacterium* in place of *Lactobacillus bulgaricus* is that *Bifidobacterium* produces the physiologically active D(+)-lactic acid.

The acidity of the yoghurt using *Bifidobacterium* and *Lactobacillus acidophilus* (pH 4.64), are lower than the yoghurt with *Lactobacillus bulgaricus* and *Streptococcus thermophilus* starters (pH 4.01); because they produced slightly lower of lactic acid and higher acetic acid.

Preference test.

For preference test, we're using 20 well trained tester, for testing the colour, aroma, taste, consistency, and texture. In Table 2, presents the results of the preference test.

Table 2. Preference test

Yoghurt	Consumer Preference					
	Colour	Aroma	Taste	Consistency	Texture	Average
A	3.25	2.70	3.05	3.20	3.15	3.07
B	3.35	3.30	2.80	2.95	3.00	3.08

Notes : A = *Lactobacillus bulgaricus* : *Streptococcus thermophilus* = 1 : 1

B = *Bifidobacterium* : *Lactobacillus acidophilus* = 1 : 1

Scale : 1 = dislike extremely

2 = dislike moderately

3 = moderately

4 = like moderately

5 = like extremely

From Table 2, the average of the preference test has no significance. It means that the consumer could accept both yoghurt. Even the yoghurt from *Bifidobacterium* and *Lactobacillus acidophilus* starter has

slightly higher score than the yoghurt with *Lactobacillus bulgaricus* and *Streptococcus thermophilus* starter, and also for colour, and aroma, but has lower score for taste, consistency and texture.

Glucose content.

The glucose content in mice blood, was tested after 7 weeks of the trial. In Table 3, presents the results of the glucose content that was tested after 7 weeks trial, and after 3 hours the mice consumpt with 1.25% and 2.00% yoghurt.

Table 3. Blood glucose content in mice (mg/100 ml)

Dosage (%)	Yoghurt			
	A	A	B	B
	Male	Female	Male	Female
1.25	171	176	136	137
2.00	176	177	165	166

Notes : A = *Lactobacillus bulgaricus* : *Streptococcus thermophilus* = 1 : 1

B = *Bifidobacterium* : *Lactobacillus acidophilus* = 1 : 1

After 7 weeks treatment with 1.25% and 2.00% dosage in male and female mice, and 3 hours after consumpt; the average of blood contents are varied between 136 mg/100ml and 177 mg/100 ml. The glucose content in the mice (male and female) from both dosage (1.25 and 2.00%), that consumed with *Lactobacillus bulgaricus* and *Streptococcus thermophilus* yoghurt are higher than the *Bifidobacterium* and *Lactobacillus acidophilus* yoghurt in both sexes. And between sexes, the male are lower than the female; and when the dosage are higher, the blood content will higher too. The yoghurt with *Bifidobacterium* and *Lactobacillus acidophilus* starters, has lower glucose content, because could raised the blood insulin so the blood glucose will decreased. Even it decreased, but the glucose content still in normal range.

CONCLUSIONS

The yoghurt that fermented with *Lactobacillus acidophilus* and *Bifidobacterium* has no effect on yoghurt quality than the yoghurt with *Lactobacillus bulgaricus* and *Streptococcus thermophilus*, and the blood glucose will decrease after 3 hours treatment even still in normal range.

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THE BIOFERMENTATION OF *LACTOBACILLUS ACIDOPHILUS* AND *BIFIDOBACTERIUM* ON MILK AGAINST THE ACTIVITY OF LIPASE AND MICE BLOOD CHOLESTEROL

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The development in food fermentation technology allowed humans to use microbes ability optimally to increase the benefit of materials that could be damaged to products that could resist and have a high economical value.

*The aim of the research is to explore the effect of the fermentation with *Lactobacillus acidophilus* and *Bifidobacterium* against the ability to increase the activity of lipase and to decrease the cholesterol value of mice blood.*

*Results indicated that the activity of lipase in milk fermented with *Lactobacillus acidophilus* and *Bifidobacterium* are 0.45 units/ml, and the cholesterol value is 169.5 mg/dl, compared with the *Lactobacillus bulgaricus* and *Streptococcus thermophilus* milk fermented that is 0.18 units/ml, and 224.3 mg/dl.*

Key words : Biofermentation, lipase, cholesterol, *Lactobacillus acidophilus*, *Bifidobacterium*

Lactobacillus acidophilus is a slow acid producer in cultures used for the production of acidophilus milk. This product has a plain acid flavour, and may be combined with other bacteria to obtain a more palatable product. Acidophilus milk is recommended as a dietary product, because *Lactobacillus acidophilus* functions in the large intestine of man (van den Berg, 1988) In bioghurt, *Lactobacillus bulgaricus* is replaced by *Bifidobacterium* in place of *Lactobacillus bulgaricus* is that *Bifidobacterium* produces the physiologically active D(+)-lactic acid. It is assumed that a high concentration of cholesterol in the diet affects the cholesterol level in the blood serum. The endogenous lipolytic activity in intestine is low so that lipases ingested will make a significant contribution to intestinal lipolysis (Cross and Overby, 1988).

Some of the milk enzymes, can be found in the skim milk, e.g. lipase and lysozyme (Cross and Overby, 1988).

The aim of this work was to get the effect of fermentation with *Lactobacillus acidophilus* and *Bifidobacterium* on gaining the lipase activities and to reduce the blood cholesterol in mice.

MATERIALS AND METHODS

The milk. We are using raw milk (cow milk) from farm animals at the faculty.

The bacteria. The bacteria are pure cultivated *Bifidobacterium*, *Lactobacillus acidophilus*, *Lactobacillus bulgaricus* and *Streptococcus thermophilus*. The identification of the cultures was based on the characteristics of the lactobacilli and streptococcus as described in Bergey's Manual of Determinative Bacteriology (Holt, et al, 1994). Bacteria total count with Total Plate Count (TPC).

Lipase activity. Lipase activities are tested according to Bergmeyer, 1981, after 7 weeks fermented.

Blood cholesterol. Blood cholesterol was tested according to Randox.

RESULTS AND DISCUSSION

The effect of the treatment on lipase activities

Lipase activities are responsible for the rancid flavour in milk and milk products. They hydrolyse glycerides, thus liberating free fatty acids. This process is called lipolysis. Lipase enzymes are still active at temperatures as low as 4 to 5^o C. Most of them are destroyed at normal or slightly increased HTST pasteurization temperatures, but rancidity already developed in raw milk will not be removed during pasteurization (van den Berg, 1988).

Table 1. Total cell counts and Lipase activities in yoghurt

Yoghurt	Total cell count (cell/ml)	Lipase activities (unit/ml)
<i>Lactobacillus bulgaricus</i> and <i>Streptococcus thermophilus</i>	13 x 10 ¹¹	0.18
<i>Bifidobacterium</i> and <i>Lactobacillus acidophilus</i>	7.6 x 10 ⁹	0.45

In Table 1. , presents the total cell counts in yoghurt and the lipase activities in yoghurt. After fermented with *Lactobacillus acidophilus* and *Bifidobacterium*, total cell count was 13 x 10¹¹ cell/ml and lipase activities was 0.45 unit/ml, compared to the *Lactobacillus bulgaricus* and *Streptococcus thermophilus* fermentation; the total cell counts was 7.6 x 10⁹ cell/ml and the lipase activities only 0.18 unit/ml.

The distribution of lipase between serum and cream fractions was altered, indicating that the equilibrium between the lipase activity of skim milk and cream is temperature dependent. The distribution of lipase in milk depends primarily on the lipid phase.

The effect of the treatment on blood cholesterol

Milk fat should only refer to the pure fat, but since the fat is associated with other fat-like substances, such as lecithine and cholesterol, the term lipids is often used for all fatty components that can be extracted from the milk by ether. Cholesterol is an alcoholic compound of complex structure. In milk, most of it is found in the fat globules. Unfortunately, milk fat has been suspected of being injurious to health. Particular attention has been given to its cholesterol content. It has been assumed that the consumption of milk fat will increase the cholesterol content of the blood.

In Table 2, presents the cholesterol content in mice blood cholesterol.

Table 2. Blood cholesterol content in mice (mg/100 ml) after 7 weeks treatment

Dosage (%)	Yoghurt			
	A	A	B	B
	Male	Female	Male	Female
1.25	172	147	153	144
2.00	166	140	143	142

Notes : A = *Lactobacillus bulgaricus* : *Streptococcus thermophilus* = 1 : 1

B = *Bifidobacterium* : *Lactobacillus acidophilus* = 1 : 1

It is assumed that a high concentration of cholesterol in the diet affects the cholesterol level in the blood serum. The cholesterol content of milk fat is relatively low compared to that of other animal foods. The average cholesterol content of milk is 13 mg/100 ml (van den Berg, 1988).

After 7 weeks treatment with 1.25% and 2.00% dosage in male and female mice, the average of blood cholesterols are varied between 140 mg/100ml and 172 mg/100 ml. The blood cholesterol content from the mice (male and female) from both dosage (1.25 and 2.00%), that consumed with *Lactobacillus bulgaricus* and *Streptococcus thermophilus* yoghurt are higher than the *Bifidobacterium* and *Lactobacillus acidophilus* yoghurt in both sexes. And between sexes, the male are higher than the female; and when the dosage are higher, the blood cholesterol will lower too. The yoghurt with *Bifidobacterium* and *Lactobacillus acidophilus* starters, has lower cholesterol content, because the cholesterol in the diet has only a limited effect on the level of cholesterol in the blood plasma, since the body has a control mechanism which ensures that the synthesis of cholesterol by the body is reduced when the amount of cholesterol consumed increases.

CONCLUSIONS

The blood cholesterol content that consumed with *Lactobacillus bulgaricus* and *Streptococcus thermophilus* yoghurt are higher than the *Bifidobacterium* and *Lactobacillus acidophilus* yoghurt. The yoghurt with *Bifidobacterium* and *Lactobacillus acidophilus* starters, has lower cholesterol content, because the cholesterol in the diet has only a limited effect on the level of cholesterol in the blood plasma, since the body has a control mechanism which ensures that the synthesis of cholesterol by the body is reduced when the amount of cholesterol consumed increases.

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