American Scientific Research Journal for Engineering, Technology, and Sciences (ASRJETS)

ISSN (Print) 2313-4410, ISSN (Online) 2313-4402

http://asrjetsjournal.org/

Performance Analysis of Industrially and Traditionally Used Cultures in Yogurt Production Considering Microbial Risk

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Abstract

Yogurt is a fermented milk product produced by two ways; traditional method where local cultures is used and industrial method where industrial starter culture is used for yogurt production in dairy industries. In this study, yogurt cultures were collected from the local markets and from modern dairy plants. This study was undertaken to analyze the performance of industrially used thermophillic, mesophillic homofermentative culture and traditionally used cultures in yogurt production. Thermophillic, mesophillic homofermentative and BAU dairy farm used yogurt culture needed 4, 5 and 3 hours to complete yogurt production and percentages of acidity were 0.76, 0.73, and 0.74 respectively. Local market culture needed highest time (6 hours) for yogurt production and percentage of acidity was 0.96. Maximum (10 coliform cfu) was found in sample-4, but coliform was absent in sample-1 and sample-2. Total yeast count was nill for all samples. Salmonella and shigellae was found in sample-3 and sample-4 yogurt sample. Syneresis and total fungal count was done to find out the shelf life quality of yogurt samples. After 12 days syneresis observed in sample-2 but for sample-1 it was found after 11 days. Syneresis observed in Sample-3 and Sample-4 after 10 days.

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This study stated that, the yogurt made by mesophillic homofermentative culture has long shelf life than other yogurt culture, so mesophillic homofermentative culture is more suitable for industrial yogurt production.

Keywords: Yogurt; starter culture; syneresis; acidity; shelf life

1. Introduction

Yogurt as a dairy food can be consumed in form of snack, thirst quenching beverages and as a desert. It is semi solid custard like product obtained from pasteurized or boiled milk by souring, natural or otherwise, by a harmless lactic acid or other bacterial culture. Yogurt may contain additional cane sugar. Sugar was added at the rate of 8 to 10% during preparation [1] Milk is very nutritious and obligatory food for human being. But in this era of industrialization, food habit of common people is changing. They are preferable as it is healthy, delicious foods to fresh raw foods. Hence, milk is converted to various milk products, like yogurt, fermented milk, cheese, butter, yogurt, milk ice-cream etc. of which yogurt is locally available dairy product in Indian sub-continent.

Like milk, yogurt is also very nutritious as it is a good source of iodine, calcium, phosphorus, zinc, riboflavin, vitamin B_5 and vitamin B_{12}. It is also nutritionally rich in protein, molybdenum and pantothenic acid [2] The food rating system adopted as the government standard for food labeling that are found in the U.S food and drug administration allow yogurt to be rated as one of the world's healthiest food. Yogurt bacteria are also capable of manufacturing the entire range of B- complex vitamins in the intestine. This is very important because many modern drugs kill intestinal flora. Another advantage of yogurt is that people who are allergic to milk can generally eat it safely. The bacterial culturing agent consumes most of the lactose, which is one of the main reasons for intolerance of milk, partially in adults. The bacteria also act on the milk protein so that it becomes in effect pre-digested. The changes to the lactose and protein of milk in yogurt render it easily digestible, even by chronic invalids. It has been demonstrated that acid milk is somewhat easily digestible than normal milk. For some individuals, yogurt has a definite therapeutic value, especially, who usually suffer from stomach and intestinal disorders. The use based on the assumption that the acid fermenting bacteria and lactose of milk are able to create conditions in the intestinal tract which are unfavorable for the growth of putrefactive, bacteria and thereby prevents the formation of gas and a condition known as autointoxication [3] Consumption of fermented milk products is associated with several types of health benefits partly because of their content of lactic acid bacteria. Several experimental observations have indicated a potential effect of lactic acid bacteria against the development of colon tumors. A wide range of other health benefits include, improved lactose digestion, prevent diarrhea, immune system modulation & serum cholesterol reduction [4].

In Bangladesh yogurt is one of the most popular fermented milk product which is made in mud pots usually from cow milk, sometimes from buffalo milk. The bacteria used are *Lactobacillus bulgaricus, Lactobacillus plantarum* and *Streptococcus thermophilus* and/or *streptococcus lactis* [5] Lower temperature (37°-42°C) and long incubation period (8-15h) is required for yogurt preparation. Yoghurt is manufactured using *Streptococcus thermophilus* and L. delbrueckii sp. bulgaricus as starter cultures. These organisms are claimed to offer some health benefits however, they are not natural inhabitants of the intestine. Therefore, for yoghurt to be considered as a probiotic product L.delbrueckii ssp. bulgaricus and S. thermophilus are at a daily dose of 10^9 cfu and

several authors have indicated that a minimal concentration of 10^{-6} cfu/g of a product is required for a probiotic effect [6]

In Turkey, yoghurt is produced by the two ways; one of them is a traditional method without using starter culture in small dairy plants and the second production method by using industrial starter culture in modern plants [7]. Many cultural bacteria can used for yogurt production i.e *Lactobacillus bulgaricus, Lactobacillus plantarum* and *Streptococcus thermophilus* and/or *streptococcus lactis* [7]. *S. thermophilus* grows more quickly a first and render he milk anaerobic and weakly acidic. *L. bulgaricus* then acidifies the milk even more. Acting together, the two species ferment almost all the lactose to lactic acid and flavor the yogurt with diacetyl and acetaldehyde. During such kind of yogurt generally prepared from mixed culture of *Streptococcus lactis, Streptococcus diacetilactis* and *Streptococcus cremoris,* however highly aromatic sweet results using *Lactobacillus planetarium* in place of *Streptococcus citrofilus* [8] in industrial production pure dry culture has been used for avoid contamination. Thermophillic yoghurt culture and Mesophillic homofermentative culture probably used most in industrial production. Different cultural bacteria used in yogurt production gives different appearance, color, flavor, test and it may favor for microbial attack. Therefore this study was conducted to analysis the performance of industrially used and traditionally used cultures in yogurt production considering microbial risk.

2. Methods and materials

2.1 Study Design

An experimental study was carried out to performance analysis of industrially used and traditionally used starter cultures in yogurt production.

2.2 Place of Experiment

The study was conducted in different laboratories at Aarong dairy laboratory, Dhaka, Bangladesh during the period of November 2013 to February 2014.

2.3 Raw Materials

Milk, pure starter yogurt culture (Thermophillic, Mesophillic) in powder form were collected from local market, BAU Dairy Farm yogurt culture and Culture used by local sweet producers were collected from Bangladesh Agriculture University (BAU) dairy farm and from local sweet markets.

2.3.1 Generation of MS-1 and MS-2 for industrial pure starter culture

For industrially used cultures i.e. Thermopfillic yoghurt culture and Mesophillic homofermentative culture were needed two times of generation. First made mother starter-1 (MS1) from pure culture powder and then made second generation (MS-2) from MS-1. But for local traditional cultures no generation time was needed.

2.3.2 Procedure of Making MS-1

At first 100 ml of distilled water was taken in conical flasks measured by a measuring cylinder and then heated the water in the water bath for about 40˚C. Then 10 gm of skim milk powder (0.5% milk fat, color less, odor less) were mixed in water and autoclaved at 121˚ C for 15 minutes in a digital autoclave machine to destroy all the unpleasant microorganisms e.i. coliform, shigella, salmonella, yeast and moulds those are responsible for contamination of yogurt, gas forming and producing bad odors. At the same time the temperature of water bath was increased and adjusted at 40 – 42 ˚C. Then 5 gm of pure thermophillic yogurt culture was added in flask, mixed culture properly and placed in the heated water bath for incubation at 40 – 42 ˚C. After 40 to 60 min incubation has stopped and culture structure reached in semi hard position. Finally the conical flasks was cooled at 4˚ C for storage and thus made of MS-1 to use.

2.3.3 Generation of MS-2

To produce MS-2 second time generation from MS-1 pasteurized milk (150 liters) in the culture generation vat was taken and heated up to 80˚ C and stand for 30 min and then cooled at 40˚C. About 2% culture of MS-1 was added in 150 liters generation vat and properly agitated. Filled up the container of 20 liters then placed for incubation at about 42˚C for 8 – 10 hours and stopped incubation when acidity reached at 0.80 percent. Then cooled the containers in the cold storage room at 4˚C and MS-2 was ready for further usage.

2.4 Yogurt production

In this study there were 4 cultures taken to compare the formation time of curd. At first 500 ml of yogurt samples were taken and poured in 500 ml raw milk and then heated in water bath to raise the temperature of milk. Then 10 gm (2%) of different culture was added to each milk pots at 42˚C temperature and mix properly. After that pots in water bath at 42˚C and stayed for incubation. Acidity and structure was checked every 30 minutes for each different culture product until full formation of yogurt.

2.5 Determination of acidity level

The growth of bacteria of yogurt culture is generally happened by increasing the acidity of milk. Increasing of acidity was identified by checking % acidity of milk or curd in titration method. The titration process was done every 30 minutes for each four different samples during incubation period and the result of % acidity was recorded. Incubation was stopped until the acidity comes 0.70 % or above because in this stage a desirable and acceptable curd structure gains but incubation will have to continue if desirable structure not found.

2.6 Microbiological Analysis

Total Coliform Count (TCC), Total Plate Count (TPC), Salmonella & Shigellae count, Yeast mould count of sample was done according to the method as described in the standard methods for the examination of dairy products [9]

2.7 Data Analysis

All analyses were performed in triplicate. All the ends of data collection, data were complied, tabulated and analyzed. The local units were converted in to the standard units. The comparisons in different sample were carried out by SPSS (16) program.

3. Results and discussions

3.1 Comparison of developing acidity in response to time for different samples

Table 1 indicates the growth rate of acidity for Sample-1 (thermophillic yogurt culture) in case of producing consumer product. First one hour development of acidity was slow but after two hours it was very fast after four hours it reached to 0.76 %. So, thermophillic yogurt culture can produce yogurt quickly within 4 hours along with full of suitable structure and acidity of yogurt. Acidity development in sample-2 (mesophillic homofermentative culture) was slower than sample-1. It needed five hours to reach the acidity at 0.7 percent and made an acceptable taste, flavor and structure of finished product. Acidity development in sample-3 (BAU dairy farm culture) was very quicker than other cultures. Only three hours needed to make a full flavored and structured yogurt for sample-3. The structure of this yogurt was very good and very hard. Table-1 also represent that, the increasing rate of acidity in sample-4 (Traditional culture used in local sweet markets) was very slow from other three culture described before. This culture took five hours to reach 0.73 percent of acidity, but achieving this acidity the yogurt structure was not good. Curd structure was not as hard as desirable. It may happen for bacterial contamination and also for use of sugar, caramel etc. Sugar act as a preservative that can inhibit the growth of bacteria. That's why bacteria cannot grow fast, acidity cannot increase quickly and for that yogurt production may slow. It took six hours to form of desirable hard yogurt structure and after six hours acidity reached at 0.96 percent.

Table 2 shows the time and average rate of acidity development for four different samples. It indicates that sample-3 (BAU Dairy farm culture) has the highest rate of acidity development and the value was 0.104/30 minutes. On the other hand sample-2 (mesophillic homofermentative culture) has lowest acidity development rate and the value was 0.06/30 minutes. This table also mention that sample-3 (BAU Dairy farm culture) needed lowest time to produce final yogurt product and sample -4 (local sweet market culture) needed highest time to produce final yogurt product.

3.2 Microbiological Quality Assessments

In this study, Total Coliform Count (TCC), Total Plate Count (TPC), Salmonella & Shigellae count, Yeast and Mould count were conducted to compare the microbiological quality of different types of prepared yogurt samples. Table 3 indicates the microbial load for four finished yogurt products made by different cultures. Highest plate count bacteria were found in Sample- 4 and lowest in sample-3 yogurt. Total coli form count bacteria was absent in sample-1 and 2 but 10 cfu was found in sample-4 yogurt. No yeast was found after 96 hours but one and two mould colonies were found in sample-3 and sample-4 respectively. Maximum amounts of salmonella were found in sample-4 but in sample-1 and 2 it was absent.

Table 1: Development of acidity in different sample

	Sample-1	Sample-2	Sample-3	Sample-4
	% acidity	% acidity	% acidity	% acidity
After 0.5 hr	0.2	0.16	0.22	0.17
After 1.0 hr	0.25	0.20	0.30	0.21
After 1.30 hr	0.31	0.25	0.39	0.28
After 2.0 hr	0.43	0.30	0.50	0.33
After 2.50 hr	0.54	0.37	0.62	0.39
After 3.0 hr	0.62	0.46	0.74	0.44
After 3.50 hr	0.68	0.53		0.50
After 4.0 hr	0.76	0.60		0.57
After 4.30 hr		0.66		0.66
After 5.0 hr		0.73		0.73
After 5.30 hr				0.82
After 6.0 hr				0.96

Table 2: Comparison of developing acidity in response to time for different samples

Sample	Time (hours)	Acidity	Average increase (acidity) rate /30 min
Sample-1	4	0.76	0.07
Sample-2	5	0.73	0.06
Sample-3	3	0.74	0.09
Sample-4	6	0.96	0.07

Table 3: Microbiological Quality Assessments of different types of Prepared Yogurt

Microbial parameter		Sample-1	Sample-2	Sample-3	Sample-4
Total plate count (cfu/ml)		(33±3.21) × 10^3	(37±3.78) × 10^3	(70±4.5) × 10^3	(109±9.6) × 10^3
Total coliform count (cfu/ml)		Nill	Nill	05±1.72	10±2.08
Total Fungal Count (cfu/ml)	Yeast	Nill	Nill	Nill	Nill
	Mould	Nill	Nill	01	02
Salmonella & Shigellae count		Nill	Nill	03	08

3.3 Shelf life of the finished product

Normally shelf life of yogurt is 10 to 12 days, but it would be more if yogurt can made under proper hygienically. Shelf life will reduce if excessive coliform and fungal bacteria (yeast/ mould) grow. It creates syneresis and produce off flavor of yogurt. In this circumstance, syneresis and total fungal count was done to find out the shelf life quality of four yogurt samples. Table-4 shows that, after 12 days syneresis observed in sample-2 but for sample-1 it was found after 11 days. Syneresis observed in Sample-3 and Sample-4 after 10 days. So it indicates that sample-2 has the highest shelf life than other cultures. That's why it will be more suitable and beneficial for industrial yogurt production. Table 4 also indicates that highest number of yeast and mould colonies was found after 12 days in sample -4 that indicates it will be syneresis with in ultimate 9-10 days. On the other hand after 15 days, lowest amount of yeast and mould colonies was found in sample-2 yogurt.

Table 4: Days to occurs syneresis and estimation of Yeast and Mould growth after 12 days

Sample	Syneresis observed	Yeast grows after 12 days	Mould grows after 12 days
Sample 1	11 days	$(13.66 \pm 3.21) \times 10^3$	10
Sample 2	12 days	$(11.33\ 0.57) \times 10^3$	9.33
Sample 3	10 days	$(20.3\pm 4.04) \times 10^3$	18.3
Sample 4	10 days	$(23.66 \pm 0.57) \times 10^3$	21.6

4. Conclusion

The study was carried out a comparison of time consumption for different types of yogurt culture. Comparison was done by identifying the increase of % acidity of product and microbial analysis also done for finished products. BAU Dairy farm culture has taken lowest time to complete finishes product but local sweet market culture has taken heights time to make final product. The yogurt made by mesophillic homofermentative culture has long shelf life than other yogurt culture and it is more beneficial for industrial business. However, further research work is needed in this field for identification/development of more suitable culture for industrial yogurt production.

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