Leonardo Electronic Journal of Practices and Technologies Issue 11, July-December 2007
ISSN 1583-1078 p. 131-142

Effect of Preservative on the Shelf Life of Yoghurt Produced from Soya Beans Milk

Uduak G. AKPAN[1], Abdullahi D. MOHAMMED[2] and Ibrahim AMINU[3]

Department of Chemical Engineering, Federal University of Technology, Minna, Nigeria
ugaekon@yahoo.co.uk, abduldyamoh@yahoo.com, ibrodya@yahoo.com

Abstract

This study concentrated on the effects of preservatives on shelf life of yoghurt produced from Soya beans milk. The yoghurt was produced by heating Soya beans milk slurry, cooled and incubated with starter culture. After the required yoghurt has been formed, sugar, flavour and preservatives were added. Study of the effect of preservatives revealed that Sodium benzoate preservative used at 20mg/ml give the best (optimum) preservation on both shelf and refrigeration storage for 15 and 21 days respectively. This is because the inhibitive ability of Sodium benzoate at lower temperature is higher than that of Potassium metabisulphate preservative. The study also revealed that 40mg/ml concentration of the combined preservatives gives the best (optimum) concentration level for both shelf and refrigeration storage with pH values of 3.92 and 4.01 respectively after 14 days fermentation. The preservatives concentration added are within the threshold values specified by Standard Organization of Nigeria (SON) and National Agency for Food Administration and Control (NAFDAC).

Keywords

Soya Beans; Yoghurt; Preservatives; Concentration; Fermentation; Culture.

Introduction

Yoghurt is a semi-fluid milk product, prepared from fresh whole or skimmed milk, boiled and concentrated by evaporation. The fermentation is caused by the addition of culture bacteria. The thickness of the product is the result of the acidification by the Lactic Acid Bacteria (LAB) [1].

Yoghurt appears as whitish liquid substance but in some cases, flavour is added to bring about alteration in the colour and taste. The transformation of milk into yoghurt with a starter culture composed of *streptococcus salivarus ssp. Thermophilus and Lactobacillus delbiueckil ssp*) [2]. *Bulgarius* can easily extend the shelf life of the milk from few days to weeks. Example of such post fermentation process includes heating, concentration, freezing and drying, but it is evident that such treatment will alter the characteristics of the end product [2].

Soya bean (Glycine Maxima), the primary material for Soya milk production has been identified to be one of the most important legumes of the tropics with high protein content. It is a potential food material that contains all essential amino acids that are very important for the proper development of the body, indeed Soya beans, has a higher content of Lysine in comparison to other plant proteins [3]. Soya bean when processed they give Soya beans milk which can be converted to yoghurt which is valuable protein supplement or substitute for adult and infant feeding [4]. Soya milk is lactose-free and can be consumed by the lactose-intolerant people as a substitute to milk [5].

Preservatives are added to improve yoghurt consistency. These are generally additives, which prolong the life span of foods and drinks by preventing micro-organisms attack. Technically, preservatives are chemicals used to poison micro-organisms and to prevent the food onto which it is added from fermentation and spoilage without causing any harmful effect to the person who consumed the food. The uses of chemical preservatives enhance food quality, reduce waste and enhance consumer acceptability [6].

These chemical preservatives are classified into three main types: Anti microbial (such as Benzoic acid, propionates, Di-methyl Pyrocirbonates), Antioxidant (such as Ascorbic acid, Butylated hydroxyl anisole), and Antibiotic (such as Oxy tetracycline, Ninsin and Lacto peroxidase). Other miscellaneous preservatives are Vinegar, Sorbet, Silica gel, Boric acid, Boras, Formaldehyde, Thiabendazole et cetera [6].

Ascorbic acid and its derivatives are added to milk and yoghurt to inhibit the rancidity and spoilage rate of the yoghurt. Likewise Ninsin is used in preserving yoghurt, as it prevent the growth of vegetable bacterial cells and end pores that are responsible for gas formation by *clostridium spp* which may cause the yoghurt packages to expand or burst. A culture of Lactoperioxidase is also used to preserve yoghurt because it damages the inner membrane of bacteria. Most preservatives are readily available since they can be synthesized in the laboratories [6].

This work is however aimed at studying the effect of preservatives on yoghurt produced from Soya beans; and majorly focuses on the following specific objectives:

- Production of the Soya milk from the Soya beans;
- Preparation of the starter culture;
- Conversion of the milk produced to yoghurt;
- Application of the preservatives on the yoghurt;
- Estimation of the best (optimum) concentration level required to preserve the yoghurt.

Methodology

Materials

Soya beans (Glycine Maxima) yellow seeds were purchased at modern Market, Minna, Niger State, Nigeria; a starter culture (developed from skimmed powdered milk); distilled water; flavouring agent; sugar and chemical preservatives (Sodium benzoate and Potassium metabisulphate).

Procedure for propagation of starter culture

0.6 kg skimmed powdered milk was weighed into a measuring cylinder. 3L clean water at ambient temperature was added to the skimmed milk and thoroughly stirred to give a homogeneous mixture. The mixture was heated to 90°C and held for 30 minutes at this temperature, following the inoculation of the milk. With the culture, the milk was then incubated at 42°C for a period of 12hrs.

Procedure for the production of yoghurt

7.0 kg of moth-free and mildew-free Soya beans grain was weighed and soaked in warm water for a period of 24h. At interval of 8h, the warm water was drained off and replaced with a new one to reduce the beanic flavour of the Soya beans grain. After soaking overnight, the beans was de-husked, washed with water and ground into paste. Three liters of clean water was added and thoroughly mixed to give slurry. The Soya beans milk was extracted by transferring the slurry to a clean white sieve cloth where filtration by suction took place. The beans dreg on the filter cloth was dried and used as livestock and poultry feed. The extracted milk was transferred into a pot and pasteurized or rather heated to 85°C for an hour and allowed to cool gradually to a temperature of about 42-45°C. The yellowish wad appeared on the surface was continually packed off using a clean spoon. The cooled homogenized milk was incubated with the already prepared starter culture. The mixture was stirred properly and kept to stand at a temperature of about 30-35^{o}C for a period of 24h. At the expiration of the incubation period, the output was yoghurt. At this juncture, sugar, flavour and preservatives are seldom added.

Procedure for addition of chemical preservatives

0.01g each of Sodium benzoate and Potassium metabisulphate was dissolved in 3mL of distilled water and shaken thoroughly inside a beaker 3mL of the solution was then added to 200mL of the prepared yoghurt. The yoghurt was stored in sample bottle with label on each bottle for easy identification of each sample.

Determination of pH of the sample

The pH meter was switched on and allowed to warm up for 20min. The pH was adjusted to neutral value by using distilled water at ambient temperature. The electrodes of the meter where then cleaned, dried and dipped into the different samples and the reading was noted. After the pH of the first sample was noted, the electrodes were re-washed with distilled water before being dipped into the second and subsequent samples until all the samples were tested.

Determination of viscosity of the sample

The viscosity of water at 27°C and 28°C were noted. The water was poured into a

flow-cup viscometer to the brim, while the discharge outlet was blocked with the index finger. This was so done to ascertain the time taken for the water to discharge on releasing. The average time taken for each sample was recorded. By using the method of linear interpolation, the viscosity of each sample was determined by comparing with standard viscosity of water at 27^{o}C and 28^{o}C respectively.

Results

The results obtained during the investigation on the effect of preservatives at different concentration of yoghurt produced from Soya beans milk kept on both shelf and refrigeration storage are tabulated in Table 1 to14.

Table 1. Results of sample preserved with Sodium benzoate at 10mg/ml on shelf storage

Days	pH	Viscosity Kg/m^2	Counts mg/ml	Temp °C	Color	Odor	Taste
1	4.4	1.572	3	31	White	Pleasant	Sweet
3	4.4	1.572	3	30			
5	4.34	1.624	4	27			
7	4.31	1.658	6	29			
9	4.31	1.671	7	27			
11	4.17	1.683	8	27			
13	4.04	1.699	9	29			
15	3.98	1.728	10	27			
17	3.8	1.749	11	29	Brownish white	Unpleasant	Sour
19	3.67	1.756	12	26			
21	3.59	1.774	12	26			

Table 2. Results of sample preserved with Sodium benzoate at 10mg/ml on refrigeration storage at 4^{o}C

Days	pH	Viscosity Kg/m^2	Counts mg/ml
1	4.4	1.572	3
3	4.4	1.572	3
5	4.43	1.572	3
7	4.37	1.559	3
9	4.37	1.611	4
11	4.3	1.621	5
13	4.23	1.629	6
15	4.14	1.634	6
17	4.01	1.639	7
19	3.84	1.645	8
21	3.79	1.648	10

- Sweet taste, pleasant odor and white coloration were maintained by the sample during the period under consideration.

Table 3. Results of sample preserved with Sodium benzoate at 20mg/ml on shelf storage

Days	pH	Viscosity Kg/m^2	Counts mg/ml	Temp ^{o}C	Color	Odor	Taste
1	4.4	1.572	3	31	White	Pleasant	Sweet
3	4.4	1.598	4	30			
5	4.34	1.612	5	27			
7	4.31	1.626	6	29			
9	4.31	1.635	6	27			
11	4.17	1.647	7	27			
13	4.04	1.652	8	29			Sour
15	3.98	1.66	10	28	Brownish white	Unpleasant	
17	3.8	1.678	11	29			
19	3.67	1.684	11	29			
21	3.59	1.67	12	26			

Table 4. Results of sample preserved with Sodium benzoate at 20mg/ml on refrigeration storage at 4^{o}C

Days	pH	Viscosity Kg/m^2	Counts mg/ml
1	4.4	1.572	3
3	4.4	1.572	3
5	4.37	1.57	4
7	4.36	1.593	4
9	4.33	1.597	5
11	4.38	1.623	6
13	4.31	1.617	7
15	4.26	1.693	8
17	4.2	1.701	9
19	4.13	1.743	10
21	4.07	1.764	10

- Sweet taste, pleasant odor and white coloration were maintained by the sample during the period under consideration.

Table 5. Results of sample preserved with Sodium benzoate at 40mg/ml on shelf storage

Days	pH	Viscosity Kg/m^2	Counts mg/ml	Temp ^{o}C	Color	Odor	Taste
1	4.4	1.572	3	31	White	Pleasant	Sweet
3	4.4	1.596	4	30			
5	4.37	1.603	5	27			
7	4.33	1.639	6	29			
9	4.24	1.647	6	27			
11	4.2	1.661	7	27			
13	4.07	1.678	9	29			
15	3.98	1.729	11	28	Brownish white	Unpleasant	Sour
17	3.91	1.743	11	29			
19	3.84	1.787	12	29			
21	3.65	1.853	12	26			

Table 6. Result of sample preserved with Sodium benzoate at 40mg/ml on refrigeration storage at 4°C

Days	pH	Viscosity Kg/m^2	Counts mg/ml
1	4.4	1.572	3
3	4.4	1.572	3
5	4.37	1.59	3
7	4.35	1.596	3
9	4.31	1.601	4
11	4.29	1.619	5
13	4.1	1.624	5
15	4.01	1.629	6
17	3.97	1.638	7
19	3.98	1.643	7
21	3.83	1.657	8

- Sweet taste, pleasant odor and white coloration were maintained by the sample during the period under consideration.

Table 7. Results of sample preserved with Potassium metabisulphate at 20mg/ml on shelf storage

Days	pH	Viscosity Kg/m^2	Counts mg/ml	Temp °C	Color	Odor	Taste
1	4.4	1.572	3	31	White	Pleasant	Sweet
3	4.35	1.572	4	30			
5	4.31	1.642	5	29			
7	4.27	1.629	6	27			
9	4.19	1.641	7	27			
11	4.13	1.678	8	29			
13	4.09	1.689	10	27			Sour
15	3.97	1.698	11	29	Brownish white	Unpleasant	
17	3.89	1.702	12	26			
19	3.79	1.719	13	29			
21	3.65	1.732	13	27			

Table 8. Results of sample preserved with Potassium metabisulphate at 20mg/ml on refrigeration storage at 4°C

Days	pH	Viscosity Kg/m^2	Counts mg/ml
1	4.4	1.572	3
3	4.4	1.572	3
5	4.37	1.589	3
7	4.29	1.59	4
9	4.22	1.612	5
11	4.17	1.642	6
13	4.11	1.661	6
15	4.08	1.715	7
17	4.03	1.729	8
19	4.0	1.763	9
21	3.93	1.781	9

- Sweet taste, pleasant odor and white coloration were maintained by the sample for 19 days, but brownish white color, unpleasant odor and sour taste were dictated on the 21st day.

Table 9. Results of sample preserved with Potassium metabisulphate at 40mg/ml on shelf storage

Days	pH	Viscosity Kg/m^2	Counts mg/ml	Temp °C	Color	Odor	Taste
1	4.4	1.572	3	31	White	Pleasant	Sweet
3	4.37	1.572	4	30			
5	4.34	1.61	5	29			
7	4.28	1.642	6	27			
9	4.2	1.661	7	27			
11	4.13	1.663	8	29			
13	3.96	1.697	10	27	Brownish white	Unpleasant	Sour
15	3.92	1.718	11	2			
17	3.86	1.772	12	26			
19	3.79	1.821	12	29			
21	3.64	1.867	13	27			

Table 10. Results of sample preserved with Potassium metabisulphate at 40mg/ml on refrigeration storage

Days	pH	Viscosity Kg/m^2	Counts mg/ml	Temp °C	Color	Odor	Taste
1	4.4	1.572	3	4	White	Pleasant	Sweet
3	4.4	1.572	3	,,			
5	4.37	1.572	4	,,			
7	4.34	1.593	4	,,			
9	4.33	1.607	5	,,			
11	4.26	1.635	5	,,			
13	4.13	1.647	6	,,	Brownish white	Unpleasant	Sour
15	4.01	1.657	7	,,			
17	3.99	1.687	8	,,			
19	3.8	1.63	9	,,			
21	3.74	1.736	9	,,			

Table 11. Sample preserved with combined preservatives at 10mg/ml on shelf storage

Days	pH	Viscosity Kg/m^2	Counts mg/ml	Temp °C	Color	Odor	Taste
1	4.4	1.572	3	29	White	Pleasant	Sweet
3	4.4	1.572	3	30			
5	4.33	1.598	4	29			
7	4.03	1.612	5	29			
9	4.39	1.628	7	28			
11	4.01	1.639	8	28			
13	4.27	1.648	10	28			
15	4.17	1.66	11	29	Brownish white	Unpleasant	Sour
17	3.83	1.678	12	29			
19	3.78	1.684	13	29			
21	3.69	1.692	14	29			

Table 12. Sample preserved with combined preservatives at 10mg/ml on Refrigeration storage at 4°C

Days	pH	Viscosity Kg/m^2	Counts mg/ml
1	4.4	1.572	3
3	4.4	1.572	3
5	4.4	1.572	3
7	4.37	1.585	4
9	4.3	1.592	5
11	4.29	1.598	5
13	4.18	1.603	6
15	4.14	1.608	7
17	4.11	1.614	9
19	4.06	1.637	10
21	3.91	1.656	11

- Sweet taste, pleasant odor and white coloration were maintained by the sample for 19 days, but brownish white color, unpleasant odor and sour taste were dictated on the 21st day.

Table 13. Sample preserved with combined preservatives at 40mg/ml on shelf storage

Days	pH	Viscosity Kg/m^2	Counts mg/ml	Temp °C	Color	Odor	Taste
1	4.4	1.572	3	31	White	Pleasant	Sweet
3	4.4	1.572	3	30			
5	4.37	1.643	5	29			
7	4.32	1.664	6	27			
9	4.17	1.678	7	27			
11	4.09	1.685	8	29			
13	3.97	1.709	9	27			
15	3.92	1.721	10	29			
17	3.87	1.756	11	26	Brownish white	Unpleasant	Sour
19	3.83	1.781	11	27			
21	3.71	1.821	12	29			

Table 14. Sample preserved with combined preservatives at 40 mg/ml on Refrigeration storage at 4°C

Days	pH	Viscosity Kg/m^2	Counts mg/ml
1	4.4	1.572	3
3	4.4	1.572	3
5	4.37	1.598	3
7	4.32	1.602	4
9	4.3	1.611	4
11	4.29	1.618	5
13	4.21	1.623	6
15	4.16	1.627	7
17	4.11	1.631	7
19	4.06	1.635	8
21	4.01	1.643	9

- Sweet taste, pleasant odor and white coloration were maintained by the sample during the period under consideration.

Discussion

It was observed that all the samples kept on refrigeration storage with preservatives at different concentrations maintained their color; odor and taste within the first seventeen days, and thereafter, some samples immediately changed their qualities. These may be as a result of rapid growth of bacteria as room temperature changes.

Parameters results obtained on the refrigeration storage of the sample preserved with 10,20,30,40 and 50mg/mL concentration of Sodium benzoate, appear almost similar with pH value ranging from 3.97-4.4, viscosity from 1.572-1.764Kg/m^2 and microbial count between 3 and 10 respectively. Perhaps, the sample color, odor and taste maintained their quality within the preservative period of twenty one days.

The samples preserved with Sodium benzoate at different concentration and kept on shelf storage maintained their quality within the first-fifteen days. After which a sour taste was observed. This, of course, is as a result of acid formation as the room temperature increases to 29^{o}C. In addition, the microbial count in the sample increased, indicating further deterioration of the yoghurt quality.

On refrigeration storage, the samples preserved with Potassium metabisulphate at different concentrations, maintained their properties within the first-nineteen days with decrease in the pH values. But at 20mg/mL the yoghourt maintained its properties for 21 days on refrigeration storage. This shows that other samples become more acidic as a result of fermentation which subsequently results into clotting or curdling (coagulation) of the samples. On shelf storage, the sample preserved with Potassium metabisulphate maintained their quality within fifteen days. Only sample preserved with 40mg/ml concentration of the preservative was noticed to change quality immediately after 13 days of storage, as the microbial count activity increases from 3-13mg/ml and the pH value decreases from 4.4-3.64 respectively.

The results obtained on refrigeration storage of the samples preserved with a mixture of Sodium benzoate and Potassium metabisulphate preservatives, revealed a slight change in the pH values from 4.4 to 4.08 within the same time. Only sample preserved with 40mg/mL

of the preservatives maintained it quality despite the slight changes in the pH value. On the other hand, the samples kept on shelf storage turned sour after 15 days with decrease in the pH value and increase in microbial count.

The samples kept on refrigeration storage with Sodium benzoate preservative, best preserved the yoghurt. This is as a result of anti-microbial activity of Sodium benzoate which results in low temperature that inhibits the growth of bacteria. However, on shelf storage, the room temperature makes the oxidation favorable for their replication and growth. In a broad sense, the preserved yoghurt should be stored at a low temperature in order to increase its life span.

Conclusions

Sodium benzoate preservatives used at 20mg/ml concentration gives the best (optimum) concentration on both shelf and refrigeration storage within 21 days. This clearly indicates that the inhibitive ability of Sodium benzoate is higher than that of Potassium metabisulphate. In addition, the combined preservatives (Sodium benzoate and Potassium metabisulphate) give the best concentration level for both shelf and refrigeration storage respectively.

The storage condition at low temperature and the proportions of the preservatives concentration increases the shelf life of the yoghurt samples above the recommended storage times (between 5-7 days). The chemical changes undergone by the yoghurt sample during storage gradually lead to loss in color, odor and taste. Moreover, the increased in acidity was due to decrease in pH that provides ideal environment for microorganisms to grow.

The fermentation of yoghurt especially on shelf storage samples was due to the oxidation of organic compounds present in the yoghurt by bacteria. The preservatives concentrations were within the threshold values specified by the Standard Organization of Nigeria (SON) and National Agency for Food and Drug Administration and Control (NAFDAC).

References

1. Adams M. R., Moss M. O., *Food Microbiology*, Cambridge U.K, Royal Society of Chemistry, p. 18, & 255-265, 1995.
2. Peterson S., Johnson M., *Food for the Orient*, 1st edition, McGraw Hill publishers, London, p. 34-42, 1978.
3. Ade-Omowaye B. I. O., Olajide J. O., Otunola E. T., Omotade V. A., *Effect of some processing parameters on the quality characteristics of Soya bean curd*, Science Focus, 2004, 7, p. 53-57.
4. Delia C., Herbert E., *Food facts*, A study of Food Nutrition, 1st edition, Macmillan Publishers Company, London, p. 101-114, 1986.
5. Okafor N., *Focus on Nutritional Values of Soya Beans*, Macmillan Publishers, Nigeria, p. 6-9, 1990.
6. Friezer W. C., *Contamination, Spoilage and Preservation of Yoghurt*, AVI Publishers, U.S.A., p. 13-29, 1989.