

Study of Antibacterial Activity and Viability of Lactic Acid Bacteria (BAL) on Fermented Milk Beverage Products Circulating in Malang City

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ABSTRACT

The results showed that eight brands of fermented milk drinks circulating in Malang City showed 6 brands of fermented milk drinks namely brands SF1, SF2, SF3, SF4, SF5, and SF6 containing live lactic acid bacteria cultures in their products. Meanwhile, the SF7 and SF8 brands do not contain live lactic acid bacteria cultures in their products. Fermented milk drink is a form of probiotic food which has long been known as a carrier food for probiotic organisms. Probiotics are food supplements in the form of live microbes that have beneficial effects on the human body by balancing microbes in digestion. Viability and functional activity of probiotics is an important thing in a probiotic supplement product. One of the functional activities of this probiotic food is the ability of the bacterial culture in it to inhibit the growth and activity of bacteria that are pathogenic to the body. showed that fermented milk drinks that had live and active cultures had higher antibacterial activity when compared to fermented milk drinks that did not have probiotic bacterial cultures in them. SF2 brand fermented milk drink has a higher activity of inhibiting the growth of pathogenic *Staphylococcus aureus* bacteria when compared to other brands. As for the inhibition of *Escherichia coli* pathogenic bacteria, fermented milk drink brand SF3 has the highest antibacterial activity.

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1. INTRODUCTION

Milk can be defined as a white liquid obtained from milking cows or other mammals, without reducing or adding other ingredients. In general, the milk consumed daily is milk from dairy cows (Hadiwiyoto, 1994).

Milk is a food ingredient that contains lots of nutrients, including high levels of protein, carbohydrates, fats, vitamins, and several minerals. Normal milk has a slightly salty and sweet taste. The smell of milk is very distinctive with a slight fishy smell. The smell of milk will disappear if left a few hours or aerated. The color of normal milk varies from white-gray to yellow-brown, depending on the amount of fat and non-fat solids (Lampert, 1975).

Fermented milk drink is a form of probiotic food which has long been known as a carrier food for probiotic organisms (Adhikari et al., 2003). Probiotics themselves are food supplements in the form of live microbes that have a beneficial effect on the human body by balancing microbes in digestion. Probiotic food has now become a food product that plays an important role for health.

Several scientific studies and previous articles have revealed the relationship between the consumption of probiotic foods derived from milk ("dairy products") and health. Most of the dairy products ("dairy products") are in the form of fermented milk beverage products.

one type of fermented milk drink that is quite popular, yogurt has antibacterial activity where the lactic acid bacteria present in yogurt can inhibit the growth of *Escherichia coli* and *Salmonella typhosa* bacteria "in vitro". Low acidity is one of the factors that can inhibit the growth of *Escherichia coli* and *Salmonella typhosa* bacteria. In addition to the pH and acidity factors in yogurt, there are also antibacterial substances, namely in the form of bacteriocins.

In fermented milk drinks, the use of "starter" cultures can be done using pure probiotic cultures such as *acidophilus* and yakult milk drinks, each of which uses probiotic lactic acid bacteria *Lactobacillus acidophilus* and *Lactobacillus casei* strain Shirota or using natural/traditional cultures that are not classified as probiotics such as *L. bulgaricus* and *S. thermophilus* which are found in the process of making yogurt.

These bacteria have a role in the formation of sour yogurt flavor. The rapid growth at the beginning of the fermentation accumulates the formation of large amounts of lactic acid, acetic acid, acetaldehyde, diacetyl and formic acid.

2. METHOD

2.1 Types of research

The research method used in this research is a survey method (descriptive) with three repetitions. Samples were obtained as many as 8 (eight) different brands of fermented milk drinks from various places in Malang City randomly. Results Data analysis was carried out by comparing the data obtained from the test results, both microbiological and chemical analysis as well as the relationship between one another and then conclusions were drawn.

2.2 Research Variables

The variables in this study consisted of independent variables which were 8 types of milk and the effect of lactic acid bacteria on milk.

2.3 Research design

Testing of Solid Samples and Liquid Samples in Design to determine the performance of the tool in direct testing. The Digital Formaldehyde Meter test method uses a solid sample (the sample is cut into 3 cm as much as 5 grams) tested placed in a closed container with a volume of 30 ml and a liquid sample with a volume of 30 ml, then heated for 1 minute and the results will be displayed on the LCD.

2.4 Sampling location

Samples were obtained as many as 8 (eight) different brands of fermented milk drinks from various places in Malang City randomly.

2.5 Time and Place of Research.

This research was conducted at the Laboratory of Biochemistry and Nutrition, Department of Agricultural Product Technology, Faculty of Agricultural Technology, University of Brawijaya, Malang, from January 2006 to April 2006.

2.6 Tools and materials

The equipment used in this study included "Laminar Air Flow", electric stove (Maspion S-300 220V), autoclave (HL-36 AE Hiramaya Japan), spectrophotometer, incubator (Binder BD53 Germany), pH meter (model pH S- 3C), digital balance (Denver Instrument M-310), vortex, volume pipette, petridish, test tube, beaker glass, erlenmeyer, stir bar, measuring pipette, measuring cup, suction ball, ose, bunsen, sprayer, micropipette (Finn pipette Labsystem), rubber bands, polypropylene plastic bags, brown paper (umbrellas), and cotton and various other microbiological equipment.

The materials used for analysis include MRSA media (de Mann Rogose Sharpe Agar), MRS (de Mann Rogose Sharpe) Broth, NB (Nutrien Broth), NA (Nutrien Agar), TSB (Triptone Soya Broth), sterile peptone solution, 70% alcohol, 1N NaOH solution, oxalic acid, pp indicator, distilled water, phosphate buffer pH 7 solution obtained from the Laboratory of Biochemistry and Nutrition, Department of Agricultural Product Technology.

2.7 Research procedure

The sampling method used is "purposive random sampling design". Fermented milk products that will be used as research samples are fermented milk products stored at low temperatures

("cold storage"). The considerations in selecting fermented milk products are brand, composition, viscosity, and expiration date and are tested with test parameters for Total Lactic Acid Bacteria (LAB), Antibacterial Activity, Total Acid and pH.

2.8 Data analysis.

Sample analysis was carried out on 8 brands of fermented milk drinks which included observations of various parameters, namely microbiological analysis, chemical analysis and surveys and sampling.

3. RESULTS AND DISCUSSION

3.1 Research result

The survey results show that fermented milk drinks circulating in Malang City are dominated by types of yoghurt, acidophilus milk, and yakult which consist of local/domestic and imported/foreign products. This fermented milk product is marketed in most supermarkets and small shops. While being marketed, this fermented milk drink is stored in a room or place/container with low temperature. Where the existing environmental conditions can ensure that the fermented milk drink does not suffer damage while being marketed.

These products are marketed using various packaging materials and sizes. Among them are using polyethylene terephthalate (PET) and polypropylene (PP) plastic packaging, most of which are made in the form of plastic cups (cups), bowls and bottles. Package sizes also vary between 65 ml to 200 ml. The viability of lactic acid bacteria present in fermented milk beverage products is an important parameter in probiotic beverage products. Fermented milk drink which is a form of probiotic food will be beneficial if it contains lactic acid bacteria in large quantities, remains alive and stable during storage.

According to Anonymous (2002), not all yogurt contains live cultures. After the fermentation process some yogurt is pasteurized again. This can kill beneficial bacteria and can reduce the health effects of yogurt, which are dependent on live and active cultures. Pasteurization carried out on yogurt after fermentation is intended to extend its shelf life, but this can damage the nutritional and health value of yogurt. According to Anonymous (2002), not all yogurt contains live cultures. After the fermentation process some yogurt is pasteurized again. This can kill beneficial bacteria and can reduce the health effects of yogurt, which are dependent on live and active cultures.

3.1.1 Antibacterial Activity

The results of the analysis of antibacterial activity showed that the level of turbidity (abs) of TSB (Tryptone Soya Broth) media overgrown with the indicator pathogenic bacteria *Escherichia coli* at the end of the observation (9th hour) ranged from 0.387 – 0.473.

Table 1. Average 9th Hour Medium Turbidity (*S. aureus* Indicator Bacteria) in Fermented Milk Beverage Products Circulating in Malang City

No	Sample	Average sample medium	
		Turbidity At 9 Hours (abs)	
1	Prime (SF1)	0.406	
2	Queens (SF2) 0.372	0.372	
3	Bull (SF3) 0.379	0.379	
4	Vitacharm (SF5)	0.420	
5	Yakult (SF6)	0.412	
6	Nice (SF7)	0.432	
7	Elle & Vire (SF8)	0.444	
8	Control	0.459	

Based on the analysis results, it can be seen that the SF2 brand fermented milk drink product has the lowest turbidity (abs) value at the 9th hour, which is equal to 0.372. While the turbidity value (abs) is the highest in the SF8 brand, which is 0.444. A high turbidity value (abs) means that the growth rate of pathogenic bacteria is high.

3.1.2 Total Acid and Degree of Acidity (pH)

The total acid (calculated as lactic acid) in fermented milk drinks circulating in Malang City ranges from 0.31 to 1.51%. Graph showing the average total acid levels in eight brands of fermented milk drinks.

Table 2. Average Total Acids of Fermented Milk Beverage Products Circulating in Malang City

No	Sample	Total Acids of Milk Beverage Products	
		Total acid %	

1	Prime (SF1) 1.51	1.51
2	Queen (SF2) 1.16	1.16
3	Emmi (SF4) 1.06	1.06
4	Vitacharm (SF5) 0.60	0.60
5	Yakult (SF6) 0.82	0.82
6	Nice (SF7) 0.31	0.31
7	Elle & Vire (SF8) 0.53	0.53

While the lowest total acid value was found in the SF7 brand of 0.31%. The different values of total acid (calculated as lactic acid) are thought to be due to the different content of lactic acid bacteria and the availability of lactose which will be broken down into lactic acid during the fermentation and storage processes. Shah (2000) stated that during the low temperature storage process, starter culture in fermented milk drinks still produces lactic acid which in the fermentation industry is known as "post acidification". The degree of acidity in fermented milk beverage products circulating in Malang City ranges from 3.9 to 4.3. Graph showing the average degree of acidity (pH) in eight brands of fermented milk drinks.

Based on the results of pH analysis, it is known that fermented milk drink products that have a low pH have a high number of lactic acid bacteria (fermented milk drinks SF1, SF2, SF3, SF4, SF5, and SF6). Meanwhile, fermented milk drink products SF7 and SF8 which did not contain lactic acid bacteria at all had a higher pH value. Live lactic acid bacteria present in fermented milk drinks will continue to break down lactose and produce lactic acid which will continue to accumulate and cause a decrease in pH.

3.2 Discussion

The test results showed that the average tool error for testing liquid samples was 2.78% while for solid samples it was 2%. Tests on several foods obtained from the market such as meatballs, sausages, and tofu have formalin levels that are safe for consumption, while marine fish are dangerous for consumption. In addition, dead protein will not be attacked by spoilage bacteria that produce acidic compounds. others are more durable.

Formaldehyde kills bacteria by making the tissue in the bacteria dehydrated (lack of water), so that the bacterial cells will dry and form a new layer on the surface. This means that formalin not only kills bacteria, but also forms a new layer that protects the layer beneath it, so that it is resistant to attack by other bacteria. If other disinfectants deactivate bacterial attacks by killing and not reacting with the protected material, formaldehyde will react chemically and remain in the material to protect against further attacks. Electronic nose technology is a data acquisition technology with a data processing link, usually done to solve the problem of a tool-making system consisting of gas sensor arrays (gas sensor array). Electronic nose is a portable system that has advantages such as small size and low operating costs. Previous research has widely used Electronic Nose for detection of environmental air safety, medical applications, and food safety (Zhang et al., 2009).

4. CONCLUSION

The antibacterial activity of eight brands of fermented milk drinks as indicated by the turbidity level of the TSB growing medium added to the pathogenic bacteria *Staphylococcus aureus* and *Escherichia coli* shows that fermented milk drinks that have live and active cultures have higher antibacterial activity when compared to fermented milk drinks that do not have probiotic bacteria culture inside.

SF2 brand fermented milk drink has a higher activity of inhibiting the growth of pathogenic *Staphylococcus aureus* bacteria when compared to other brands. As for the inhibition of *Escherichia coli* pathogenic bacteria, fermented milk drink brand SF3 has the highest antibacterial activity.

Results Data analysis was carried out by comparing the data obtained from the results testing both microbiological and chemical analysis as well as the relationship between one another to then draw conclusions. The results showed that eight brands of fermented milk drinks circulating in Malang City showed 6 brands of fermented milk drinks namely brands SF1, SF2, SF3, SF4, SF5, and SF6 containing live lactic acid bacteria cultures in their products. Meanwhile, the SF7 and SF8 brands do not contain live lactic acid bacteria cultures in their products.

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In consuming fermented milk drinks, consumers should pay attention to the type of fermented milk drinks consumed, especially related to the content of probiotic cultures in them. Because only products that have probiotic cultures can have a positive effect on health and further research is needed to find out how much intake or minimum daily dose of fermented milk drinks is to meet its antibacterial effectiveness for the body and the need for identification of the most dominant antibacterial compounds in inhibiting the growth of pathogenic bacteria.

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