

# The Dynamics of Microorganisms in Dried Yogurt Storages at Different Temperatures

Jaruwan Chutrtong

**Abstract**—Yoghurt is a fermented milk product. The process of making yogurt involves fermenting milk with live and active bacterial cultures by adding bacteria directly to the dairy product. It is usually made with a culture of *Lactobacillus* sp. (*L. acidophilus* or *L. bulgaricus*) and *Streptococcus thermophilus*. Many people like to eat it plain or flavored and it's also use as ingredient in many dishes. Yogurt is rich in nutrients including the microorganism which have important role in balancing the digestion and absorption of the boy. Consumers will benefit from lactic acid bacteria more or less depending on the amount of bacteria that lives in yogurt while eating. When purchasing yogurt, consumers should always check the label for live cultures. Yoghurt must keep in refrigerator at 4°C for up to ten days. After this amount of time, the cultures often become weak. This research studied freezing dry yogurt storage by monitoring on the survival of microorganisms when stored at different temperatures. At 30°C, representative room temperature of country in equator zone, number of lactic acid bacteria reduced 4 log cycles in 10 week. At 40°C, representative temperature in summer of country in equator zone, number of lactic acid bacteria also dropped 4 log cycle in 10 week, similar as storage at 30°C. But drying yogurt storage at 40°C couldn't reformed to be good character yogurt as good as storage at 40°C only 4 week storage too. After 1 month, it couldn't bring back the yogurt form. So if it is inevitable to keep yogurt powder at a temperature of 40°C, yoghurt is maintained only up to 4 weeks.

**Keywords**— Dynamic, dry yoghurt, storage, temperature.

## I. INTRODUCTION

YOGHURT classified as dairy product refers to milk that is produced by fermentation with lactic acid bacteria [3], [7] such as *Streptococcus thermophilus*, *Lactobacillus* subsp. *bulgaricus*) or other sub-species [2]. When fermentation ends, Yogurt may have sold or may alter shape, texture, flavor or other properties [6]. Sometimes take to heat, dry, concentrate, sweeten, add additives or processed into a frozen yogurt. It's also filled concentrate *Lactobacillus acidophilus* inoculum in the yogurt after fermentation to make acidophilus yogurt. It's also used to make yogurt powder.

Drying or dehydration is a method of food preservation, commonly used. Reduce the humidity (moisture content) of food by dehydration. Freeze-dried or "lyophilization" is the drying process which drying in low temperature and high pressure condition. It can save nutritional values, structure, texture, color, smell and taste of product as close to fresh product. The advantage of lyophilized process is that the product has low damage [4]. Product structure is very porous. As a result, the product can be recovered (rehydration)

Jaruwan Chutrtong is with the Industrial Microbiology Department, Faculty of Science and Technology, Rajabhat Suansunandha University, Institute of Standards and Technology, Bangkok, CO 10300 Thailand (phone: 662-160-1143; fax: 662-160-1143; e-mail: jaruwan.ch@ssru.ac.th).

quickly. Therefore, this study investigated the yogurt storage process by studying the dynamics of microorganisms in dry yogurt storage at different temperatures, 4°C and 40°C. Moreover, also study the change of curd character when yogurt powder is rehydrated after storage to provide the storage time.

## II. PROCEDURE

### A. Making Yogurt

1. Warm pasteurized milk in water bath at 90°C (Do not let milk boiling because it will make denatured).
2. Remove milk from heat and reduce the temperature by flow cold water through the outside of the container or Immerse in ice until the temperature dropped to about 45°C.
3. Fill yogurt inoculum into 100ml of milk, 3-10%; stir gently to mix the ingredients.
4. Measure the initial pH before incubation. Close the container tightly. Incubate at 42-45°C for 4-6 hours.
5. Measure pH after fermentation and observe characters such as flavor, color, texture.

### B. Cell Counting

Spread plate method used for counting the number of cells in yogurt [1].

1. Pipette yogurt 1ml. Dilute with 9ml of distilled water, and make serial dilution.
2. Pipette 0.1ml of the dilution to culture medium, *Lactobacillus bulgaricus* agar and *Streptococcus thermophilus* agar [8].
3. Dip triangle glass rod in alcohol, then fire and wait to cool. Use glass rod to spread dilution on surface of medium until dry.
4. Incubate medium at 40-45°C for 48 hours in anaerobic jar which have Anaerocult ® A. Recorded colony numbers.

### C. Yogurt Drying (to Make Yogurt Powder)

1. Put yogurt 10 grams in sterile tube.
2. Froze at -20 degrees Celsius.
3. Dry with freeze- dry machine takes about 6-7 hours to make yogurt powder.
4. Weigh the remaining.
5. Closed tightly package and wrapped with parafilm. Then store at 30°C and 40°C in incubator.

### D. Recombine Yogurt Powder

1. Recombine stored yogurt powder from the drying to the amount of 10 grams before the freeze dry by mix UHT milk to yogurt powder.

2. Stir gently until homogeneous. Incubate at 42-45°C for 4-6 hours [9].
3. Measure pH, count the number of microorganism and observe character such as flavor, color, texture. Compare with initial yogurt.

### III. RESULT

#### A. Number of Survivors Microorganism

TABLE I

NUMBER OF LACTIC ACID BACTERIA AT START TIME OF REFORMING

Week	Number of Microorganism (cfu/ml)	
	Storage at 30°C	Storage at 40°C
0	$2.0 \times 10^8$	$2.0 \times 10^8$
1	$1.6 \times 10^7$	$1.56 \times 10^7$
2	$1.24 \times 10^7$	$1.2 \times 10^7$
3	$1.04 \times 10^7$	$1.02 \times 10^7$
4	$8.8 \times 10^6$	$8.6 \times 10^6$
5	$7.6 \times 10^6$	$7.2 \times 10^6$
6	$6.3 \times 10^5$	$6.0 \times 10^5$
7	$3.7 \times 10^5$	$3.0 \times 10^5$
8	$4.6 \times 10^4$	$4.4 \times 10^4$
9	$2.3 \times 10^4$	$2.0 \times 10^4$
10	$1.9 \times 10^4$	$1.8 \times 10^4$

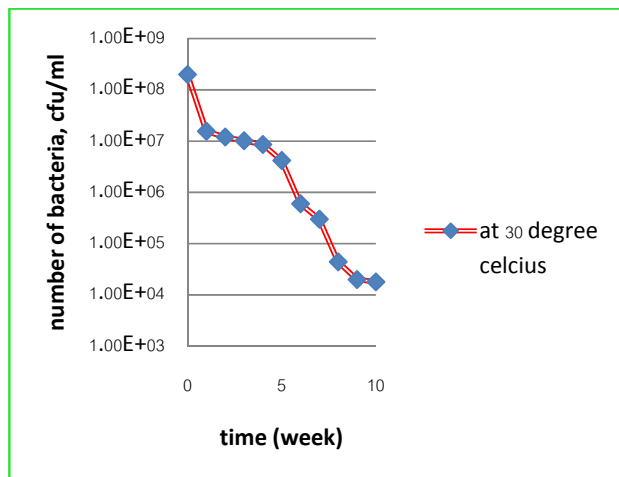


Fig. 1 Number of Lactic acid bacteria storage at 30°C

#### B. Rate of Reforming

TABLE II

NUMBER OF LACTIC ACID BACTERIA AND pH BETWEEN REFORMING AFTER STORAGE 1 WEEK

Time (hour)	Storage at 30°C		Storage at 40°C	
	pH	Number (cfu/ml)	pH	Number (cfu/ml)
0	6	$1.6 \times 10^7$	6	$1.56 \times 10^7$
1	6	$1.8 \times 10^7$	6	$1.72 \times 10^7$
2	5.5	$2.64 \times 10^7$	5.5	$2.35 \times 10^7$
3	5.5	$5.1 \times 10^7$	5.5	$4.4 \times 10^7$
4	5.5	$7.51 \times 10^7$	5.5	$7.34 \times 10^7$
5	5	$9.22 \times 10^7$	5	$8.6 \times 10^7$
6	4	$1.26 \times 10^8$	4	$1.02 \times 10^8$

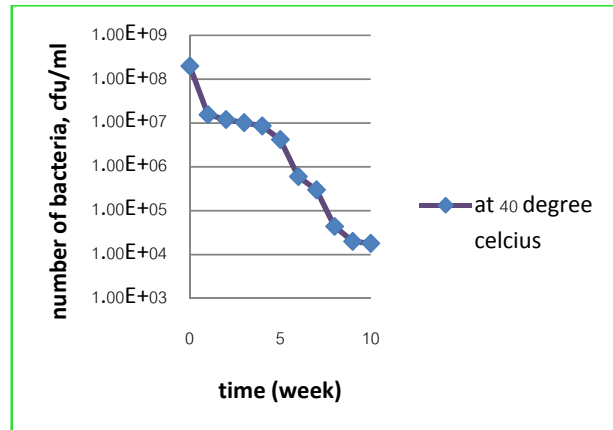


Fig. 2 Number of Lactic acid bacteria storage at 40°C

TABLE III

NUMBER OF LACTIC ACID BACTERIA AND pH BETWEEN REFORMING AFTER STORAGE 2 WEEK

Time (hour)	Storage at 30°C		Storage at 40°C	
	pH	Number (cfu/ml)	pH	Number (cfu/ml)
0	6	$1.24 \times 10^7$	6	$1.2 \times 10^7$
1	6	$1.7 \times 10^7$	6	$1.62 \times 10^7$
2	5.5	$2.2 \times 10^7$	5.5	$2.05 \times 10^7$
3	5.5	$3.05 \times 10^7$	5.5	$2.7 \times 10^7$
4	5.5	$3.9 \times 10^7$	5.5	$3.6 \times 10^7$
5	5	$5.54 \times 10^7$	5	$5.2 \times 10^7$
6	5	$6.66 \times 10^7$	5	$6.45 \times 10^7$
7	4	$8.6 \times 10^7$	4	$7.0 \times 10^7$

TABLE IV

NUMBER OF LACTIC ACID BACTERIA AND pH BETWEEN REFORMING AFTER STORAGE 3 WEEK

Time (hour)	Storage at 30°C		Storage at 40°C	
	pH	Number (cfu/ml)	pH	Number (cfu/ml)
0	6	$1.04 \times 10^7$	6	$1.02 \times 10^7$
1	6	$1.2 \times 10^7$	6	$1.1 \times 10^7$
2	5.5	$1.42 \times 10^7$	5.5	$1.34 \times 10^7$
3	5.5	$2.7 \times 10^7$	2.4	$2.4 \times 10^7$
4	5.5	$3.5 \times 10^7$	5.5	$3.0 \times 10^7$
5	5	$5.7 \times 10^7$	5	$4.3 \times 10^7$
6	5	$8.2 \times 10^7$	5	$5.5 \times 10^7$
7	4.5	$9.1 \times 10^7$	4.5	$7.6 \times 10^7$
8	4	$1.07 \times 10^8$	4	$8.2 \times 10^7$

TABLE V

NUMBER OF LACTIC ACID BACTERIA AND pH BETWEEN REFORMING AFTER STORAGE 4 WEEK

Time (hour)	Storage at 30°C		Storage at 40°C	
	pH	Number (cfu/ml)	pH	Number (cfu/ml)
0	6	$8.8 \times 10^6$	6	$8.6 \times 10^6$
1	6	$1.047 \times 10^6$	6	$1.02 \times 10^6$
2	6	$2.6 \times 10^6$	6	$2.16 \times 10^6$
3	5.5	$3.3 \times 10^7$	5.5	$3.25 \times 10^7$
4	5.5	$3.95 \times 10^7$	5.5	$4.44 \times 10^7$
5	5.5	$5.2 \times 10^7$	5	$5.0 \times 10^7$
6	5	$6.7 \times 10^7$	5	$5.22 \times 10^7$
7	5	$7.0 \times 10^7$	5	$6.6 \times 10^7$
8	4.5	$7.94 \times 10^7$	4.5	$7.2 \times 10^7$
9	4	$8.6 \times 10^7$	4	$9.4 \times 10^7$

TABLE VI  
NUMBER OF LACTIC ACID BACTERIA AND pH BETWEEN REFORMING AFTER  
STORAGE 5 WEEK

Time (hour)	Storage at 30°C		Storage at 40°C	
	pH	Number (cfu/ml)	pH	Number (cfu/ml)
0	6	$7.6 \times 10^6$	6	$7.2 \times 10^6$
1	6	$8.0 \times 10^6$	6	$7.6 \times 10^6$
2	6	$9.2 \times 10^6$	6	$7.9 \times 10^6$
3	5.5	$9.7.6 \times 10^6$	5.5	$8.2 \times 10^6$
4	5.5	$1.2 \times 10^7$	5.5	$8.6. \times 10^6$
5	5	$1.46 \times 10^7$	5.5	$9.0 \times 10^6$
6	5	$2.0 \times 10^7$	5	$9.2 \times 10^6$
7	5	$2.4 \times 10^7$	5	$9.4 \times 10^6$
8	5	$2.6 \times 10^7$	5	$1.0 \times 10^7$
9	4.5	$3.5 \times 10^7$	4.5	$1.35 \times 10^7$

TABLE VII  
NUMBER OF LACTIC ACID BACTERIA AND pH BETWEEN REFORMING AFTER  
STORAGE 6 WEEK

Time (hour)	Storage at 30°C		Storage at 40°C	
	pH	Number (cfu/ml)	pH	Number (cfu/ml)
0	6	$6.3 \times 10^5$	6	$6.0 \times 10^5$
1	6	$6.6 \times 10^5$	6	$6.2 \times 10^5$
2	6	$7.2 \times 10^5$	6	$6.5 \times 10^5$
3	6	$7.7 \times 10^5$	6	$6.9 \times 10^5$
4	5.5	$9.0 \times 10^5$	5.5	$7.1. \times 10^5$
5	5.5	$1.12 \times 10^6$	5.5	$7.3 \times 10^5$
6	5.5	$1.3 \times 10^6$	5.5	$8.0 \times 10^5$
7	5	$1.5 \times 10^6$	5	$8.2 \times 10^5$
8	5	$1.74 \times 10^6$	5	$8.6 \times 10^5$
9	5	$1.9 \times 10^6$	5	$9.1 \times 10^5$

TABLE VIII  
NUMBER OF LACTIC ACID BACTERIA AND pH BETWEEN REFORMING AFTER  
STORAGE 8 WEEK

Time (hour)	Storage at 30°C		Storage at 40°C	
	pH	Number (cfu/ml)	pH	Number (cfu/ml)
0	6	$4.6 \times 10^4$	6	$4.4 \times 10^4$
1	6	$4.7 \times 10^4$	6	$4.5 \times 10^4$
2	6	$4.9 \times 10^4$	6	$4.7 \times 10^4$
3	6	$5.2 \times 10^4$	6	$5.2 \times 10^4$
4	5.5	$7.1 \times 10^4$	5.5	$5.9 \times 10^4$
5	5.5	$9.8 \times 10^4$	5.5	$8.8 \times 10^4$
6	5.5	$3.8 \times 10^5$	5.5	$4.0 \times 10^5$
7	5.5	$6.4 \times 10^5$	5.5	$5.9 \times 10^5$
8	5	$6.9 \times 10^5$	5	$6.1 \times 10^5$
9	5	$7.1 \times 10^5$	5	$6.3 \times 10^5$

TABLE IX  
NUMBER OF LACTIC ACID BACTERIA AND pH BETWEEN REFORMING AFTER  
STORAGE 10 WEEK

Time (hour)	Storage at 30°C		Storage at 40°C	
	pH	Number (cfu/ml)	pH	Number (cfu/ml)
0	6	$1.9 \times 10^4$	6	$1.8 \times 10^4$
1	6	$2.1 \times 10^4$	6	$1.8 \times 10^4$
2	6	$3.0 \times 10^4$	6	$2.0 \times 10^4$
3	6	$4.3 \times 10^4$	6	$2.9 \times 10^4$
4	5.5	$6.1 \times 10^4$	5.5	$5.9 \times 10^4$
5	5.5	$6.6 \times 10^4$	5.5	$6.3 \times 10^4$
6	5.5	$7.0 \times 10^4$	5.5	$6.5 \times 10^4$
7	5.5	$7.3 \times 10^4$	5.5	$6.9 \times 10^4$
8	5.5	$7.8 \times 10^4$	5.5	$7.1 \times 10^4$
9	5	$3.3 \times 10^5$	5.5	$8.1 \times 10^4$

#### IV. CONCLUSION

From the results found that after dried yogurt by freeze drying and kept it in incubator, bacteria in yogurt which storage at 30 and 40°C decreased rapidly in the first week. The number of microorganisms was down 2 log cycle in 4 week. Microorganisms continual decreased in week 5-10. At the end of week 10<sup>th</sup> lactic acid bacteria decreased 4log cycle.

When reformed yogurt powder stored at 30 and 40°C, yogurt still have good character in 1- 4 week. They have sour taste, acetaldehyde smell, and smooth - stick together sedimentary wedge which is close to common yogurt. pH is approximately 4 but fermentation time is longer as a week of storage. After 4 week, yogurt from yogurt powder starts to have separate cluster. In the 5<sup>th</sup> week curd is loosely and has weakly flavor of yogurt and high pH. From 6<sup>th</sup> to 10<sup>th</sup> week reformed yogurt did not set curd. It could be only thicken milk. The results were identical in both samples which storage at 30 and 40°C.

Experimental results indicated that storing yogurt powder at 30 and 40°C made microorganisms decreased significantly. Consumers will benefit from yogurt more or less depending on the amount of lactic acid bacteria that lives in yogurt while eating [5], [7]. Thereby, Keep yogurt in dry conditions should be stored at low temperatures. But if it is necessary to store at high temperatures, it is good quality just not more than one month.

#### ACKNOWLEDGMENT

The author would like to thank Suan Sunandha Rajabhat University, Bangkok, Thailand for providing fund, necessary equipment, and laboratory area.

#### REFERENCES

- [1] AOAC, *Official methods of Analysis of AOAC International*, 17th ed. AOAC International. Strunk, W., Jr., & White, E. B. (1979), *The elements of style* (3rd ed.), New York: MacMillan, (2000).
- [2] E.S. Bautista, R.S Dahiya and M.L. Speck, Identification of Compound Symbiotic Growth of *Streptococcus thermophilus* and *Lactobacillus bulgaricus* in milk, *J. of Dairy Res.* 33: 299-307. (1966).
- [3] H.C. Deeth and A.Y. Tamime, Yoghurt: Nutritive and therapeutic aspects, *J. Food Protect.*44: 78-86. 1981.
- [4] L.M. Medina and R. jordano, Survival of constitutive in commercially fermented milk containing bifido- bacteria during refrigerated storage, *J.Food Prot.* 56: 731-733. 1994.
- [5] M. Aquirre and M.D. Collins, A review: Lactic acid bacteria and human clinical infection. *J. App.Bacterio.* 75: 95-107. 1993.
- [6] M.I. Gurr, Health and nutrition aspects of dairy products: An up-to-the-minute report, *Food Australian*, 44(9): 421-425. 1992.
- [7] O. Orihara, I. Sakauchi and Y. Nakazawa, Type M. Young, Types and standards for fermented milk and lactic drinks, pp.1-16. In Y. Nakazawa and A. Hosano(eds.), translated by B.W. Howells, Functions of Fermented Milk. *Elsevier Applied Science USA*. 1992.
- [8] R.M. Atlas, *Handbook of Microbiological Media for the Examination of Food*, CRD Press, Inc., USA. 310 p., 1995.
- [9] W.W. Hamann and E.H. Marth, Survival of *Streptococcus thermophilus* and *Lactobacillus bulgaricus* in commercial and experimental yoghurts. *J. Food Prot*, 47: 781-786. 1984.