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THE EFFECT OF LACTIC ACID BACTERIA STARTER CULTURES ON THE QUALITY OF YOGHURT AND GASTROINTESTINAL TRACT ECOSYSTEM IN MICE

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ABSTRACT

This research was aimed to evaluate the implication effect of yoghurt microbes: *Lactobacillus bulgaricus, Streptococcus thermophillus, Lactobacillus acidophilus, Bifidobacterium* and their mixtures to intestine microflora on mice gastrointestinal tract. The method of manufacture and the composition of the cultures used for the fermentation of the milk vary from area to area. The time of fermentation depends on the temperature, the percentage of inoculation and the type of the culture. The percentage may vary from 0.5 to 5.0%, but usually the lower percentage is applied. This trial used a Complete Random Design (CRD) with 4 treatments and each treatment was repeated 4 times; so there were 16 units and each unit consist of 5 mice and the total number of mice used were 80 . Results indicated that yoghurt with correct measurement of probiotic mixtures (*Lactobacillus bulgaricus, Streptococcus thermophillus, Lactobacillus acidophilus,* and *Bifidobacterium*) has a good implementation in mice microfloral intestine; which increased the population of non patogenic microfloral, especially the mixture contained *Lactobacillus acidophilus,* and *Bifidobacterium.* They also decreased the population of pathogenic microflora such as micrococci in the mice colon , also *Escherichia coli* and *Staphylococcus* spp.on sensitivity reaction

Key words : gastrointestinal tract ecosystem; *Bifidobacterium, Lactobacillus acidophilus, Lactobacillus bulgaricus,* and *Streptococcus thermophilus.*

INTRODUCTION

A functional food that can promote human health and well-being better than a similar traditional food. The most widely used and recognized functional food is yoghurt, a product made by fermenting milk with bacteria. Prior to the discovery of specific microbial pathogens, scientists recognized that the intestinal flora could be improved by the ingestion of yoghurt. The newest, and perhaps the most important group of foods in the category of functional foods are the probiotics. Probiotic is a Greek word which means "for life." It is used to describe the "friendly" bacteria that normally live in the intestinal tract and which contribute to good health. Unfortunately, probiotics can be destroyed or depleted by stress, aging, antibiotics, changes in one's diet, or ingestion of foreign bacteria . Probiotics is also a label for food products that contain health-promoting bacteria – primarily lactic-acid bacteria (LAB) that have a positive influence on the metabolic activity of the intestinal flora.

Over the past several years the relationship of the intestinal tract health to the overall health of the body has become increasingly appreciated. The human gastrointestinal (GI) tract is home to a vast and complex bacterial ecosystem, hosting over 400 different species (1,2). The GI tract in a healthy adult is about 30 feet long and contains anywhere from 5 to 15 pounds of

living bacteria. In healthy individuals the most common of these organisms includes *Escherichia coli.*

Lactobacillus constitute a major part of the microflora throughout the gastrointestinal tract. These bacteria have been proposed as candidate probiotic microorganisms to reinforce the barrier effect in the gut. Consumption of probiotic bacteria can alleviate intestinal inflammation, normalize increased intestinal permeability, and strengthen the intestine's immunologic barrier function (3,4). These gut microflora play a vital role in human health and perform important metabolic functions that support the digestive system. Research reveals that the gut lining is primarily nourished by nutrients produced from favorable bacteria – not by our blood supply, as was previously believed. We are completely reliant upon the activities of these bacteria for the manufacture of key vitamins, the assimilation and distribution of nutrients, and for the suppression of pathogenic and putrefactive bacteria.

MATERIALS and METHOD

a. Materials :
   - Eighty mice which placed randomly on 16 plot experimental cages, and each cages for 5 mice.
   - *Bacteria: Lactobacillus bulgaricus, Streptococcus thermophilus Lactobacillus acidophilus*, and Bifidobacterium were mixed as yoghurt starter in this research.

b. The dietary treatments consisted of :
   Y1 = *Lactobacillus bulgaricus* + *Streptococcus thermophilus*
   Y2 = *Lactobacillus bulgaricus*+ *Streptococcus thermophilus* + *Lactobacillus acidophilus*
   Y3 = *Lactobacillus bulgaricus* +*Streptococcus thermophilus* +*Bifidobacterium*
   Y 4= *Lactobacillus bulgaricus* +*S. thermophilus* + *L. acidophilus* + *Bifidobacterium*
   Yoghurt quality was tested according the preference test of aroma, taste and consistency.

c. The number of pathogenic and non pathogenic bacteria in mice colon :
   Pathogenic bacteria are *Staphylococcus aureus* and *Bacillus*; the non pathogenic bacteria are *Lactobacillus bulgaricus, Streptococcus thermophilus, Lactobacillus acidophilus*, and Bifidobacterium.

d. Antibiogram of starter isolates :
   The starter isolates were inoculated into MRS broth individually, according to the starter consentration, and incubated for 24 hr. Abou 20ml MRS agar was seeded with th cultures of yoghurt starter, mixed well, poured into sterile Petri dish and stored at 4C for 1hr to solidify the media. OCTA-discs (OXOID) were placed upside down, pressed on the top pf the agar plates and kept again at 4C for 1hr. The plates ere incubated at 37C over night. Resistance was defined as the absencce of a growth inhibition zone aound the discs.

RESULTS and DISCUSSION

1. Pathogenic and non-pathogenic bacteria ratio.

Table 1, The ratio of pathogenic and nonpathogenic bacteria

Colon	Bacteria	Y1	Y2	Y3	Y4
Pathogenic bacteria	Stp	$4{,}0x10^{1}$	-	-	
	B	$4{,}0x10^{2}$	-	-	$3{,}5x10^{1}$
Non Pathogenic bacteria	Lb	$5.50\ x10^{2}$	$4.37\ x10^{2}$	$1.20\ x10^{0}$	$1.00\ x10^{0}$
	St	$8.51\ x10^{1}$	$1.00\ x10^{0}$	$1.30\ x10^{0}$	$9.12\ x10^{2}$
	La	$1.95\ x10^{3}$	$4.61\ x10^{3}$	$1.9\ 5\ x10^{7}$	$2.82\ x10^{6}$
	B	$5.62\ x10^{1}$	$4.00\ x10^{3}$	$4.51\ x10^{4}$	$9.50\ x10^{6}$

Notes :
Stp = *Staphylococcus aureus.*
B = *Bacillus*
Lb = *Lactobacillus bulgaricus*
St = *Streptococcus thermophilus*
La = *Lactobacillus acidophilus*
B = Bifidobacterium
Y1= *L. bulgaricus : S. thermophillus*
Y2= *L.bulgaricus : S. thermophillus : L. acidophillus*
Y3= *L. bulgaricus : S. thermophillus : Bifidobacterium*
Y4= *L bulgaricus : S. thermophillus: L. acidophilus : Bifidobacterium*

Results indicated that the pathogenic bacteria in the colon, after the mice was fed with yoghurt was decreased and the non-pathogenic was increased in the mice, especially that have been feed with *Bifidobacterium* and *Lactobacillus acidophilus.* The pathogenic bacteria in colon was decreased because the probiotic bacteria in yoghurt has inhibitory effect and on the other side, they grown well in the colon as non-pathogenic bacteria. This is similar according to Moore et al; 1974 and Kaila, et al, 1992; that Lactobacillus constitute a major part of the microflora throughout the gastrointestinal tract. These bacteria have been proposed as candidate probiotic microorganisms to reinforce the barrier effect in the gut. Consumption of probiotic bacteria can alleviate intestinal inflammation, normalize increased intestinal permeability, and strengthen the intestine's immunologic barrier function (Moore, et al, 1974 and Kaila,et. al, 1992). So, as mixture starter in yoghurt it has positive function to the consumer that consume yoghurt with probiotic starters.

2. Yoghurt quality.

In Table 2, present the results of organoleptic test of aroma, taste and consistency.

Tabel 2. Organoleptic Tests

	Consumer Preferency		
	Aroma	**Taste**	**Consistency**
Y 1	2.80[a]	3.05[a]	3.20[a]
Y 2	3.25[b]	2.90[a]	3.26[a]
Y 3	3.45[b]	3.15[a]	3.27[a]
Y 4	3.55[b]	3.10[a]	3.45[a]

Notes: Y1= *L. bulgaricus : S. thermophillus*
Y2= *L.bulgaricus : S. thermophillus : L. acidophillus*
Y3= *L. bulgaricus : S. thermophillus : Bifidobacterium*
Y4= *L bulgaricus : S. thermophillus: L. acidophilus : Bifidobacterium*

Criteria :1. Exteme dislike ; 2. Like; 3. Neutral; 4. Extreme Like; and 5. Exellent

From Table 2, results indicated that there is no difference for consumer preferency in

taste and consistency for all mixture of starters, but for aroma, yoghurt content *Lactobacillus bulgaricus, Streptococcus thermophilus, Lactobacillus acidophilus* and *Bifidobacteria* gave better aroma than the yoghurt with *Lactobacillus bulgaricus* and *Streptococcus thermophillus* starters.

3. **Sensitivity reaction of starter mixture to *E.coli and S. aureus*.**

In Table 3, present the results of starter mixture of starters on *E. coli and Stapylococcus aureus.*

Tabel 3. Sensitivity reaction of starter mixture to *E. coli and S. aureus*

	Mixture starters	Inhibition zone (mm)	
		E coli	*S. aureus*
1	Y1(Lb +St)	0.0	0.0
2	Y2 (Lb+St+La)	0.0	0.2
3	Y3 (Lb+St+B)	0.2	0.4
4	Y 4 (Lb+St+La+B)	0.4	0.6

Notes : Y1= *L. bulgaricus : S. thermophillus*
Y2 = *L.bulgaricus : S. thermophillus : L. acidophillus*
Y3 = *L. bulgaricus : S. thermophillus : Bifidobacterium*
Y4 = *L bulgaricus : S. thermophillus: L. acidophilus : Bifidobacterium*

Results indicatd that Y1 has no sensitivity effect to *Escherichia coli* and *Staphylococcus aureus*, and the Y4 mixtures gave more sensitivity both to *Escherichia coli* and *Staphylococcus aureus*. It means that starter mixtures of *Lactobacillus bulgaricus, Streptococcus thermophilus,Lactobacillus acidophilus* and Bifidobacterium) could inhibit the pathogenic bacteria (*Escherichia coli* and *Staphylococcus aureus*).(lovita, A.,2009)

CONCLUSION

1. The pathogenic bacteria in the colon was decreased , and non pathogenic bacteria was increased because the consorsium bacteria that added with *Lactobacillus acidophilus and Bifidobacterium than the yoghurt* only content *Lactobacillus bulgaricus and Streptococcus thermophilus* starters.

2. The quality, according to organoleptic test has no significancy for taste and consistency, but for aroma, the yoghurt with *Lactobacillus bulgaricus, Streptococcus thermophilus, Lactobacillus acidophilus* and *Bifidobacteria* consortium, gave better aroma than the yoghurt ccontent *Lactobacillus bulgaricus* and *Streptococcus thermophillus* starters.

3. Yoghurt content *Lactobacillus bulgaricus, Streptococcus thermophilus* has no sensitivity effect to *Escherichia coli* and *Staphylococcus aureus*, but when added with Lactobacillus acidophilus and Bifidobacterium mixtures gave more sensitivity both to *Escherichia coli* and *Staphylococcus aureus*. It means that starter mixtures of *Lactobacillus bulgaricus, Streptococcus thermophilus,Lactobacillus acidophilus* and *Bifidobacterium* could inhibit the pathogenic bacteria

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