#### CHAPTER IV

#### RESULT AND DISCUSSION

## 4.1 Microbiological, chemical, organoleptic and enzymatic qualities of raw and pasteurized milk

4.1.1 The microbiological properties of raw and pasteurized milk.

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The microbiological properties of raw and

pasteurized milk from each dairy were analysed and shown in Table 4.1

	DAIRIES	RAW MILK		PASTEURIZED MILK			ĸ		
		1	2	3	AVERAGE	1	2	3	AVERAGE
log no. of	A	5.447	5.677	5.531	5.552	2.000	2.041	2.079	2.040
standard plate	В	5.663	5.748	5.954	5.778	3.000	2.556	4.519	3.358
counts (cfu/ml)	с	6.919	5.204	4.954	5.692	3.041	3.162	4.045	3.416
	D	5.978	6.447	5.198	5.874	4.033	4.265	3.672	3.757
	E	5.954	5.786*	5.491	5.744	3.954	2.845	3.204	3.334
log no. of	A	5.215	5.633	5.531	5.460 <sup>a</sup>	1.311*	1.460	0.845	1.205
psychrotrophic	в.	5.544	6.3 69	4.778	5.564 <sup>a</sup>	2.236	2.568	3.398	2.734
counts (cfu/ml)	с	5.898	5.279	5.415	5 .53 1 <sup>a</sup>	1 .3 01	2.079	3.322	2.23 4
	D	4.699	3.000	3.000	3.566 <sup>b</sup>	3.330	3.651	2.079	3.020
	Е	5.681	5.968	4.903	5.517 <sup>a</sup>	3 .3 16	1.000	1.000	1.772
log no.of coliform	A	3.979	2.477	3 .3 80	3.279	nil	nil	nil	
counts (cfu/ml)	в	3.352	3.135*	3.362	3.283	0.114	-	1.820	
	с	3.484	3.000	2.699	3.061	nil	0.3 01	1.826	
	D	3.602	3.477	3.507*	3.529	1.544	-	1.000	
	E	3.813	4.544	4 .3 22	4.226	nil	nil	0.477	
log no.of lactic	A	4.477	5.265	4.638	4.793 <sup>a</sup>	0.362	0.778	0.574	0.571
acid bacteria	в	3.889	3.863	3.602	3.785 <sup>b</sup>	0.903	1.5 05	2.982	1.797
(cfu/ml)	c	4.204	4.477	4.833	4.505 <sup>a</sup>	0.602	0.568	1.792	0.987
	D	4 3 72	4.813	4.255	4.480 <sup>a</sup>	0.904*	1.778	1.3 01	1.3 28
	Е	4.724	4.623	4.586*	4.644 <sup>a</sup>	0.778	0.699	1.476*	0.984

Table 4.1 Microgiological properities of raw and fresh pasteurized milk

\* = missing value (determined using statistical analysis as shown in Appendix E 1 due to error in the experiment)

= error in the experiment

nil = negative result in 1 ml of milk

The experiment was conducted by using randomize complete block design (RCBD)

(see appendix E)

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The difference of mean values of each type among dairies were determined by using Duncan's multiple range test (DMRT) at 95% significant level with different alphabets. (see appendix E)

From the results, it is seen that the microbiological qualities of raw milk collected can be summarized as below.

total bacterial counts	10 <sup>5</sup> cfu/ml
psychrotrophic counts	10 <sup>5</sup> cfu/ml
coliform counts	10 <sup>4</sup> cfu/ml
lactic acid bacterial counts	$10^3  \text{cfu/ml}$

The different quantity of psychrotrophic and lactic acid bacterial counts of raw milk among dairies was found. This was due to the different method of handling and transportation of milk. It could be implied that using an aluminium can of dairy D, raw milk supplier for dairies B and E can cause more contamination than using a truck tanker of dairy C, raw milk supplier for dairy A.

For pasteurized milk, results of dairy A showed the lowest counts for all items. This was of course the good hygiene and sanitation conditions. Using cream separator and deodorizing process might be another reason helped to reduce the microbiological content. It was also observed that the good hygiene and sanitation of dairy E resulted the lowest count of coliform bacteria.

When considering the coliform count which is generally regarded as an index of good hygiene and sanitation, the results revealed that dairies B, C and D should give more attention in sanitation practising.

#### 4.1.2 Chemical properties of raw and pasteurized milk

The composition, titratable acidity and somatic cell count (SCC) of raw and pasteurized milk from each dairy were analyzed by Milko Scan 104 and Fossomatic 90, the results were shown in Table 4.2

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	DAIRIES		RAW MILK			PASTEURIZED MILK			
		1	2	3	AVERAGE	1	2	3	AVERAG
	A	4.60	4.49	4.33 .	4.47ª	3.30	3.29	3.40	3.11 <sup>°</sup>
Fat (%)	в	4.14	3.96	4.63	4.24	4.03	4.45	4.49	4.32 <sup>ab</sup>
	c	4.51	3.21	3.95	3.89 <sup>a</sup>	4.53	4.51	4.52	4.52
	. D	5.38	5.91	5.59	5.63 <sup>b</sup>	4.14	4.13	4.07	4.11 <sup>b</sup>
	E	4.35	4.31	4.87	4.518	3.84	4.24	4.30	4.13 <sup>b</sup>
	A	3.32	3.30	3.30	3.31ª	3.32	3.29	3.27	3.29
Protein (%)	в	3.20	3.30	3.33	3.28ª	3.21	3.24	3.26	3.24
	с	3.26	3.36	3.16	3.26ª	3.25	3.35	3.38	1.33
	D	3.57	3.68	3.76	3.67 <sup>b</sup>	3.30	3.30	3.24	3.28
	E	3.26	3.30	3.40	3.32ª	3.01	3.34	4.34	3.56
	x	4.67	4.65	4.56	4.63	4.69	4.64	4.54	4.62
Lactose (%)	В	4.72	4.73	4.83	4.76	4.72	4.69	4.70	4.70
	с	4.62	4.64	4.45	4.57	4.58	4.57	4.66	4.60
	D	4.96	4.74	4.74	4.81	4.76	4.74	4.71	4.74
	E	4.73	4.67	4.92	4.77	4.31	4.72	4.74	4.59
1	۸	86.3	86.5	86.8	86.53 <sup>b</sup>	87.5	87.8	87.8	87.70
Water (%)	В	86.7	86.9	87.3	86.97 <sup>ab</sup>	86.7	86.7	86.7	86.70
	с	86.6	87.8	87.9	87.43 ª	86.4	86.4	85.9	86.21
	D	86.3	85.1	84.6	85.33 C	87.3	86.8	86.7	86.91
	E	86.6	87.1	87.2	86.97 <sup>ab</sup>	87.5	86.9	86.7	87.03
	٨	13 .3 1	13.21	12.90	13.14 <sup>a</sup>	12.06	12.00	11.95	12.00 <sup>C</sup>
Total solid	В	12.73	12.75	12.96	12.81ª	12.68	13.08	13.10	12.95 ab
(%)	с	13.17	13.18	12.02	12.79 <sup>a</sup>	13.16	13.17	13.30	13.21ª
	D	14.24	14.99	14.81	14.68 <sup>b</sup>	12.79	12.83	12.81	12.81 ab
	E	12.96	12.86	13.17	13.00 <sup>8</sup>	11.90	12.96	13.02	12.63 b
	A	8.72	8.69	8.59	8.67 <sup>8</sup>	8.74	8.83	8.52	8.70
folid not	В	8.67	8.77	8.75	8.73 <sup>a</sup>	8.67	8.65 .	8.67	8.66
at (%)	c	8.68	8.71	8.29	8.56 <sup>a</sup>	8.64	8.67	8.77	8.69
	D	9.14	9.10	9.24	9.16 <sup>b</sup>	8.77	8.73	8.71	8.74
	E	8.71	8.68	8.86	8.75 <sup>a</sup>	8.05	8.77	8.78	8.53
itratable cidity	A	0.16	0.16	0.15	0.16	0.15	0.16	0.15	0.15
lactic acid)	В	0.16	0.17	0.18	0.17	0.16	0.17	0.17	0.17
· lactic acid)	с	0.16	0.16	0.15	0.16	0.16	0.14	0.16	0.15
	D	0.17	0.16	0.17	0.17	0.26	0.15	0.15	0.15
	E	0.16	0.17	0.17	0.17	0.15	0.17	0.16	0.16
omatic cell	Α	7.17×10 <sup>5</sup>	5.4×10 <sup>5</sup>	8.83 ×105	7.13×10 <sup>5</sup>	J.79x10 <sup>5</sup>	2.53×10 <sup>5</sup>	3 x 10 <sup>5</sup>	3.11×10 <sup>5 be</sup>
ilk	в	4.93×10 <sup>5</sup>	5.21×10 <sup>5</sup>	6.09×105	5.41x10 <sup>5</sup>	5.0x10 <sup>5</sup>	4.92×10 <sup>5</sup>	4.16x10 <sup>5</sup>	4.69×10 <sup>5 al</sup>
	с	3.96×105	8.78×10 <sup>5</sup>	3.78x10 <sup>5</sup>	5.58x10 <sup>5</sup>	9x10 <sup>4</sup>	4.1×10 <sup>4</sup>	1.75 x10 <sup>5</sup>	2.25×10 <sup>5</sup> C
1	D	2.43 x10 <sup>5</sup>	5.26x10 <sup>5</sup>	1.626x10 <sup>5</sup>	7.98×10 <sup>5</sup>	5.09x10 <sup>5</sup>	7.55×105	4.79×10 <sup>5</sup>	5.81x10 <sup>5 a</sup>
· · /	E	4.94z10 <sup>5</sup>	6.72x10 <sup>5</sup>	6.29x10 <sup>5</sup>	5.98×10 <sup>5</sup>	5.8x10 <sup>4</sup>	1.79x10 <sup>5</sup>	1.14×10 <sup>5</sup>	2.91x10 <sup>5 bc</sup>

Table 4.2 Chemical composition and somatic cell count of raw and pasteurized milk from various dairies.

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The experiment was conducted by using RCBD

The difference of mean values of each item among dairies were determined by using DMRT at 95% significant level with different alphabets.

• = \*

From the results, the average compositions of raw milk from various dairies were almost the same except the milk from dairy D which showed higher percentage of fat, protein and lactose. The average composition of pasteurized milk among dairies were different especially fat content, but milk from all dairies were within the standard for ready-to-drink milk set by Ministry of Public Health no. 26, B.E.2522 (14).

The titratable acidity of raw and pasteurized milk from each dairy were not significantly different. It was in the range of 0.15-0.17% as lactic acid.

The somatic cell counts of raw milk among dairies were not significantly different at 95% level but those of pasteurized milk were significantly different at 95% level. Both raw and pasteurized milk had somatic cell counts in the level of  $10^5$  cells/ml. This is rather high compared to the limitation of Norway (not exceed  $2.5 \times 10^5$  cells/ml)(22), but it seemed to have no effect on mastitis and composition change.

#### 4.1.3 Organoleptic properties of pasteurized milk

The color, odor, flavor, mouthfeel and overall quality of pasteurized milk from each dairy were evaluated and results shown in Table 4.3

#### DAIRIES Scores Properties 1 2 3 Mean (scores) 3.0 2.5 3.0 2.83 A 2.5 2.9 2.63 2.5 color в 2.8 2.80 С 2.9 2.7 (1-3) 2.5 2.9 2.80 3.0 D 2.8 2.87 2.8 3.0 Е 7.0 7.0 6.0 6.67 A 6.8 6.43 6.0 6.5 odor В 6.60 С 6.8 6.5 6.5 (1-7)7.0 5.5 6.17 6.0 D 6.3 · 5.97 5.8 5.8 Е 7.0 6.77 6.3 7.0 Α flavor В 6.0 6.5 6.0 6.17 6.57 (1 - 7)6.5 6.8 6.4 С 6.8 5.8 6.30 6.3 D 6.3 6.9 6.8 6.67 E 2.5 3.0 2.5 2.67 A mouthfeel 2.2 2.5 2.8 . 2.50 В 2.70 (1-3) С 2.8 2.8 2.5 2.6 2.7 2.5 2.60 D Е 2.5 2.6 2.7 2.60 8.20 8.25 7.50 7.98 Α overall 7.50 7.00 7.25 7.25 в quality C 8.50 8.00 8.50 8.33

#### Table 4.3 Organoleptic properties of raw and pasteurized milk

from various dairies.

The experiment was conducted by using RCBD

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(1-9)

There is mo significant difference of each property considered.

7.50

8.00

8.50

7.75

7.75

8.25

7.92

8.00

The average organoleptic properties of pasteurized milk from various dairies can be summarized as shown in Table 4.4

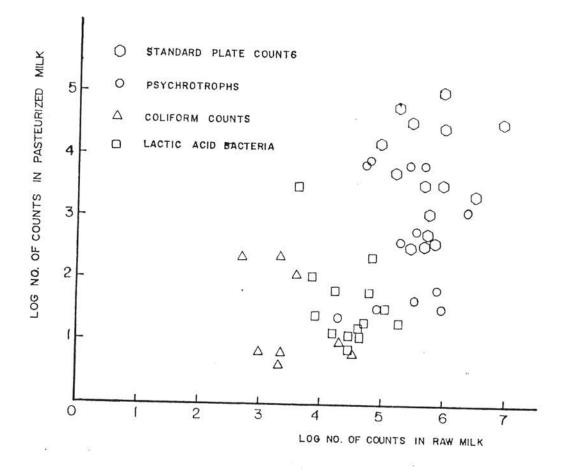
Table 4.4 Average scores of organoleptic properties of	Table 4.4	Average	scores	of	organoleptic	properties c	of
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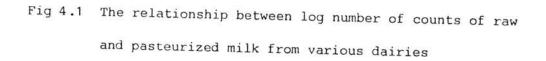
pasteurized milk

Properties	Mean scores
color (1-3)	2.79
odor (1-7)	6.37
flavor (1-7)	6.50
mouthfeel (1-3)	2.61
Overall quality (1-9)	7.90

4.1.4 Relationship of counts between raw and pasteurized milk

Log no.of counts (Standard plate count, psychrotrophic count, coliform count and lactic acid bacterial count) of raw milk were plotted against log no.of counts of pasteurized milk as shown in Fig. 4.1. The results shown no correlations of counts between raw and pasteurized milk. It might be from the uniformity of microbiological quality of raw milk. If its quality was different, the correlations of counts between raw and pasteurized milk might be found.





#### 4.1.5 Enzymatic properties of raw and pasteurized milk

No protease was observed in both raw and pasteurized milk while the presence of lipase was observed in raw milk and remain constant in pasteurized milk as shown in Table 4.5

Table 4.5	Protease	and	lipase	in	raw	and	pasteurized	milk	from
	various d	lairi	ies.						

		D	iamete	r of d	clear Z	one (m	m.)
Enzymes	DAIRIES	R	AW MIL	к	PASTEURIZED MILK		
12		1	2	3	1	2	3
	A	- -	-	-	-	-	-
protease	в	-	-	Œ	Ξ	-	-
	с	-	-	-		-	
	D	1.000	- *	-	-	-	
	Е	-	-		-	-	-
	A	6.0	5.5	*	6.0	6.0	*
lipase	в	6.0	*	*	6.0	*	*
	С	6.0	*	*	6.0	*	*
	D	*	*	*	*	*	*
	E	*	*	*	*	*	.*

- = negative result

\* = Not analysed because of the shortage of reagent

It can be said that, raw and pasteurized milk in Thailand had no problem related with protease but the results of lipase in pasteurized milk still in doubt. This is due to the experiment of Luhtala & Antila in 1968 who pointed out that HTST pasteurization generally believed to inactivate endogenous milk lipase completely while the report of Harper and Gould in 1959 observed that 15% of milk lipase activity remained after pasteurization (29).

### 4.2 Microbiological, chemical, organoleptic and enzymatic qualities of pasteurized milk at various storage temperatures and times.

From the experiment, five dairies were divided into three groups according to their packaging materials and sanitation conditions. It is because these two factors affected the counts and shelf-life of pasteurized milk at various storage temperatures and times (37). These three groups were :

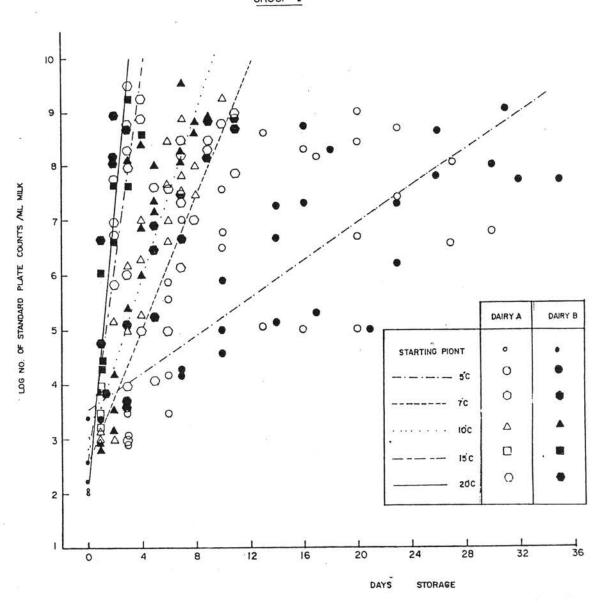
Group I : Dairy A dnd B-using paperboard (carton) as packaging material.

Group II : Dairy C and D-using polyethylene sachet as packaging material and fair sanitation condition.

Group III : Dairy E-using polyetheylene sachet and having a good sanitation condition.

4.2.1 <u>Relationship between microbiological counts and</u> storage times at various temperatures

For all groups of dairies, every type of counts (standard plate count, psychrotrophic count, coliform count and lactic acid bacterial count) increased linearly with storage time. At the same storage-time, there was an increasing of count at higher storage temperature as shown in Fig. 4.2 a, b, c; Fig 4.3 a, b, c; Fig 4.4 a, b and Fig 4.5 a, b and c. The linear correlation coefficients were highly significant as shown in Table 4.6, 4.7, 4.8 and 4.9. It showed that microbiological growth rate corresponded with the first order autocatalytic chemical reaction (33).





a The relationship between log number of standard plate count and storage time of pasteurized milk at various storage temperatures

GROUP I

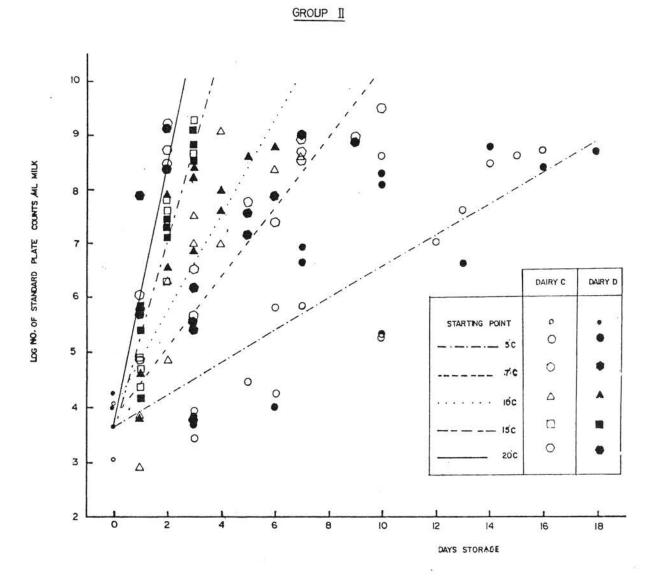
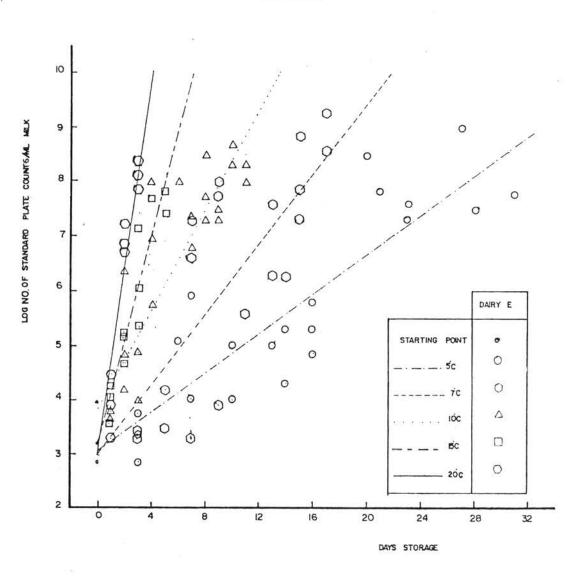


Fig 4.2 b The relationship between log number of standard plate count and storage time of pasteurized milk at various storage temperatures



GROUP II

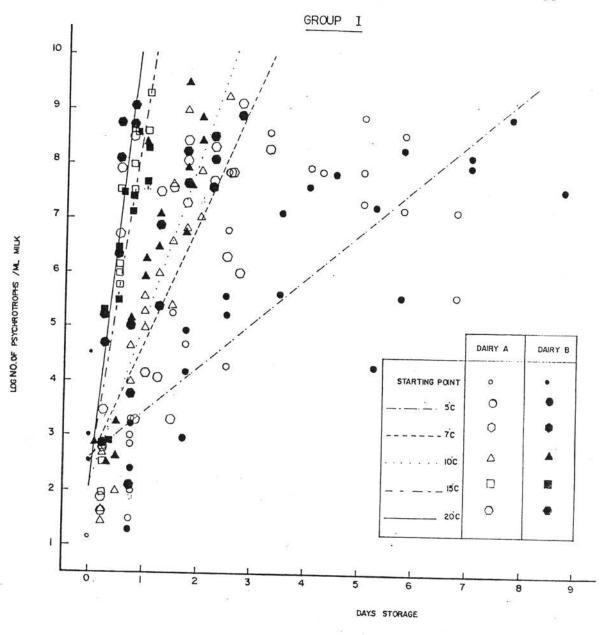
Fig 4.2 c The relationship between log number of standard plate count and storage time of pasteurized milk at various storage temperatures .

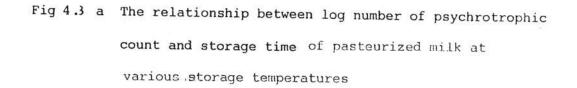
Sample group	Storage temp.('C)	Linear correlation coefficients	Significance (P)
I	5	0.78	<0.01
	7	0.92	<0.01
	10	0.92	<0.01
18	15	0.94	<0.01
	20	0.93	<0.01
II	5	0.87	<0.01
	7	0.96	<0.01
	10	0.87	<0.01
	15	0.96	<0.01
	20	0.93	<0.01
III	5	0.89	<0.01
	7	0.83	<0.01
	10	0.89	<0.01
	15	0.95	<0.01
	20	0.95	<0.01

Table 4.6 Linear correlation coefficients between log.no.of standard plate

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counts and storage time of pasteurized milk at various temperatures.





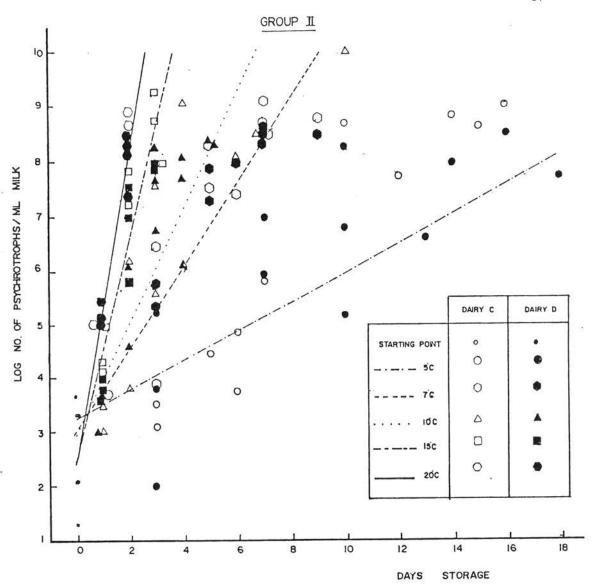
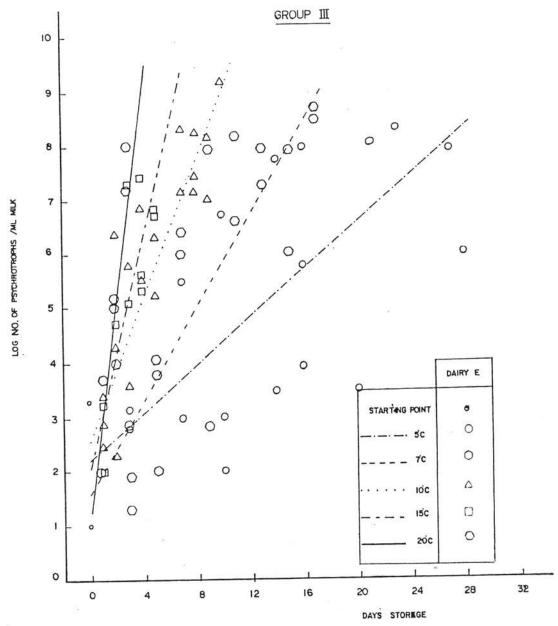


Fig 4.3 b The relationship between log number of psychrotrophic count and storage time of pasteurized milk at various storage





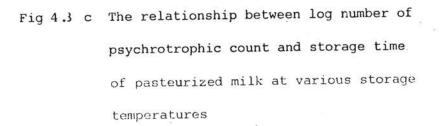
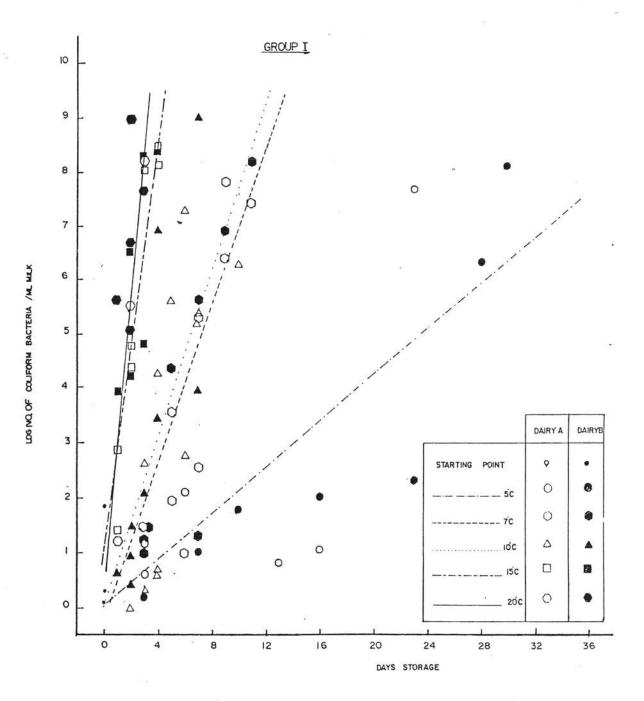
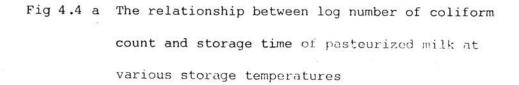


Table 4.7 Linear correlation coefficients betweeen log number of psychrotrophs and storage time of pasteurized milk at various temperatures.

Sample group	Storage temp	Linear correlation	Significant
	(°C)	coefficients	
group I	5	0.81	<0.01
	7	0.83	<0.01
	10	0.89	<0.01
	15	0.90	<Ø.01
	20	0.89	<0.01
group II	5	0.75	<0.01
	7	0.95	<0.01
	10	0.89	<0.01
	15	0.96	<0.01
	20	0.96	<0.01
group III	5	0.78	<0.01
a. 2	7	0.86	<0.01
	10	0.90	<0.01
5	15	0.89	<0.01
	20	0.91	<0.01





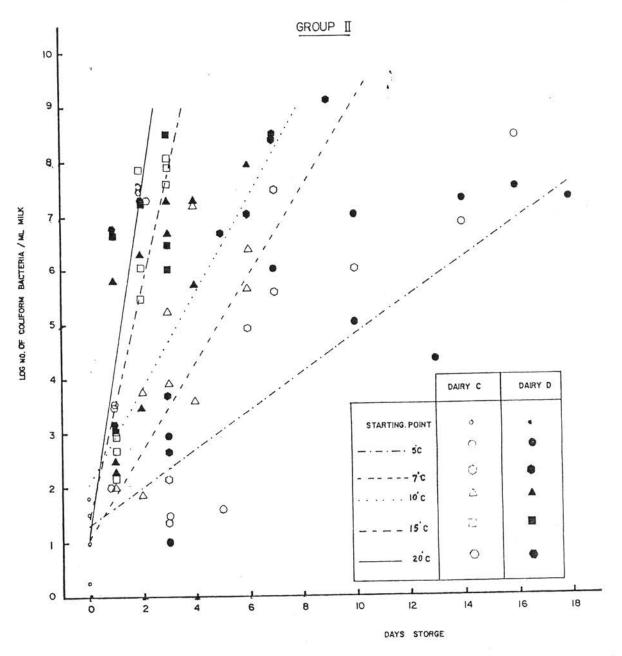
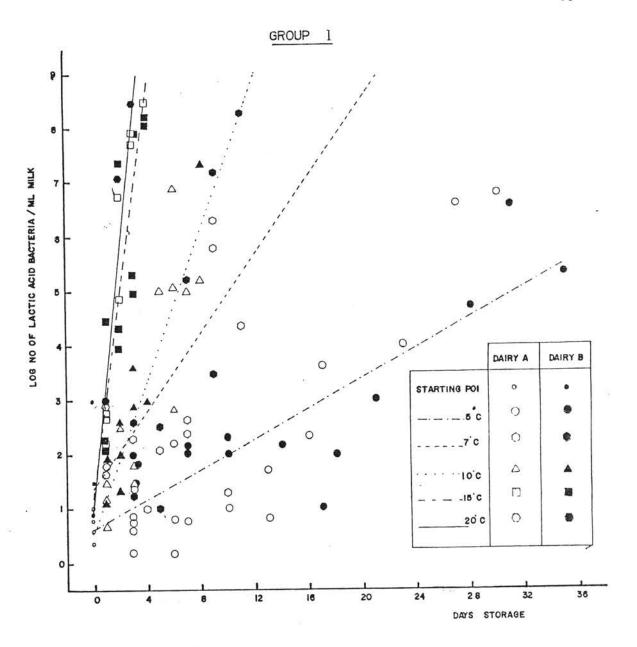


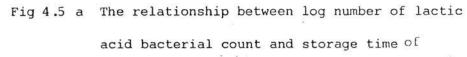
Fig 4.4 b The relationship between log number of coliform count and storage time of pasteurized mild; at various storage temperatures

# Table 4.8 Linear correlation coefficients between log number of coliform counts and storage time of pasteurized milk at various temperatures.

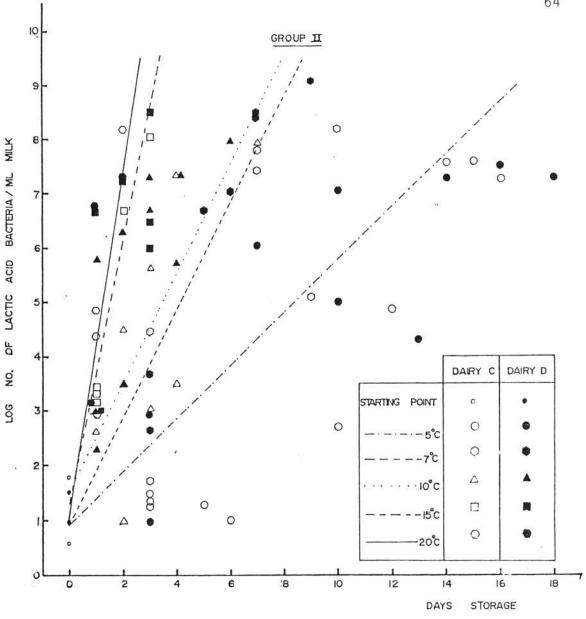
Sample group	Storage temp	Linear correlation	Significant
	(°c)	coefficients	
gboup I	5	0.85	<0.01
	7	0.85	<0.01
	10	0.76	<0.01
	15	0.93	<0.01
	20	0.87	<0.01
group II	5	0.87	<0.01
	7	0.88	<0.01
	10	0.75	<0.01
	15	0.91	<0.01
	20	0.92	<0.01

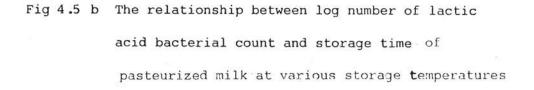
The coliform count of dairy E was very low. Most of the counts at all storage temperatures and times were less than 10 cfu/ml. So, 'the correlation between coliform count and storage time couldn't be found.

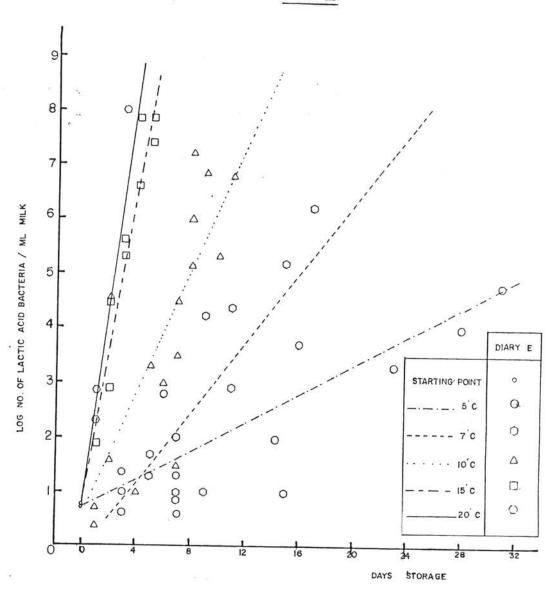


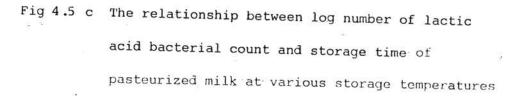


pasteurized milk at various storage temperatures









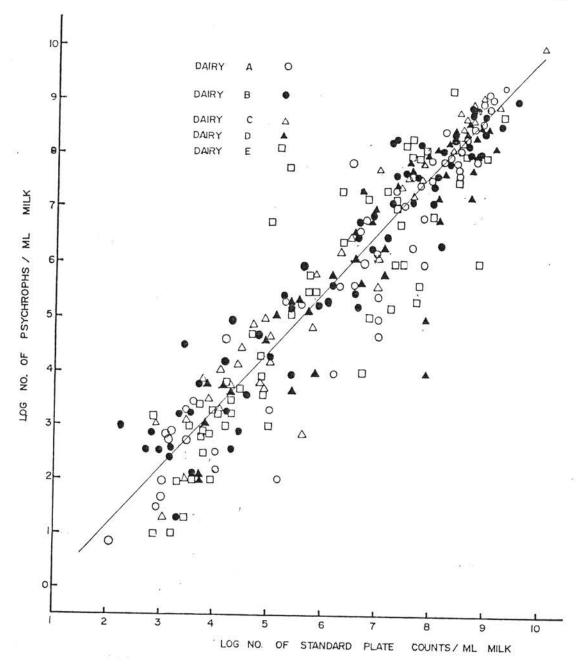
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Table 4.9 Linear correlation coefficients between log number of lactic acid bacteria and storage time of pastourized milk at . various temperatures.

	1	1	
Sample group	Storgge temp (°C)	Linear correlation	Significant
		coefficients	
group I	5	0.78	<0.01
	7	0.70	<0.01
8	10	0.91	<0.01
2	15	0.92	<0.01
	20	0.91	<0.01
group II	5	0.91	<0.01
	7	0.89	<0.01
	10	0.84	<0.01
	45	0.98	<0.01
	20	0.94	<0.01
group III	5	0.91	<0.01
	7	0.85	<0.01
	10	0.86	<0.01
	15	0.98	<0.01
	20	0.98	<0.01

At all storage temperatures and dairies, panelists didn't accept the milk when standard plate count and psychrotrophic count reached log 7-9 cfu/ml. Because the microorganisms studied in milk were mesophile and the storage temperatures were in the range between 5 and 20°C, the microorganisms grew faster at higher storage temperature and consequently causing shorten shelf-life at higher storage temperature. 4.2.2 Relationship between standard plate count, psychrotrophic count, coliform count and lactic acid bacteria.

At all storage temperatures times and dairies, the psychrotrophic count, coliform count and lactic acid bacteria increased linearity with standard plate count as shown in Fig. 4.6, 4.7 and 4.8 These linear correlation coefficients were highly significant as shown in Table 4.10 (see appendix F for calculation)



#### Fig 4.6

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Relationship between log number of standard plate count and log number of psychrotrophic count of pasteurized milk at all storage temperatures and times among various dairies

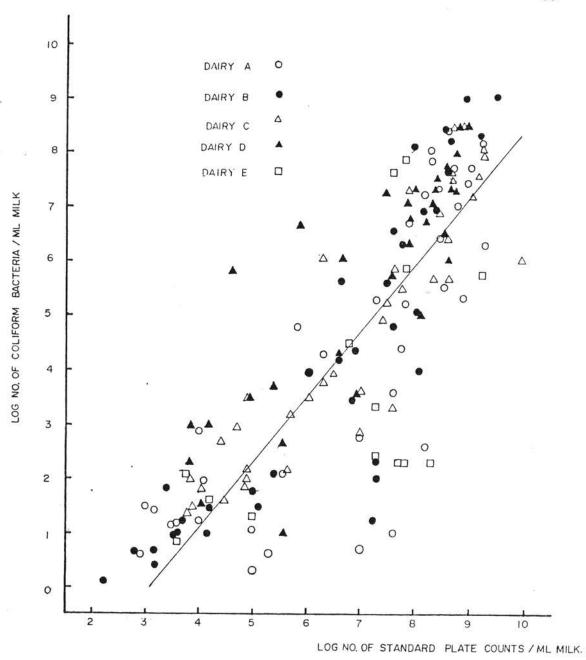
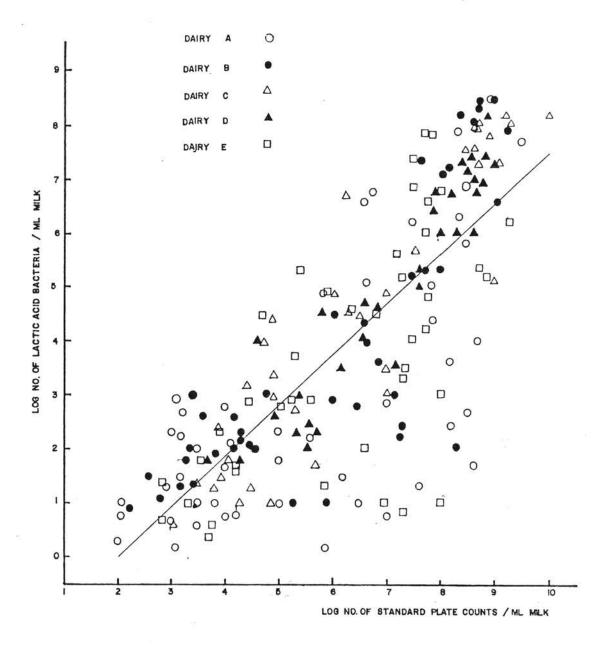


Fig 4.7 Relationship between log number of standard plate count and log number of coliform count of pasteurized milk at all storage temperatures and times among various dairies



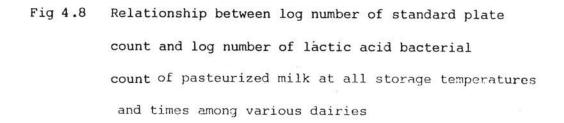


Table 4.10 Linear correlation coefficients between log number of standard plate counts and psychrotrophs, coliform counts and lactic acid bacteria of pasteurized milk at all storage temperatures and times from various dairies.

Kińds of bacteria	Linear correlation coefficients	Significant
Psychrotrophs	0.94	<0.01
Coliform counts	0.84	<0.01
Lactic acid .	0.80	<0.01
bacteria		

From the data obtained, the relationship between various kinds of count can be calculated and expressed as in the equations shown below:

log P/ml	= -1.00+1.08 log SPC/ml
log C/ml	= -2.97+1.12 log SPC/ml
log L/ml	= -1.85+0.93 log SPC/ml

Where SPC is standard plate counts, P is psychrotrophic counts, C is coliform counts and L is lactic acid bacterial counts.

Besides, multicorrelation among various kinds of count could also be calculated and expressed as shown in Table 4.11 (see appendix F for calculation)



Table 4.11 Equations and multiple correlation coefficients among standard plate count, psychrotrophic count, coliform count and lactic acid bacterial count of pasteurized milk at all storage temperatures and times from various dairies.

Equations	multiple correlation coefficients	Significant
log SPC/ml = 1.1689+0.7294 log P/ml +0.1775 log L/ml	0.967	<0.01
log SPC/ml = 1.9791+0.7027 log P/ml +0.1941 log C/ml	0.970	<0.01
log SPC/ml = 3.0591+0.393 log C/ml +0.3844 log L/ml	0.911	<0.01
log P/ml = 2.9297+0.7390 log C/ml +0.0116 log L/ml	0.928	<0.01

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From the equations expressed, it can be concluded that milk had a constant proportion of each type of counts at all storage temperatures between 5 and 20°C for all dairies at all storage times.

4.2.3 Composition of pasteurized milk at various storage

temperatures.

The compositional analysis of milks at various storage temperatures from each dairy are shown in Table 4.12

Table 4.12	Compositional	analysis	of pasteurized	milk	from	various	dairies	at	various	storage	temperatures
			,	MALK	11011	various	dairies	at	various	storage	temperatures

DAIRY	STORAGE TEMP						
	('c)	FAT	PROTEIN	LACTOSE	WATER	TOTAL SOLID	SOLID NOT FA
	5	3.25+0.09	3.29+0.08	4.63 ±0.09	a 87.66+0.26 <sup>a</sup>	11.89+0.11	8.65 +0.13
	7	3.30+0.08ª	3.29+0.07	4.58+0.11	a 87.60+0.14ª		8.59+0.11
A	10	3.40+0.26ª	3.28+0.09	4.58+0.14	87.59+0.16ª	_	- 8.60+0.14
	15	3.31+0.10 <sup>a</sup>	3.33 +0.10	4.53 +0.12	87.61+0.16ª	11.89+0.08	- 8.58+0.12
	20	3.36+0.10 <sup>a</sup>	3.33 +0.14	4.54+0.21ª	87.72 <u>+</u> 0.19 <sup>a</sup>		- 8.58+0.14
	5	3.68+1.61 <sup>a</sup>	3.29+0.07	4.74+0.09 <sup>a</sup>	87.41+1.61 <sup>a</sup>	12.42+1.53	8.74+0.11
	7	3.50 <u>+</u> 1.51 <sup>a</sup>	3.28+0.11	4.69+0.15ª	87.71+1.54ª	2875 28	8.69+0.21
В	10	3.68+1.21 <sup>a</sup>	3.31+0.10	4.71+0.10 <sup>a</sup>	87.50+1.16ª	12.40+1.15	- 8.73+0.08
	15	3.97+0.94 <sup>a</sup>	3.29+0.08	4.71+0.02ª	87.16+1.04 <sup>a</sup>	12.66+0.91	B.72+0.07
	20	4.41+0.28 <sup>a</sup>	3.23 +0.05	4.67±0.10ª	86.78+0.17ª	13.06+0.26	- 8.67+0.04
	5	4.47+0.06 <sup>a</sup>	3.33+0.08	4.57+0.10 <sup>a</sup>	86.37+0.23 <sup>a</sup>	13.10+0.13	8.62+0.11
	7	4.47 <u>+</u> 0.07 <sup>a</sup>	3.34+0.06	4.55+0.10 <sup>a</sup>	86.34+0.21 <sup>a</sup>	13.07+0.12	8.62+0.12
с	10	4.49+0.04 <sup>a</sup>	3.34+0.08	4.59+0.06 <sup>a</sup>		- 13.15+0.10	8.66+0.09
	15	4.53 ±0.04 a	3.33 +0.07	4.52+0.16 <sup>a</sup>	- 86.34+0.40 <sup>a</sup>	- 13.14+0.12	8.63 +0.12
	20	4.65 <u>+</u> 0.3 7 <sup>a</sup>	3.55+0.62	4.67±0.18ª	85.81+1.19 <sup>a</sup>	-	8.67 <u>+</u> 0.09
	5	3.87 <u>+</u> 0.26 <sup>a</sup>	3.34+0.09	4.70+0.07 <sup>a</sup>	87.02+0.24 <sup>a</sup>	12.65+0.20	8.76+0.07
	7	3.87+0.28 <sup>a</sup>	3.35 <u>+</u> 0.10	4.68+0.08 <sup>a</sup>	- 87.09+0.29 <sup>a</sup>	12.60+0.35	8.75+0.07
D	10	4.07+0.11ª	3.32+0.06	4.70+0.06 <sup>a</sup>	- 86.87+0.18 <sup>a</sup>	12.70+0.24	8.74+0.02
8	15	4.11+0.08 <sup>a</sup>	3.30+0.05	4.71+0.05ª	- 86.84+0.17 <sup>a</sup>	12.85+0.10	8.74+0.03
	20	4.08±0.10 <sup>a</sup>	3.30+0.05	4.72+0.06ª	86.87+0.21 <sup>a</sup>	- 12.77+0.06	8.73 +0.02
	5	3.98+0.27 <sup>a</sup>	3.27+0.27	4.56+0.19 <sup>a</sup>	87.29+0.62 <sup>a</sup>	-	
Е	7	4.06+0.46ª	3.28+0.26	- 4.59+0.19 <sup>a</sup>	87.22+0.63 a	12.58+0.64	8.51 <u>+</u> 0.34
	10	4.05±0.19 <sup>a</sup>	3.31 <u>+</u> 0.30	4.60+0.18ª	87.18+0.47 <sup>a</sup>	12.59+0.49	8.55 ±0.33 8.56+0.32
	. 15	4.14 <u>+</u> 0.27 <sup>a</sup>	3.30±0.33	4.58+0.19 <sup>a</sup>		12.59+0.55	8.52+0.35
	20	4.12+0.21ª	3.35+0.43	94235	-	12.60+0.54	8.52+0.35

The experiment was conducted by using RCBD

The difference of mean values of each item among various temperatures were determined by using DMRT at 95% significant level with different alphabets.

There was no difference of all items of composition among various storage temperatures which showed that the storage temperature had no effect on the changes in composition. The difference of each item among various dairies was found; It was due to the different process used, some dairies had milk standardized while some dairies had not. Mean composition of milk from various dairies was shown in Table 4.13.

Table 4.13 Mean composition of pasteurized milk from various dairies at all storage temperatures between 5 and 20°C

DAIRIES		Mean composition (%)										
	fat	protein	lactose	water	total solid	solid not fat						
Α					11.90 <u>+</u> 0.10 <sup>d</sup>							
В	3.64+1.42 <sup>c</sup>	3.30+0.09 <sup>a</sup>	4.71+0.11 <sup>a</sup>	87.53 +1.42	12.36 <u>+</u> 1.36 <sup>c</sup>	8.73 ±0.13 a						
С			2.225	· · · · · · · · · · · · · · · · · · ·	13.09 <u>+</u> 0.11 <sup>a</sup>							
D			1 A A		12.70 <u>+</u> 0.24 <sup>b</sup>							
Е	4.04+0.31 <sup>b</sup>	3.26+0.21 <sup>a</sup>	4.58+0.19 <sup>b</sup>	87.23 +0.56	12.56 <u>+</u> 0.54 <sup>b</sup>	8.54+0.32 <sup>c</sup>						

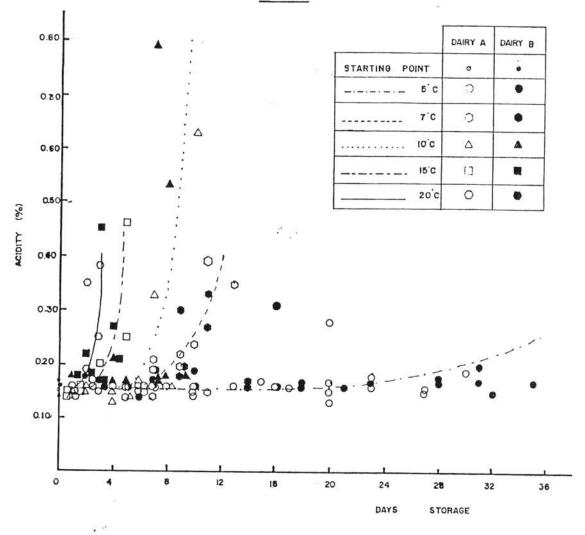
The experiment was conducted by using RCBD

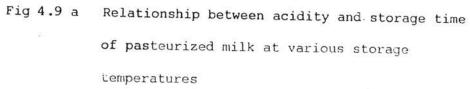
The difference of mean values of each item among various dairies were determined by using DMRT at 95% significant level with different alphabets.

4.2.4 Acidity of stored pasteurized milk at various storage temperatures.

The acidity of milk was rather constant at the first period of storage, then it increased with storage time. At the same storage time, the constant period of acidity was shorter when storage temperature was higher as shown in Fig. 4.9 a, b and c. This may be from the more growth of acid producing bacteria and/or the faster rate of lipolysis at higher temperature.







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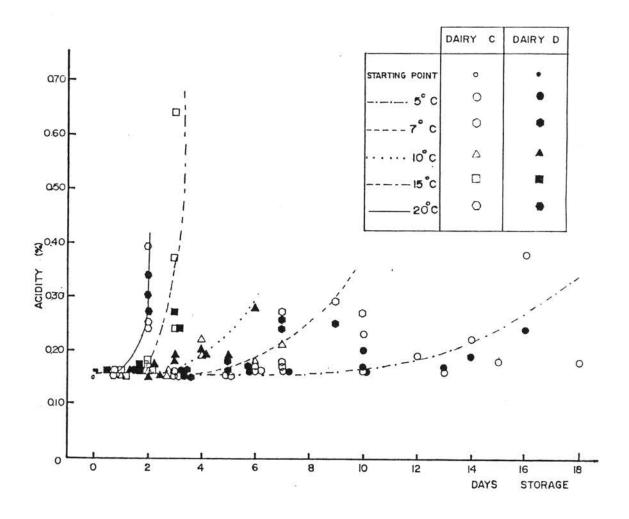


Fig 4.9 b Relationship between acidity and storage time of pasteurized milk at various storage temperatures

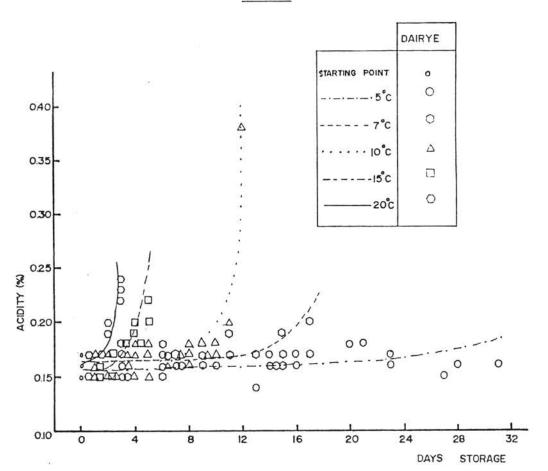


Fig 4.9 c Relationship between acidity and storage time of pasteurized milk at various storage temperatures

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4.2.5 Somatic cell counts of pasteurized milk at various storage temperatures.

Somatic cell count in this study showed a fluctuation. It was in the range between  $10^3$  and  $10^6$  cells/ml,but most of them was in the range of  $10^5$  cells/ml. This may be that the distribution of somatic cells in milk was not equal. The somatic cell counts of pasteurized milk from various dairies at various storage temperatures were shown in Table 4.14

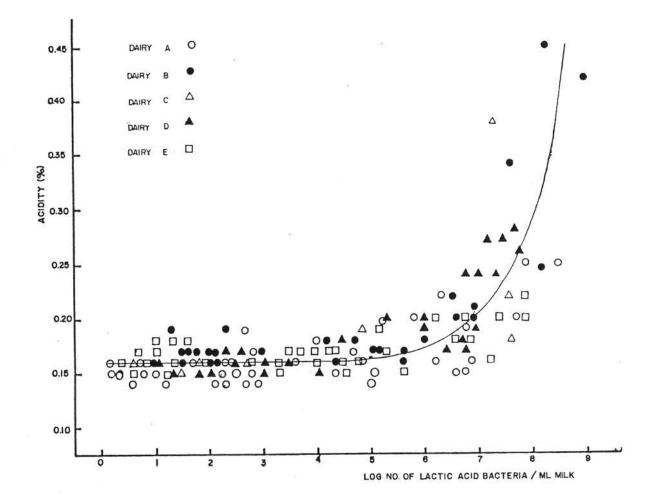
Table 4.14 Somatic cell counts of pasteurized milk from various dairies at various storage temperatures.

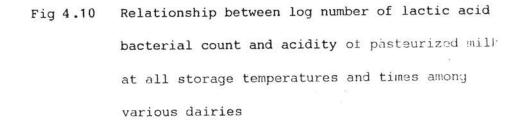
Storage temp (°C) Dairies	Somatic cell counts x 10 <sup>5</sup> /ml milk										
	5	7	10	15	20	average					
А	3.51+1.12	3.73 <u>+</u> 1.17	3.16+0.98	2.96+1.04	3.69+1.09	3.40+1.09					
В	3.59+2.25	3.03 + 2.02	3.55 <u>+</u> 1.56	4.49+1.73	4.15 <u>+</u> 1.98	3.49+2.04					
С	2.04 +4 .43	0.81 <u>+</u> 0.79	1.16+2.29	1.07+0.73	1.06+0.77	1.49+2.71					
D	2.67+3.88	1.46+1.48	2.22+5.27	1.18+0.43	1.02+0.50	1.96+3.59					
E	3.93+2.56	3.57+2.67	4.07+2.15	3.78+2.24	4.76+2.20	3.56+2.31					

In fact, the somatic cell should be destroyed by HTST pasteurization; but from the experiment, somatic cell was observed at various storage temperatures and times for all dairies. The presence of these cells was still doubtful and unfortunately there was no report involved. It is quite interesting to conduct study about the somatic cell in pasteurized milk.

## 4.2.6 The relationship between acidity and lactic acid bacteria

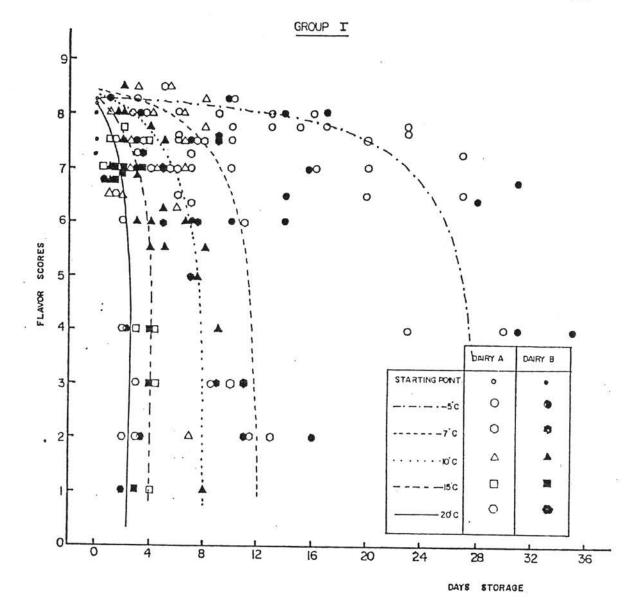
For all storage temperatures, times and dairies, the acidity was rather constant as lactic acid bacteria counts increased gradually. When lactic acid bacteria reach log 5 cfu/ml (colony forming units/ml milk), pronounced acidity in milk was observed as shown in Fig. 4.10. It should be that the first period of growth of lactic acid bacteria couldn't produce acid high enough to change the acidity of milk.





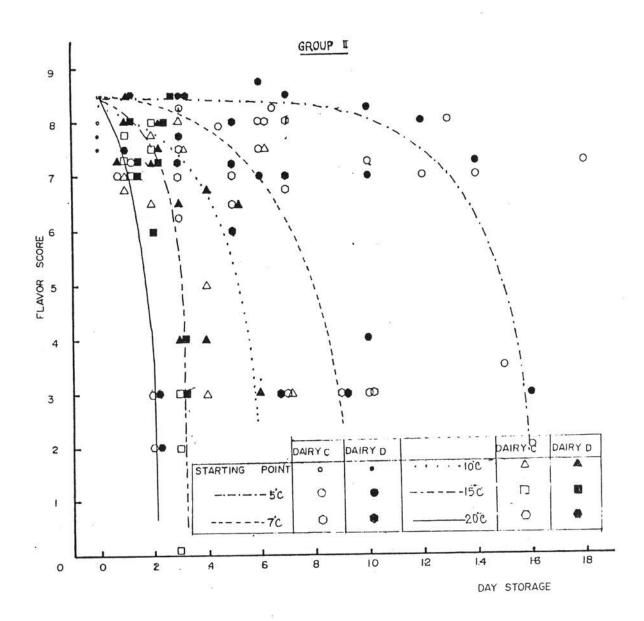
4.2.7 Organoleptic quality of stored pasteurized milk at various temperatures.

Flavor score for overall quality decreased as the storage time increased at all storage temperatures between 5 and 20°C. At the same storage time, the higher storage temperature decreased more flavor score as shown in Fig 4.11 a, b and c. When the score was equal or less than 4, (from a 9 Hedonic Scale) milk was graded as unacceptable.





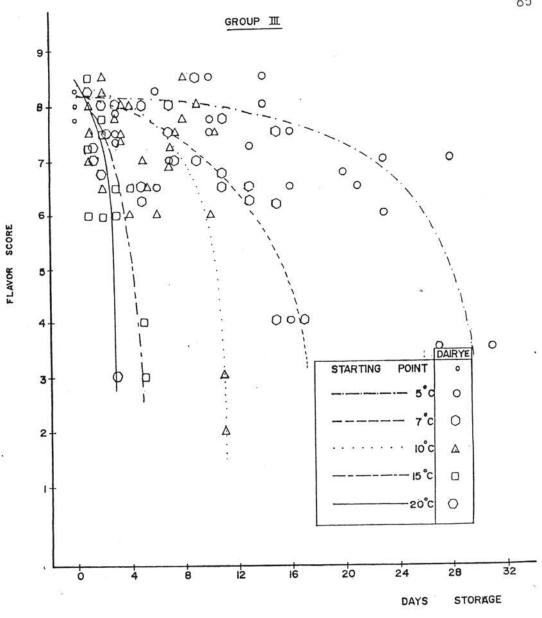
Relationship between flavor score and storage time of pasteurized milk at various storage temperatures

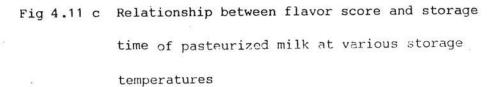




b Relationship between flavor score and storage time of pasteurized milk at various:storage

temperatures





4.2.8 The relationship between flavor score and standard plate count.

For all storage temperatures, times and dairies, panelists rejected the product (flavor score  $\leq 4$  from a 9 Hedonic Scale) which corresponded to the standard plate count about log 7-9 cfu/ml as the graph shown in Fig 4.12.

According to previous work (19, 26), it was reported that spoilage of pasteurized milk occurred when the counts reach 10<sup>7</sup> cfu/ml. So, this study showed something higher than previous reports. This might come from the fact that milk is not our daily food. The threshold of panelists to detect the unacceptability of milk would be at higher level of bacterial count than Europeans and Americans who are familiar with milk.

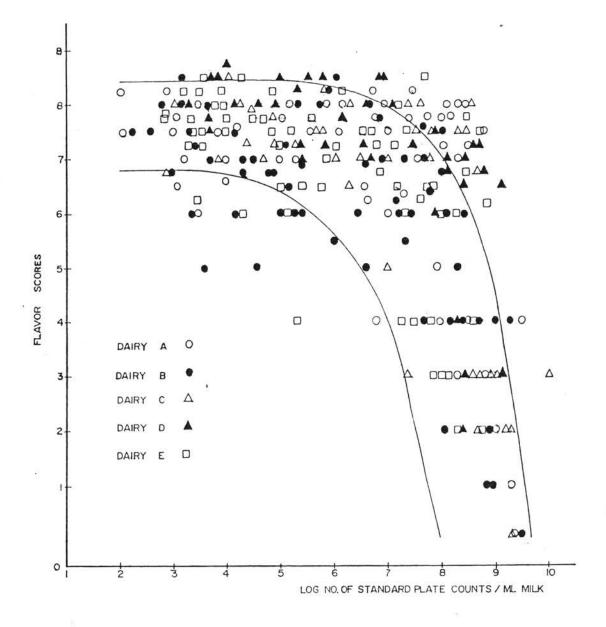


Fig 4.12

Relationship between flavor score and log number of standard plate count of pasteurized milk at all storage temperatures and times among various dairies

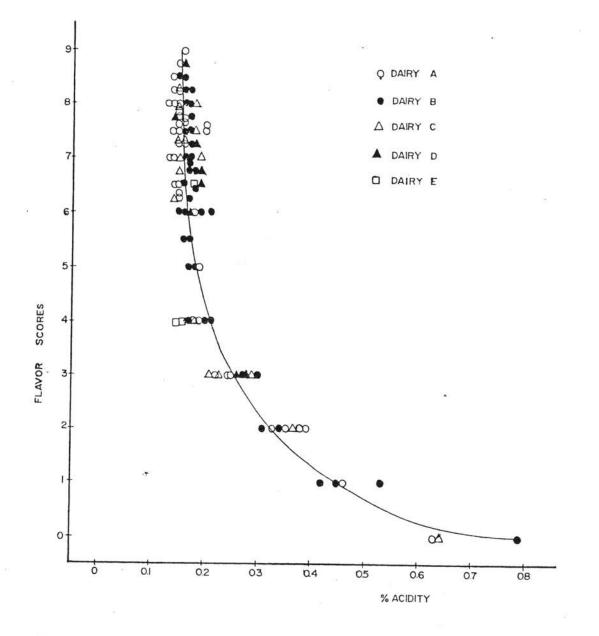


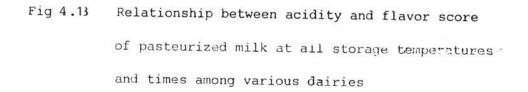
4.2.9 The relationship between flavor score and titratable acidity.

For all storage temperatures, times and dairies, panelists rejected the product (flavor score  $\leq$  4 from a 9 Hedonic Scale) when the acidity was equal or more than 0.2% as shown in Fig 4.13.

The panelists detected either grassy, unclean, cowy or rancidity flavor when they rejected the product which corresponded to the acidity in the range of equal or more than 0.2%, and detected the sourness of milk when the acidity was correspondingly more than 0.2%. These results are similar with the previous work (3.2, 3.6) which reported that a trained grader would detect rancidity in milk with an acid degree value (ADV = % acidity X 10) greater than 1.5 while most people found milk with an ADV over 2.0 unacceptable. At low levels of acidity, hydrolytic rancidity in milk was often perceived as old, unclean or cowy flavors. Increasing ADV gave bitter, butyric and finally definite rancid flavor.

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## 4.2.10 Enzymatic property of pasteurized milk at various storage temperatures and times.

From the experiment, there was mostly no protease found in milk from each dairy at all storage temperatures and times. However, it was found that two lots of samples, one from dairy B and the other from dairy E, kept at 20°C storage temperature contained this enzyme on the day the milk was not accepted (clot). At other storage temperatures, no protease was found even though the milk was clotted. Law (1979) suggested that counts of at least  $5 \times 10^6$  cfu/ml were necessary before proteolysis could be detect in milk (20). In this study, there was no enzyme protease detected even when psychrotrophic counts reached about  $10^{7-8}$  cfu/ml. It was probably due to relative low level of biochemical activity of these bacteria (19).

For lipase, the present of this enzyme was rather constant at all storage temperatures and times. It showed that the growth of microorganisms during storage produced not enough enzyme to give a clear zone in the experiment. It was reported that the lipolytic bacteria would not affect the flavor of milk until the counts reach  $10^6$  cfu/ml or more. It was also reported that the counts had to exceed  $10^6$  cfu/ml to cause rancid flavor (20, 36). In this study, it may said that presence of lipolytic bacteria in milk was not enough to produce lipase so that it was found unchange during storage, but the study of lipolytic bacteria was not carried out in this experiment.

The presence of both enzymes was shown as the clear zone in Table 4.15.

Storage	Dairies	Enzymes	Day Sampling																				
temp('C)		0	1	2	3	4	5		7 arz	8 one	9 (mm)	10	11	13	15	16	20	23	27	30	33	15	
	A	Protease	-	+	+	2		-	-	-	-	-	-	-	-	-	-	-	-	-		-	$\vdash$
1		Lipase	6.0			6.0			5.5	5.5			5.5		5.75				5.75				
	в	Protease	-			-			-				-		-		-	_	_	1	1	-	_
		Lipase	6.0	)																			1
5	с	Protease	-			- 2			-				-		-		_						
	D	Protease	12			-			-				-		-		_						
	Е	Protease	-			-			-				-		-		_	-	-	_			
	Λ	Protease	-	1	1	-	- 1	-	-	-	-		-			12				-	-	-	-
		Lipase	6.0			5.5	6.0		6.0	5.5	6.0		5.5					1					
	в	Protease				12		-		4		-		-									
		Lipase	6.0	1																			
7	с	Protease	-	1		-		-		-		-							-				
	D	Protease	-			-		-		-		-											
	Е	Protease	-			-		-		022		1		-	_	-							
	Λ	Protease	-	-	-	-			-	-	-	-	-			-	-+				-	-	-
		Lipase	6.0	5.5	6.0	5.75	6.0		5.5	6.0	6.0												
	в	Protease		-	-	-	-	-	-	-													
		Lipase	6.0	5.5	6.0														1				
10	с	Protease	-	-	-	-	12	2	-													1	6
	D	Protease	-	2	-	-	-																
	Е	Protease	-	-	-	-	-	-	-	-	-	-	-	-								- {}	
	Λ	Protease	-	-	-	-	-					-		-			+	-		-+	-	-	-
		Lipase	6.0	5.5	6.0	5.75	5.5	2	1														
	в	Protease	-	12	-	_																- 6	
		Lipase	6.0	5.5	5.5																		
15	с	Protease	-	-	-	-																- 22	
	D	Protease	-	сња Г	-	-		- 8														2	
	E	Protease	-	123	-	- 2 1	-	-	-														
	λ	Protease	-	-	-	-			-	1000		-		-+			+	+		-	-		-
		Lipase	6.0	6.0	5.5	5.5				527													
	В	Protease	-	-		5.5	- 1		1 1														
		Lipase	6.0	6.0	5.5																		
20	C I	Protease	-	-	-		- 1																
			-	-	-																		
		Protease	-	_		5.0																	
					222				- 3														
																							L

## Table 4.15 Enzyme protease and lipase of pasteurized milk at various storage temperatures and times from various dairies.

- = Negative result



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## 4.3 <u>Keeping quality of pasteurized milk from various dairies at</u> various storage temperatures.

4.3.1 Loading of microorganisms from various dairies at various storage temperatures.

From the experiment, there was no difference of count, both standard plate count and psychrotrophic count, for all dairies at various storage temperatures. But the difference of psychrotrophic count occurred among various dairies while dairy E which had a good sanitation had low count of psychrotroph on the unaccepted day. The results of standard plate count and psychrotrophic count obtained when the milk was not accepted were log 8.618 and 8.293 cfu/ml as shown in Table 4.16 and 4.17 respectively.

Table 4.16 Log number of standard plate count in pasteurized milk kept at various storage temperatures from various dairies.

Dairies Temp(°C)	A	В	с	D	Е	Mean value
5	8.159	8.494	9.3 03	9.233	8.786	8.795
7	8.495	8.813	9.113	8.739	8.599	8.752
10	8.645	9.3 01	8.704	9.098	8.440	8.838
15	8.434	8.836	8.3 27	8.655	8.664	8.583
20	8.015	8.3 83	8.159	7.935	8.124	1.457
Mean value	8.350 <sup>a</sup>	8.765 <sup>a</sup>	8.721 <sup>a</sup>	8.732 <sup>a</sup>	8.522 <sup>a</sup>	8.618

The experiment was conducted by using 5x5 factorial complete block design. (see appendix E).

The mean values among various dairies were determined by using DMRT at 95% significant level with different alphabets.

Dairies 'Temp('C)	A	В	С	D	E	Maan valué		
5	8.159	8.022	8.742	8.366	7.306	8.119		
7	7 8.581		9.286	8.519	7.392	8.506		
10	9.322	8.929	8.584	8.004	7.469	8.462		
15	9.233	8.336	8.847	8.437	6.997	8.370		
20	8.786	7.923	8.311	8.3 01	6.725	8.009		
Mean value	8.816 <sup>a</sup>	8.3 93 <sup>a</sup>	8.754 <sup>a</sup>	8.3 25 <sup>a</sup>	7.178 <sup>b</sup>	8.293		

Table 4.17 Log number of psychrotrophic count in pasteurized milk

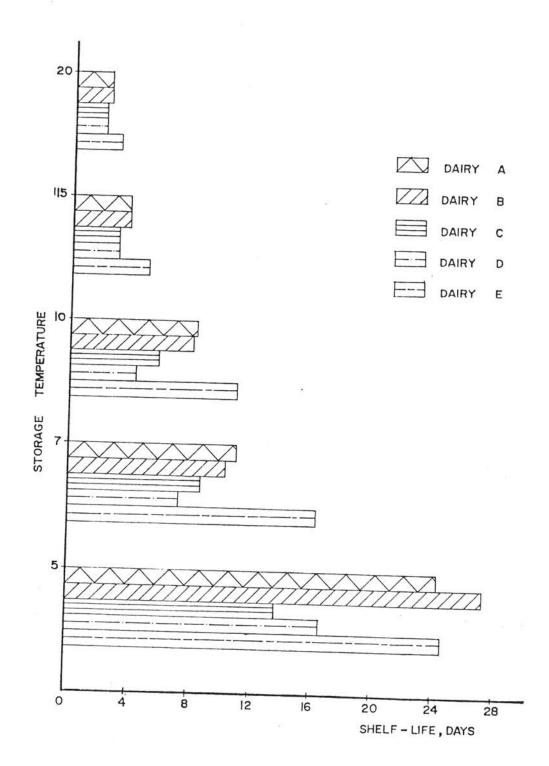
kept at various storage temperatures from various dairies.

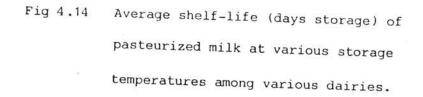
The results obtained are higher than those reported on previous studies (19, 26, 30) which was cited in 4.2.8.

The psychrotrophic count of dairy E was lowest. This might be that this type of bacteria was from post-pasteurization contamination only. If it was so, it could be considered that the sanitation of this dairy was the best.

Both kinds of count on the day the milk was not accepted were higher than previous work. It was reported that the spoilage of milk occurred when the count reach 10<sup>7</sup>(log 7)cfu/ml (19, 26). In addition, the study of Chander et al (1984) which reported that the detection of organoleptic changes in most dairy products occurred when psychrotrophic counts reached log 7.5cfu/ml (30). Both of the counts are higher than the reported value, this may result from the reason that milk is not our daily food, thus, the sensitivity and threshold for detecting the spoilage of milk is lower than that of reported. 4.3.2 <u>Shelf-life of pasteurized milk at various storage</u> temperatures from various dairies.

From the experiment, it was found that shelf-life of pasteurized milk at various storage temperatures from various dairies was different. The degree of difference was depending upon the storage temperature. At temperature between 15 and 20°C, there was no difference of shelf-life among dairies. At temperature <10°C, shelf-life of milk was considered difference depending upon packaging material and sanitation condition. The results are shown in Table 4.18 and Fig 4.14.





STORAGE TEMP(°C)	Shelf-life of pasteurized milk (days)									
DAIRIES	' <del>'</del>	7	10	15	20					
X	A <sub>24.3</sub> a	<sup>B</sup> 11.0 <sup>b</sup>	AB8.3 bc	<sup>A</sup> 3.7 <sup>c</sup>	A2.3d					
В	A27.3 a	<sup>B</sup> 10.3 <sup>b</sup>	AB8.0 <sup>bc</sup>	<sup>A</sup> 3.7 <sup>c</sup>	<sup>A</sup> 2.3 <sup>d</sup>					
С	<sup>B</sup> 13.7 <sup>a</sup>	<sup>B</sup> 8.7 <sup>b</sup>	AB5.8 <sup>bc</sup>	A3.0 <sup>bc</sup>	A2.0 <sup>c</sup>					
D	<sup>B</sup> 15.3 <sup>a</sup>	<sup>B</sup> 7.7 <sup>b</sup>	B4.3 bc	A3.0 <sup>bc</sup>	A2.0 <sup>c</sup>					
Е	A24.7 <sup>a</sup>	A16.3 b	A11.0 <sup>C</sup>	A5.0 <sup>d</sup>	A3.0 <sup>d</sup>					

Table 4.18 The average shelf-life of pasteurized milk at various storage temperatures and various dairies.

The experiment was conducted by using 5x5 factorial complete block design.

The difference of mean values among storage temperatures was determined using DMRT at 95% significant level with small alphabet.

The difference of mean values among dairies was determined by using DMRT at 95% significant level with big alphabet.

From the results obtained, it can be seen that the reasons to divide these five dairies into three groups as stated in 4.2.1 are quite reasonable. However, there was a little difference of shelf-life of group II (dairy C and D) at 10°C storage temperature but the same packaging material was used.

In the experiment, there was a fluctuation of shelf-life of milk at 5°C-storage temperature between replication. This was due to the fluctuation of electricity during storage. The fluctuation of storage temperature would obviously shorten shelf-life of milk, but the data were also collected. So, the shelf-life of pasteurized milk shown above was the minimum.