

FILTRATION METHODS FOR
MAKING TURKISH SÜZME
(THICK) YOGURT

FILTRATION METHODS FOR MAKING TURKISH SÜZME (THICK) YOGURT

By

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CONTENTS

Page

Acknowledgements	x
Abstract	xi
1.0 INTRODUCTION.....	1
2.0 LITERATURE REVIEW.....	5
2.1 Historical background of yogurt.....	5
2.2 Varieties of yogurt and their classification.....	6
2.2.1 Varieties of yogurt.....	6
2.2.2 Classification of yogurt	7
2.3 Yogurt consumption statistics	8
2.4 Properties of milks.....	10
2.4.1 The chemical composition of milks	10
2.4.2 Buffer capacity of ultrafiltered milk	11
2.5 The basic production technology of yogurt.....	12
2.5.1 Procure raw milk supply.....	12
2.5.2 Pre-treatment.....	13
2.5.2.1 Clarification.....	13
2.5.2.2 Standardisation methods of total solids content of milk.....	13
2.5.2.3 Heat treatment.....	15
2.5.3 Inoculation and incubation	17
2.5.3.1 Fermentation during incubation.....	17
2.5.3.2 The principle of acid coagulation	18
2.5.4 Cooling.....	20
2.5.5.Packaging	21
2.5.6 Storage.....	21
2.6 Some properties of natural and thick yogurts.....	21
2.6.1 Standard natural yogurt and its chemical and physical properties	22
2.6.2 Thick yogurt, production methods of thick yogurt and its chemical and physical properties.....	30
2.6.2.1 Concentrated yogurt ("süzme or torba " yogurt).....	32
2.6.2.2 The production methods of thick yogurt and its chemical and physical properties.....	33
2.6.2.2.1 Concentration by traditional method	33
2.6.2.2.2 Concentration by mechanical separators	34

	Page
2.6.2.2.3 Concentration by membrane filtration methods.....	35
2.6.2.2.4 Production method of concentrated yogurt from reconstituted milk	40
2.6.3 Microbiological properties of yogurt.....	41
2.6.3.1 Taxonomy of yogurt starter cultures.....	42
2.6.3.2 Biological aspect of the association between <i>S.thermophilus</i> and <i>L. bulgaricus</i>	43
2.6.4 Some important aspects of metabolic activity of yogurt starter cultures	46
2.6.4.1 Carbohydrate metabolism.....	46
2.6.4.2 Protein metabolism.....	47
2.6.4.3 Formation of flavour compound of yogurt	50
2.6.4.3.1 Flavour compounds of yogurt.....	50
2.6.4.3.2 Formation of flavour compound from glucose	51
2.6.4.3.3 Formation of flavour compound from protein	53
2.6.4.3.4 Formation of flavour compound from DNA component.....	53
2.6.5 Nutritional and therapeutic properties of yogurt	54
2.6.5.1 Protein.....	54
2.6.5.2 Lactose	55
2.6.5.3 Fat	57
2.6.5.4 Minerals.....	58
2.6.5.5 Therapeutic properties of yogurt.....	59
 3.0 MATERIAL AND METHODS	 61
3.1 Material	61
3.1.1 Raw materials	61
3.1.1.1 Raw cow's and ewe's milk	61
3.1.1.2 Starter cultures.....	61
3.1.1.3 Filter medium	61
3.2 Methods.....	62
3.2.1 Starter culture propagation	62
3.2.2 Concentration methods of raw milk	63
3.2.3 Heat treatment	63

	Page
3.2.4 Production methods of high total solids Turkish style thick yogurt.....	64
3.2.4.1 Production method of stirred style Turkish thick yogurt by using modified traditional method.....	64
3.2.4.2 Production method of set style Turkish thick yogurt by using fortified ultrafiltered milk retentate	65
3.2.5 Analysis methods.....	70
3.2.5.1 Analysis methods of milk	70
3.2.5.1.1 Determination of pH.....	70
3.2.5.1.2 Determination of titratable acidity	70
3.2.5.1.3 Total solids.....	70
3.2.5.1.4 Protein.....	71
3.2.5.1.5 Lactose	73
3.2.5.1.6 Fat	74
3.2.5.1.7 Ash.....	74
3.2.5.1.8 Buffer capacity.....	74
3.2.5.1.9 Mineral analysis	75
3.2.5.1.10 Microbiological analysis	76
3.2.5.1.10.1 Total count of initial bacteria in raw milk.....	76
3.2.5.1.10.2 Coliform bacteria count	77
3.2.5.1.10.3 Yeast and Mould count	77
3.2.5.2 Analysis methods of yogurt	77
3.2.5.2.1 Determination of pH.....	77
3.2.5.2.2 Determination of titratable acidity	77
3.2.5.2.3 Determination of total solids.....	78
3.2.5.2.4 Determination of protein.....	78
3.2.5.2.5 Determination of lactose.....	78
3.2.5.2.6 Determination of fat.....	78
3.2.5.2.7 Determination of ash.....	78
3.2.5.2.8 Determination of firmness	78
3.2.5.2.9 Water holding capacity	80
3.2.5.2.10 Volatile aroma compounds	80
3.2.5.2.11 Free amino acids	83
3.2.5.2.12 Mineral analysis	86
3.2.5.2.13 Microbiological analysis	86
3.2.5.2.13.1 Total count	86

	Page
3.2.5.2.13.2.Starter cultures count in yogurt.....	86
3.2.5.2.13.3 Coliform bacteria count	87
3.2.5.2.13.4 Yeast and Mould	87
3.2.5.3. Chemical analysis of UF permeate and whey from modified traditional filtration.....	87
3.2.5.4. Statistical analysis.....	87
4.0 RESULT AND DISCUSSION.....	88
4.1 Raw materials.....	88
4.1.1 Chemical composition of milk	88
4.1.2 Microbiological quality of raw milk.....	89
4.1.3 Buffer capacity of milk.....	90
4.2 Changes of titratable acidity and pH of whole milk and fortified UF retentate during incubation period.....	94
4.3 Some chemical, physical and microbiological properties of thick yogurt produced from cow's and ewe's milk using different production process.....	100
4.3.1 Production method of set and stirred style thick yogurt.....	100
4.3.1.1 Transfer rate of total solid during the concentration.	101
4.3.2 Chemical composition of yogurt and permeate.....	102
4.3.2.1 Chemical composition of fortified set style thick yogurt made from different milk.	102
4.3.2.2 Chemical composition of thick yogurt made using the modified traditional method.....	103
4.3.2.3 Chemical composition of permeate from UF and modified traditional filtration.....	105
4.3.2.4 Mineral contents of yogurt.....	108
4.3.3. Titratable acidity and pH of thick yogurts.....	108
4.3.4 Water holding capacity of yogurt.	112
4.3.5 Firmness of yogurt.....	116
4.3.6.Volatile aroma compounds of yogurt.	119
4.3.6.1 Analysis method of some volatile aroma compounds.....	119
4.3.6.2 Volatile aroma compounds in set and stirred style thick yogurt.	120
4.3.7 Free amino acids analysis in yogurt.....	124
4.3.7.1 The principle of free amino acids analysis.	124

	Page
4.3.7.2 Free amino acids content of set and stirred style thick yogurt.....	126
4.3.8 Microbiological properties of thick yogurt.....	130
4.3.8.1 Some microbiological changes in set and stirred style thick yogurt.	130
4.3.8.2 Non- lactic acid, coliform bacteria, yeast and moulds in yogurt.	131
4.4 Some chemical, physical and microbiological changes in thick yogurt produced from cow's and ewe's milk during storage period	132
4.4.1 Changes of titratable acidity and pH of set and stirred style thick yogurt during storage period	133
4.4.2 Changes of water holding capacity of set and stirred style thick yogurt during storage period.....	139
4.4.3 Changes of firmness of set and stirred style thick yogurt during storage period.....	143
4.4.4 Changes of volatile aroma compounds of set and stirred style thick yogurt during storage period	146
4.4.5 Changes of free amino acids of set and stirred style thick yogurt during storage period.....	154
4.4.6 Changes of microbiological properties of set and stirred style thick yogurt during storage period	156
4.5 Thick yogurt production by using modified traditional filtration method.....	160
4.5.1. Theory of batch cake filtration method	160
4.5.2. Whey drainage results of batch cake filtration	161
5.0 CONCLUSION	166
6.0 APPENDICES	169
Appendix I.....	173
Appendix II	179
Appendix III	181
Appendix IV.....	189
Appendix V.....	193
Appendix VI.....	199
Appendix VII	213
Appendix VIII	215
Appendix IX.....	219
7.0 REFERENCES.....	220

List of Figures

Fig 2.1-Changes in micelle structure in the course of acidification.....	20
Fig 2.2-The flow diagram outline for the production of set and stirred type natural yogurts in Turkey	23
Fig 2.3-Flow diagram outlining the production of concentrated Turkish "suzme/torba" yogurt.....	31
Fig 2.4-The schema of symbiosis of yogurt bacteria	45
Fig 2.5-Schema of lactose utilisation by homofermentative bacteria	48
Fig 2.6-Diagrammatic representation of the known reactions involving acetaldehyde	52
Fig 3.1-The two type of weaves	62
Fig 3.2-The flow diagram of starter cultures propagation.....	63
Fig 3.3-The flow diagram of concentration of yogurt by using modified traditional method (vacuum batch filtration)	66
Fig 3.4-Modified traditional filtration apparatus	67
Fig 3.5-A generalised schema for production method of set style Turkish thick yogurt produced from UF retentate	68
Fig 3.6-Ultrafiltration unit used in experimentation	69
Fig 3.7-Penetrometer apparatus	79
Fig 3.8-Steam distillation apparatus.....	81
Fig 4.1-Buffer capacity of fortified UF retentate for cow's milk.	92
Fig 4.2-Buffer capacity of fortified UF retentate for ewe's milk.....	93
Fig 4.3-Changes of pH from different total solids UF cow's milk retentate during incubation period.	95
Fig 4.4-Changes of pH from different total solids UF ewe's milk retentate during incubation period	96
Fig 4.5-Titratable acidity measurement during incubation time from cow's milk.	98
Fig 4.6-Titratable acidity measurement during incubation time from ewe's milk.	99
Fig 4.7-Mineral content of set and stirred style yogurt from cow's milk.	109
Fig 4.8-Mineral content of set and stirred style yogurt from ewe's milk.	110
Fig 4.9-Water holding capacity of cow's and ewe's milk yogurt produced using different production method.	114
Fig 4.10- Free amino acid content of set and stirred style thick cow's milk yogurt produced by using different production methods.	128

	Page
Fig 4.11- Free amino acid content of set and stirred style thick ewe's milk yogurt produced by using different production methods.	129
Fig 4.12- Changes of titratable acidity in yogurt produced from cow's milk during storage period.....	134
Fig 4.13- Changes of titratable acidity in yogurt produced from ewe's milk during storage period.....	135
Fig 4.14-Changes of pH in yogurt produced from cow's milk during storage period.	136
Fig 4.15-Changes of pH in yogurt produced from ewe's milk during storage period.	137
Fig 4.16-Changes of water holding capacity of set and stirred style yogurt produced from cow's milk during storage period.....	141
Fig 4.17-Changes of water holding capacity of set and stirred style yogurt produced from ewe's milk during storage period.	142
Fig 4.18-Changes of firmness of set and stirred style yogurt produced from cow's milk during storage period.	144
Fig 4.19-Changes of firmness of set and stirred style yogurt produced from ewe's milk during storage period.....	145
Fig 4.20-The changes of acetaldehyde level of yogurt produced from cow's milk during storage period.	146
Fig 4.21-The changes of acetaldehyde level of yogurt produced from ewe's milk during storage period.	147
Fig 4.22-The changes of acetone level of yogurt produced from cow's milk during storage period.....	150
Fig 4.23-The changes of acetone level of yogurt produced from ewe's milk during storage period.....	151
Fig 4.24-The changes of diacetyl level of yogurt produced from cow's milk during storage period.....	152
Fig 4.25-The changes of diacetyl level of yogurt produced from ewe's milk during storage period.....	153
Fig 4.26-Plots of filtrate volume versus time for cow's and ewe's milk yogurt using the cotton fiber cloth.....	162
Fig 4.27-Plots of filtrate volume versus time for cow's and ewe's milk yogurt using the synthetic filter cloth.	163

List of Tables

Page

Table 2.1- The consumption of yogurt in certain countries	9
Table 2.1-Average chemical composition (%) of milk of different species of mammals	11
Table 2.3-Heat treatment of milk and yogurt	15
Table 2.4-The amount of free amino acids in milk and yogurt (mg/100g)	29
Table 2.5-Required and optional bacteria of yogurt starter cultures	42
Table 2.6-The main taxonomic properties of <i>S. thermophilus</i> and <i>L. bulgaricus</i>	44
Table 2.7-Production of carbonyl compounds (ppm) by yogurt starter cultures	51
Table 4.1- Coding system key for products.	88
Table 4.2- Chemical composition of milk used in this study (% w/v).	89
Table 4.3- The microbiological quality of raw cow's and ewe's milk (cfu/ml).	90
Table 4.4- Chemical composition of whole milk and thick yogurts produced using different production methods.	104
Table 4.5- Chemical composition of commercial Süzme (thick) yogurt produced in Turkey.	105
Table 4.6- Chemical composition of permeate obtained from UF and modified traditional filtration methods.	107-
Table 4.7- Titratable acidity and pH in different types of yogurts.	111
Table 4.8- Penetration results of different total solid thick yogurt produced from fortified UF milk retentate and using modified traditional filtration method.	117
Table 4.9- The volatile aroma compounds of yogurt samples.	121
Table 4.10- The changes of total free amino acids amounts during storage period (mg / 100 ml).	155
Table 4.11-Changes of <i>L. bulgaricus</i> in set and stirred style yogurt produced from cow's and ewe's milk using different production method during storage period.	158
Table 4.12-Changes of <i>S. thermophilus</i> in set and stirred style yogurt produced from cow's and ewe's milk using different production method during storage period.	159
Table 4.13- Constants from fit of data to $[V=(A+B\Delta p)(\Theta^c)]$	161

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ABSTRACT

Süzme or thick yogurt, a concentrated yogurt, is produced from cow's and ewe's milk by a traditional method used in Turkey. This traditional method of thick yogurt production consist of putting natural yogurt into bags, made from cotton cloth produced for this purpose, and allowing the yogurt to concentrate by natural drainage as a batchwise operation.

In this study, two types of thick yogurt were produced from cow's and ewe's milk using a commercial yogurt starter cultures coded CH-1.

a) A thick yogurt, called a set type yogurt in this thesis, was produced from ultrafiltered milk retentate, the ultrafiltered milk retentate being fortified with fresh permeate to five different (24-32 %) total solid levels .

b) A thick yogurt was produced using a modified traditional method which is referred to as stirred type yogurt in this thesis. The modified traditional method was, essentially, a type of batch filtration. Two different filter media, the traditional cotton filter cloth and a synthetic fibre filter cloth were used as a filter membrane. Natural drainage and two levels of vacuum suction were applied to the filter membrane to improve the drainage rate. Total solids level of 26.5 % were obtained using this method.

After incubation, samples were stored in refrigerator at 4°C for 42 days. Samples were analyzed after 24 hrs of storage and every seven days thereafter. The advantages of ultrafiltration method are: higher yield (up to 10%), a shorter production time, a smoother body, texture and better palatable test. The vacuum filtration was faster than natural drainage, but slower than ultrafiltration. The filtration time was 60 min for ewe's milk yogurt and 90 min for cow's milk yogurt with vacuum filtration and 25 min for ewe's milk and 40 min for cow's milk with UF. The filtration time was affected by initial total solid level.

The total solids content of products varied from 12 to 32 % depending on the process. Protein content was 3.42 to 12.1 %. Fat content was 4 to 14.7 %. Ash content 0.71 to 1.5 % (w/w). Mineral content increased in set-type thick yogurt and decreased in stirred type thick yogurt. The total solid content of permeate was 4.94 to 6.03 % for UF and 5.76 to 7.2 % for cotton filter media and 5.86 to 10 % for synthetic filter media for vacuum filtration. Protein and mineral losses was higher in permeate for vacuum filtration. The titratable acidity was between 1.00 to 1.77% lactic acid and pH was between 4.35 to 4.72. The end of storage period titratable acidity increased about 20 % and pH decreased about 0.30 unit. The production of thick yogurt from high total solid milk retarded production time significantly. Post-acidity development was also affected during storage period. Water holding capacity was higher in high total solid yogurt. It was not affected significantly during storage period. The firmness of set and stirred type yogurt showed positive correlation with the total solid content and yogurt types. The set-type thick yogurt had higher firmness. Firmness was not affected significantly during storage period. Acetaldehyde was predominant as an aroma compound in all types of yogurts. Its amount was between 10 to 23 ppm and increased when total solid level increased in set type thick yogurt. Diacetyl level was about 0.5 to 1.0 ppm. Acetone level was 1.5 to 3.5 ppm. The amount of volatile aroma compounds decreased during storage period. Lowest aroma compounds were obtained in stirred type thick yogurt. The free amino acids content were increased during storage period. Their amount also correlated with total solid level and production methods. The viable bacteria count decreased by considerable amount for all yogurts during storage. In most cases different types of milk did not affect yogurt quality. Good quality thick yogurt can be produced with both methods. However, the optimum acidity, higher yield, more hygienic product and firmest thick yogurt can be produce from fortified UF retentate.

Key Words: Thick yogurt, Süzme yogurt, Vacuum filtration, Ultrafiltration, Storage, Quality properties, Aroma compound, Free amino acids, Water holding capacity, Firmness, Acidity, Viable bacteria count.

CERTIFICATE OF ORIGINALITY

This is to certify that I am responsible for the work submitted in this thesis, that the original work is my own except as specified in acknowledgements or in footnotes, and that neither the thesis nor the original work contained therein has been submitted to this or any other institution for a higher degree.

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Date :.....

1.0 INTRODUCTION

According to IDF (1969) the definition of fermented milks is as follows: "Products prepared from whole, partially or fully skimmed, concentrated milk or milk substituted from partially or fully skimmed dried milk, homogenised or not, pasteurised or sterilised and fermented by means of specific microorganisms." It is obvious from the microorganisms present in the starter culture that they have to be taken into consideration in order to ensure a better quality end product and consequently these aspects can affect the overall characteristics of each type of fermented milk.

The exact names given to the fermented milk products are not well established but it is likely to be several hundred. In general fermented milks are manufactured from cow's milk and the names of other cultured dairy products should imply the type of milk used. The term "cultured" is frequently substituent for "fermented" when considering fermented milks such as yogurt because these products were the first to be produced commercially on a large scale by pure bacterial cultures (Kosikowski, 1977).

Although the names of the most well-known traditional fermented milks come from Turkish language (yogurt and ayran) other products have names based on their characteristics such as the method of preparation (eg. whipped milk).

At present new fermented milks have names based on scientific or microbiological origin (acidophilus milk); however, some other names are given mainly by industrial firms. For instance, the prefix "bio" is given to products signifying the biological and natural state eg., "biodrink" "biokys". "Jog" or "yogi" is used to replace a popular product like yogurt when incorporated in a drink (Kurmann, 1984).

Thick yogurt in Turkey is produced in either animal skin or cloth bags but generally a cloth bag is used. The scale of production of these yogurts used to be small, *i.e.* household scale and was really a means of utilising the surplus of summer milk. However, even with centralisation of milk production and the processing of milk in dairies, the concentrated yogurt is still produced in a traditional fashion, *i.e.*, by the use of the cloth bag. The use of this system by most dairies in Turkey is not only because the concentrated product retain its traditional qualities but also the cloth bag is easy to wash, sterilise and cheap.

The süzme yogurt is a thick type of yogurt. "Çökelek" is also a form of thick yogurt. These can be retained for winter consumption. The süzme yogurt is unique in itself with the exact character of the product directly affected by the level of moisture retained, the degree of acidity, the microorganisms involved, as well as the type of milk used. This implies that with homebased production, different batches of concentrated yogurt totally lack uniformity.

Some of the concentrated products have now gained access to large urban populations. However, in spite of some standardisation of the process, consecutive batches of concentrated yogurt still shows wide variations in character. In some countries an attempt has made to introduce Laws or Standards eg. Turkey (Turkish Food Acts and Turkish yogurt standard-1330), where it is stipulated that "süzme yogurt" should have a specific chemical composition based on fat, total solids and salt. Nevertheless, adherence to these acts poses problems.

The most important factor in the production of thick yogurt is the volume of whey removed. Hence, the removal of excess whey yields a product that is crumbly in texture, dry and rather sharp in flavour. On the other hand, the removal of too little whey yields a product that is too thin and very mild in flavour. Therefore, the ideal characteristics of concentrated yogurt are a smooth texture, homogenous consistency and a proper aromatic flavour. The attainment of these characteristics is governed, to some extent, by the level of moisture in the final product, a feature closely coupled, of course, with the economics of processing.

The time of whey drainage varies from 12-24 hours, however, the amount of filling is critical because if too much pressure is exerted, losses of milk solids into the whey are high. Furthermore, if the whey drainage is very fast only the yogurt adjacent to the cloth bag becomes concentrated at the quicker rate and the inner content of the bag remains in a virtually liquid state. Although the resulting products are not homogeneous, the problem can be overcome to some extent by thorough mixing prior to packaging. However, excessive mixing can cause pronounced shearing of the coagulum and the product quickly shows signs of syneresis upon even limited storage.

In some respects the process for producing set or stirred style yogurts, at the present, have few problems but some areas do appear to be in need of examination. These are as following:

- i) the definition of the product in Turkish Food Act (Anon, 1980) states that "süzme/torba" yogurt shall designate the semi solid dairy product derived from natural yogurt by straining away part of its water and some water soluble compounds" and from this definition, it can be concluded that there is no standard production process to meet the growing demand.
- ii) although the chemical composition of the products laid down by the above legislation, e.g. the amount of fat minimum 5%, the amount of the total solid minimum 30 %, titratable acidity 2.25 lactic acid % and the amount of salt 1 % maximum the achievement of this composition in set or stirred style thick yogurt is dependent on the raw material yogurt. However, the above legislation also stipulates the chemical composition of natural standard yogurt as ; fat 3 % minimum, the solid non fat content 9 % minimum and titratable acidity 0.80 - 1.60 lactic acid %. If such a yogurt is used directly for the production of thick yogurt (süzme / torba yogurt), then the concentrated product either does not comply with the above specification or it is quite unacceptable organoleptically or economically; e.g. the cost of labour to meet the above legislation. Hence, it would seem essential that the statutory composition of concentrated thick yogurt should be re-examined in conjunction with proposed legislation concerning natural yogurt.

It is these aspects of the production of natural yogurt and stirred style thick yogurt that form the subject of the present study. There is, of course, a degree of overlap between the areas but for reasons of practical convenience the investigation was divided into the following main sections, namely the production of natural and set or stirred style thick yogurt by using different production method, using cow's and ewe's milk to produce the yogurt, examine the shelf life of the products and sensory and chemical properties of the products.

Therefore, it was decided that the aims of this research would be to:-

- i) Compare and contrast the qualities of stirred type thick or "süzme" yogurt obtained from the modified traditional filtration method with set type of thick yogurt produced from fortified ultrafiltered milk retentate.
- ii) Study the effects of cold storage on the quality of thick yogurt.
- iii) Study and characterise different concentration levels (24, 26, 28, 30, 32% total solids) for the manufacture of thick yogurt by using cow's and ewe's milk and fortified UF milk with whole cow's and ewe's milk for set yogurt and for stirred yogurt of 26.5% total solids.
- iv) Determination and identification some of flavour characteristics of such different type of thick yogurt, such as yogurt produced by using vacuum batch filtration and from fortified UF retentate.
- v) Provide more exact definition of this yogurt taking into consideration the chemical, microbiological, rheological and some nutritional characteristics.
- vi) The feasibility of manufacturing of set and stirred type of yogurt by using different production methods.

2.0 LITERATURE REVIEW

2.1 Historical background of yogurt

In order to write a historical background about yogurt it would be necessary to use the respective knowledge from various branches of science. These include ethnology, linguistics, geography, ancient history, the history of economics, the history of diets, archaeology, legends, literary, religious and legal texts (Kurmann, 1984).

The exact origin of yogurt is difficult to establish. It may date back thousands of years to the beginning of the domestication of animals. A great deal of today's known reputed fermented milks come from Central Asian nomadic cattle breeders. Yogurt and related dairy products formed an important ingredient in their diet (Yöney, 1979).

The oldest references to any fermented milks are the following (Kurmann, 1984): Yogurt in Turkey in the year 8 th century AD (Rasic and Kurmann, 1978; Yöney, 1979), Ayran in Turkey and central Asian countries in the year 1253-55 AD and Leben occurs in an arabian medical text book in year 633 AD.

According to legend, yogurt was first made by the ancient Turkic people in Central Asia where they lived as nomads. Another legend tells that an angel brought down the pot that contained the first yogurt; while yet another claims that the ancient Turks, who were Buddhist, use to offer up yogurt to the angels and stars who protected them (Yaygin, 1969; Rasic and Kurmann, 1978).

Helferich and Westhoff (1980) mention another classical story about the beginning of fermented milks, in which a desert nomad wandering into the desert with fresh milk held in a goat skin bag placed beside the body of his camel where the heat from his camel's body set up optimum temperature for the growth of lactic acids

bacteria. After sometime, the nomad found the milk had coagulated into semi-solid mixture.

In 1712 E. Kaempfer on his travels to Asia described the palaces of the Emperor of Persia, which had a special room denominated as "yogurt house". Yogurt was used in the preparation of human foods and in the feeding of birds and cattle. Other data mention leben of ancient Arabs or yogurt of Turks, as fermented milks, which may become too sour during storing and subsequent drying (Oberman, 1985).

Originally, yogurt was made from ewe's milk and buffalo's milk and partly from goat's and cow's milk in containers of wood or argil. Propagation was carried out by using a small quantity of previously coagulated milk to seed the next batch of boiled milk. Yogurt was used primarily in the human diet for direct consumption, either as a natural product or fortified with various vegetables and spices but also for cooking and baking purpose.

After the second world war and particularly since 1950, the technology of yogurt and understanding of the factors affecting its organoleptic properties advanced rapidly. The use of pure cultures enabled the manufacture of consistent fermented products. The application of modern equipment has resulted in continuous processes. The introduction of fruit into manufacture, followed by a wide range of flavoured yogurts, promoted further growth in the consumption of the product. From the above information it can be concluded that yogurt is one of the oldest dairy products.

2.2 Varieties of yogurt and their classification

2.2.1 Varieties of yogurt

Over the past 70 years and especially in the last 30 years, yogurt has evolved into

many variations. There are many "type / kinds" of yogurt offered for sale. These are plain yogurt, fruit flavoured yogurt (made with fruit pulp or puree with added sugar), flavoured yogurt (it is usually made with artificial flavours and sugar and other sweetening agents), sundae-style yogurt, Swiss-style yogurt, natural yogurt and drinkable yogurt (ayran) with its lower total solids content. In addition to these now well known variations, there are some others such as frozen yogurt (soft, hard), dried yogurt (sun dried, freeze dried, spray dried), pasteurized / UHT yogurt, concentrated yogurt (with cloth bag, with UF before coagulation or after coagulation), low energy yogurt, lactose hydrolysed yogurt also yogurt with added vitamins or cereal products (Bottazzi, 1983; Vedamuthu, 1991; Akin and Rice, 1994). An aseptic yogurt with an extended shelf-life (Barua and Hampton, 1986) and a dietetic or therapeutic yogurt are also produced.

2.2.2 Classification of yogurt

Yogurts have a long history and since their development, have implications for nutritional, health and physiology as well as from the point of view of taste. Tamime and Deeth (1980) and others have proposed classification of yogurts. There is no single universally accepted classification at present. However, the following criteria can be used for these classification of yogurts.

In terms of chemical composition, yogurt is mainly classified into three types on the basis of fat content, *i.e.*, full-fat, half-fat, low-fat yogurt. In the Turkish Food Act and Turkish yogurt standards 1330, the presence of "live" starter organisms in the end product is stipulated. The main type of yogurt and yogurt-related products can be classified based on moisture content of yogurt type as follows:

<u>Total solids level</u>	<u>Products type</u>
Liquid or semi-liquid	Set and stirred standard plain yogurt, buttermilk, "Ayran" / yogurt drink
Semisolid	"Süzme or Torba" yogurt (high concentrated yogurt) "Çökelek" or Lor" (Cooked yogurt)
Powdered or solid	Dried yogurt ("Kurut") Frozen yogurt

2.3 Yogurt consumption statistics

Yogurt and related dairy products are the most widespread and popular of fermented milks. Wide range of data and statistic is available for them. The popularity of yogurt has increased all over the world. The yogurt boom is ascribed largely to its image of health and low fat, from a sour or tart food to a convenient, wholesome and "natural" food.

The total and per capita consumption of yogurt and other fermented dairy products in various countries are shown in Table (2.1). It can be seen that some of the countries listed in the table have a low yogurt consumption but some others have high yogurt consumption. As it can be seen the highest yogurt consumption is in the Netherlands and Switzerland (21, 17 kg / capita / year respectively). It may well reflect similar ethnic and cultural heritage of countries. The high consumption of fermented milks in Scandinavian countries might be the result of the climate that has always limited the diversity of plant food produced there.

Table (2.1) The consumption of yogurt in certain countries

Countries	Annual per capita consumption (kg)		Annual total consumption (1000 tonne)	
	1989**	1991***	1989**	1991***
Austria	7.3	8.7	57.3	67.1
Australia	3.6	4.1	60.7	72.0
Belgium	6.9	6.2	68.4	62.0
Canada	3.2	3.4	83.9	90.7
Switzerland	17.2	17.3	117.0	120.3
Chile	4.1	4.1	53.0	54.3
Czech-Slovak Rep.	3.3	3.3	51.6	50.8
Germany	10.6	12.2	657.0	972.0
Denmark	7.9	8.6	40.6	44.3
Spain	7.7	8.3*	297.7	315.4*
Finland	11.4	11.8	56.8	59.1
France	15.9	16.8*	892.7	949.6*
UK	4.1	4.4	234.0	254.0
Greece	-	6.0	-	60.0
Hungary	1.1	1.0	11.6	10.9
Ireland	3.4	3.3*	11.9	11.7
Israel	9.5	-	43.3	-
India	4.4	4.3	3570.0	3950.0
Iceland	8.7	9.5	2.2	2.5
Italy	2.4	4.8*	138.9	260*
Japan	4.0	3.9	487.0	485.0
Luxembourg	5.1	7.0	2.0	2.8
Netherlands	21.1	21.9	314.2	329.6
Norway	4.6	6.5	17.2	27.8
Russia	-	8.4*	-	909.5*
Sweden	7.3	7.0	62.0	59.5
USA	-	2.0	354.0	493.0
South Africa	1.6	1.7	49.3	54.4

* All fermented milks

** IDF (1991) Consumption statistics for milk and milk products

*** IDF (1993) Consumption statistics for milk and milk products

The UK yogurt market has shown a strong steady growth during the past ten years. Between 1980-1991 the market grew 201 % by volume, to reach 256000 tonnes. The consumption per capita reached 4.4 kg in 1991 (IDF,1993), which is 4 % greater than 1990 but still lower than in neighbouring European countries. Between 1987-91 average increases of total consumption was 6.6 %.

The potential for future growth of yogurt consumption in the UK may be indicated by comparison to the per capita consumption of yogurt and other fermented dairy products in certain European countries. The data given in Table (2.1) shows that the yogurt consumed in some European countries is 2-5 times greater than the yogurt consumed in the UK.

2.4 Properties of milk

2.4.1 The chemical composition of milks

The chemical composition of milks is influenced by many factors such as type of mammals, stage of lactation, climate, diet of animals and season of the year (Juarez, and Ramos, 1984). Table (2.2) shows the chemical composition of milk from different species of mammals. It is evident that in gross chemical composition goat's milk is closely related to that of cow's milk (see table 2.2). Ewe's milk contains high levels of fat, protein and ash when compared with goat's and cow's milk. Thus yogurt and other fermented dairy products which are manufactured from ewe's milk have a richer and creamer texture with an excellent mouth-feel and a firm coagulum.

Table (2.2) Average chemical composition (%) of milk of different species of mammals (Anifantakis, 1989; Juárez and Ramos, 1984).

Components (%)	Cow	Ewe
Total solids	12.80	15.80 - 19.30
Protein	3.58	4.30 - 5.20
Fat	4.14	4.50 - 7.90
Lactose	4.69	4.20 - 4.80
Ash	0.71	0.79 - 0.90
Minerals (mg / L)		
Na	580 - 480	410 - 1320
K	950 - 1570	1010 - 1520
Mg	120	140 - 190
Ca	1230 - 1200	1620 - 2590
P	1410 - 1570	820 - 1830
S	300 - 250	-

2.4.2 Buffer capacity of ultrafiltered milk.

Ultrafiltration increases the buffer capacity of milk (Brule et al, 1974; Covacecich and Kosikowski, 1979) because of increases in protein and insoluble salts (Mistry et al, 1987). As a result, the amount of lactic acid required by starter bacteria to produce a unit change in pH is considerably increased (Mistry and Kosikowski, 1983 and 1985) and the coagulation time for yogurt milk is lengthened. The higher the degree of UF the more pronounced this increased will be (Mistry and Kosikowski, 1985).

2.5 The basic production technology of yogurt

It is difficult to provide a general production method for yogurt to suit every variation in the final products marketed by industry. Also, it is impossible to prescribe universal incubation conditions because dairy factory schedules, turnover times for batch incubation and the necessary equipment required differ from one operation to another. Whatever the procedure is used, it is essential that the final product conforms to the requirement of the national and international regulations, *i.e.* safe for public consumption, in addition to having satisfactory sensory properties and consistency. Yogurt production consists of several basic steps and inputs as follows:

- i) Procure raw milk supply
- ii) Pre-treatment such as standardisation of the total solids and fat contents and heat treatment.
- iv) Inoculation and incubation
- v) Cooling
- vi) Storage
- vii) Packaging

2.5.1 Procure raw milk supply

Milk for production of yogurt should comply with the following requirements: low bacteria counts, absence of pathogenic organisms and absence or negligible levels of various inhibitory substances. These may find their way into milk, such as residues of antibiotics from the treatment of mastitis and residues of sanitation chemicals.

2.5.2 Pre-Treatments

2.5.2.1 Clarification

Milk should be clarified prior to heat treatment to remove undissolved solids, debris, leucocytes and epithelial cells from the milk. Clarification can be done either in a specially designed centrifuge or in a standard milk separator. In most cases, a standard milk separator is used both for clarification and standardization.

2.5.2.2 Standardization methods of total solids content of yogurt milk

In some cases, milk has to be standardized by composition to meet legal requirements, to improve the economy of production or to modify certain characteristics of the final product. Standardization, usually means adjustment of fat content and, sometimes, non-fat solids content.

The solid-non fat (SNF) content of milk (lactose, protein and ash) ranges between 8.2-8.4 % (Tamime and Robinson, 1985). Usually for yogurt, the SNF content is increased to improve the texture and consistency of product. The consistency of yogurt can also be improved when the basic mix is followed by homogenisation and heat treatment of the milk prior to fermentation (Kessler, 1981). The level of total solids in milk for yogurt production can vary from 9 % in skimmed milk yogurt to over 20 % in other types of yogurt. The total solids content of commercial yogurts usually falls between 14-15 % (Tamime and Deeth, 1980). The relationship between the level of solids in the milk and the consistency of yogurt was studied by Tamime (1977 a), It was concluded that the physical characteristics of yogurt (e.g. consistency / viscosity of the coagulum) were greatly improved with increase of the total solids from 12 to 14 -16 %.

Standardization of the fat content and milk non-fat solids content can be achieved in several different ways (Abrahamsen and Holmen, 1980; Kessler, 1981; Bottazzi, 1983; Tamime and Robinson, 1985; Glover, 1985) such as;

- i) Traditionally, in Turkey, milk is boiled to reduce the volume to 2/3 of the original value, although such a method is not used in commercial practice.
- ii) The addition of milk powder (full cream, skimmed or buttermilk) is mostly employed to fortify liquid milk for the manufacture of thick, smooth yogurt. The level varies from 1-6 % and the recommended level is about 3-4 % milk powder. The solubility of skimmed milk powder is important in its use; generally spray dried or freeze-dried skimmed milk powder has better solubility (98-99.9%) (Rasic and Kurmann, 1978).
- iii) Concentration of the liquid milk before homogenisation and heat treatment by evaporation, ultrafiltration (UF) and reverse osmosis (RO)
- iv) Addition of whey powder at rate of 1-2%.
- iv) Addition of caseinate which enhances the hydrophilic nature of the protein in the mix and acts as a stabiliser.
- v) The addition of skimmed milk to whole milk or the addition of whole milk or cream into skimmed milk. It can also be done by separation of some cream from whole milk.

Membrane filtration, especially ultrafiltration, is increasingly being used to concentrate milk for producing yogurt. Glover (1985) has summarized the main advantages of using UF retentate in the production of yogurt:

- i) increased consistency,
- ii) reduced syneresis,
- iii) increased product yield due to a partial inclusion of whey proteins.
- iv) quite often better taste due to absence of chalkiness frequently associated with the use of milk powder.

Total solids content in milk can also be increased by evaporation which usually removes 20-40 % water. In Turkey many manufacturers use vacuum evaporated milk to produce yogurt.

2.5.2.3 Heat treatment:

The temperature and time employed during heat treatment of milk are shown in Table (2.3). Heating at 85°C for 30-45 min is considered a convenient industrial practice especially for yogurt production.

Heat-induced changes in milk constituents are described by Rasic and Krumann (1978); Puhan (1979); Tamime and Deeth (1980); Kessler (1981); Robinson and Tamime (1986).

Table (2.3) Heat treatment of milk and yogurt

(Adapted from: Rasic and Krumann, 1978; Tamime and Robinson, 1985).

Temperature (°C)	Holding Time	Comment
65	30 min	Pasteurisation.
72	15 s	
85	30-45 min	High Temperature
90-95	5-10 min	
115	3 s	
135	16 s	(UHT) Ultra High Temperature
145	3 s	(UHT)

The main beneficial effects of heat treatment of yogurt milk are:

- i) to improve the consistency and firmness of yogurt and reduce syneresis. Heating the milk is sufficient to denature the whey protein and induce interaction between β -lactoglobulin and possibly α -lactalbumin with k-casein. Such interactions are considered to be the most important physio-chemical change which improve the consistency of the protein.
- ii) to make the milk a more nutritious growth medium for the starter organisms due to release of some amino acids and inactivation of inhibitory substances.
- iii) to inactivate lipase enzymes and thus prevent formation of hydrolytic rancidity.
- iv) to kill all pathogenic and the majority of other microorganisms
- v) redistribution of minerals, particularly calcium, between the soluble and colloidal forms thus leading to the reduction of coagulation time.
- vi) the amount of oxygen is reduced so providing the microaerophilic condition required by the starter organisms.
- vii) production of volatile compounds which contribute to, for example, the formation of lactones, methyl ketones and other volatile ketones from fat.

To achieve the above objectives, it is common practice to vat-pasteurize milk at 80-95°C for 5-40 min. (Rasic and Kurmann, 1978; Bottazzi, 1983). Heating of milk to the required temperature in a plate heat exchanger is followed by pumping into a holding unit where the temperature is maintained for the required period of time. Milk is heated in some production units in a continuous process in a closed system at 90-95°C with a holding time of 2-5 minutes or more. Heat exchangers are used also to cool milk down to the inoculation temperature.

The denatured whey proteins interact with casein micelle aggregates. This increases their hydrophilic properties and facilitates a stable coagulum. Furthermore, the heat treatment shortens the coagulation time which is partly due to decreases in pH or

the result of the formation of the whey proteins casein complex. Also the heat treatment reduces the nutritive value of some milk constituents (Kessler, 1981). For example, the amount of lysin is reduced to a limited degree (*i.e.* 0.3 %) due to the interaction between the amino group (-NH₂) of the lysine with the lactose. Some vitamin are also destroyed with the most affected being ascorbic acid (vit.C), and B type vitamins (Tamime and Deeth, 1980).

2.5.3 Inoculation and Incubation

Inoculation with the starter cultures is generally at the rate of 0.5-4 % and the incubation temperature of 40-45°C usually does not differ much from the optimum growth temperature of the starter organisms. Incubation temperature must remain constant during the incubation period. While incubating the starter culture bacteria produce lactic acid and flavour compounds. Incubation processing units can be vastly different, *e.g.*, cabinet, warm room, tunnel or vat units. They can be heated by hot air, steam, or water jacket. Cabinets are often used for the production of yogurt.

2.5.3.1 Fermentation during incubation

In general, there are many different type of fermentation reactions that can occur in milk. The nature of their end products is different and can adversely affect the food quality. Among these the lactic acid fermentation is the most important and popular in the dairy industry (Kosikowski, 1977).

Fermentation of milk is achieved as the result of the biological activity of the microorganisms and their enzymes and the starter culture may consist of bacteria, yeast and moulds or their combinations.

Therefore, the role of lactic acid starter culture during the manufacture of fermented milk products as reported by Dellaglio (1988) could be summarised as follows:

- i) The production of lactic acid from lactose in milk and the formation of antibacterial or inhibitory substances which prevent the growth of pathogenic microorganisms in milk, so extending the shelf-life of product.
- ii) The production of aromatic flavour compounds, *i.e.* diacetyl, acetone or acetaldehyde, which are essential for flavour and taste of fermented milks.
- iii) The drop in pH is a result of acid formation which helps to transform the rheological properties of milk and the formation of coagulum.
- iv) The production of proteolytic enzymes which are important in some dairy products.
- v) Some lactic acid bacteria produce polysaccharides which are useful in preventing serum separation and can improve the texture of the product.

2.5.3.2 The principle of acid coagulation

Casein comprises about 80 % of the total protein of milk (*i.e.* cow's milk) and consists of a number of fractions namely α_s -, β -, k -, and δ -casein (Swaigood, 1992). The casein in milk is present in small particles called micelles which are built up of sub-units bridged together by calcium and phosphate complexes. The casein-phosphate particles exhibit great stability in fresh milk of normal composition and maintain an equilibrium with the liquid phase of milk. This is due to electrical charges carried by the micelle particles and the polymer interaction between α_s -casein and k -casein in a ratio of 3:1 (Webb and Johnson, 1965). However, the stability of casein micelles may be affected by changes in the milk composition related to ions especially hydrogen ion and salt concentration (Rasic and Kurmann, 1978).

According to Scott (1986) casein may be coagulated by three different methods namely salt-, enzyme-, acid coagulation. The mechanism of coagulation is due to the addition of acids (acetic, citric, lactic *etc*) and the starter cultures to milk to form a coagulum. This method of coagulation has been used widely by cheesemakers for many years to make cheese or to produce casein. With respect to yogurt or other fermented dairy products the lactic acid which is produced by the starter culture also produces a similar precipitation of casein. The first initial precipitation of casein appears at pH 5.3 and complete precipitation at pH 4.6 which is the isoelectric point.

The effect of lowering pH is to reduce the ionization of the acidic functional groups of the casein (aspartic, glutamic and phosphoserine residues). This reduction of ionization causes a lowering of the surface potential (Darling and Dickson, 1979) and leads to a decrease in the sequestering capacity of α_s -casein and β -casein and an increase in the water solubility of the calcium salts. This results in a progressive displacement into the aqueous phase (Davies and White, 1960; Brule et al, 1974) and hence in demineralisation of the micelle. Total demineralization occurs at a pH of less than 5. Given the predominant role of calcium and phosphate in the micella structure, the loss of these inorganic constituents is accompanied by the disaggregation of the micelles into their micellar subunits.

At pH value of 5.7-5.8, about 50 % of the colloidal calcium has passed into solution (Brule et al, 1974). A change in the rheological characteristics of the milk is then observed. This probably corresponds to a disorganization of the micelles, though the subunits remain particularly associated with one another by calcium and phosphate linkages (see Fig 2.1). The structure of the micelle subunits was caused by the presence of a hydrophilic and a hydrophobic segment in β - and κ -casein. By lowering the pH the ionization of polar sites located on the exterior of the subunits is reduced. This probably results in changes in organisation of the casein polymer. Furthermore, the production of lactic acid as well as the resulting increase in solubility of micellar calcium and phosphate lead to a significant increase in ionic

strength. This may favour depolymerization of α_s -casein.

In the course of acidification there occurs a profound disorganization of micelle, accompanied by a change in the quaternary structure of casein. As the isoelectric pH is approached, the change is neutralized. The hydration is also significantly reduced so that the casein are rendered insoluble.

The coagulum obtained is the result of the formation of an insoluble protein network which encloses the whole of the aqueous phase in its meshes. The junction point of the network are the submicelles, totally demineralized and to some extent modified in their structure. The intermolecular linkages involved in the formation of this network are electrostatic and hydrophobic in nature.

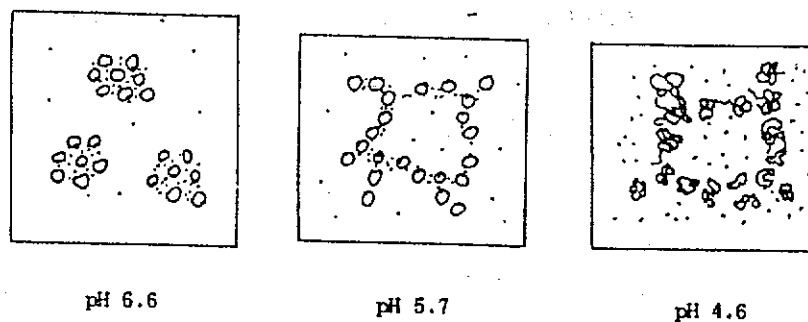


Fig (2.1) Changes in micelle structure in the course of acidification

2.5.4 Cooling:

Incubation is ended by cooling which depends on whether the product is stirred or set. This can be achieved by several techniques. Cooling of the product, usually to 4-5°C, allows the producer to control the metabolic activity of starter organisms. Cooling period and temperature are often critical to quality of the final product.

They are usually determined empirically by each manufacturer to provide the most suitable yogurt for the market.

2.5.5 Packaging:

Semi-solid or set type yogurt is packaged in the container before incubation and cooling. The stirred or fluid yogurt drink is incubated in bulk and is packaged after stirring or mixing. Different types of container or packaging materials can be used for packaging (e.g. glass, waxed paper, plastics, laminated paper, or re-usable metal container). Packaging material selection becomes more important for long-time storage of the product.

2.5.6 Storage:

Storage at about 5°C for 24-48 hrs develops several desirable product properties such as body and texture. Low temperature ensures desirable shelf-life by slowing down physical, chemical and microbiological degradation. Cold storage and careful stock rotation play an important role in ensuring that the consumer always receives a uniform high- quality yogurt.

2.6 Some properties of natural standard, set and stirred style thick yogurt.

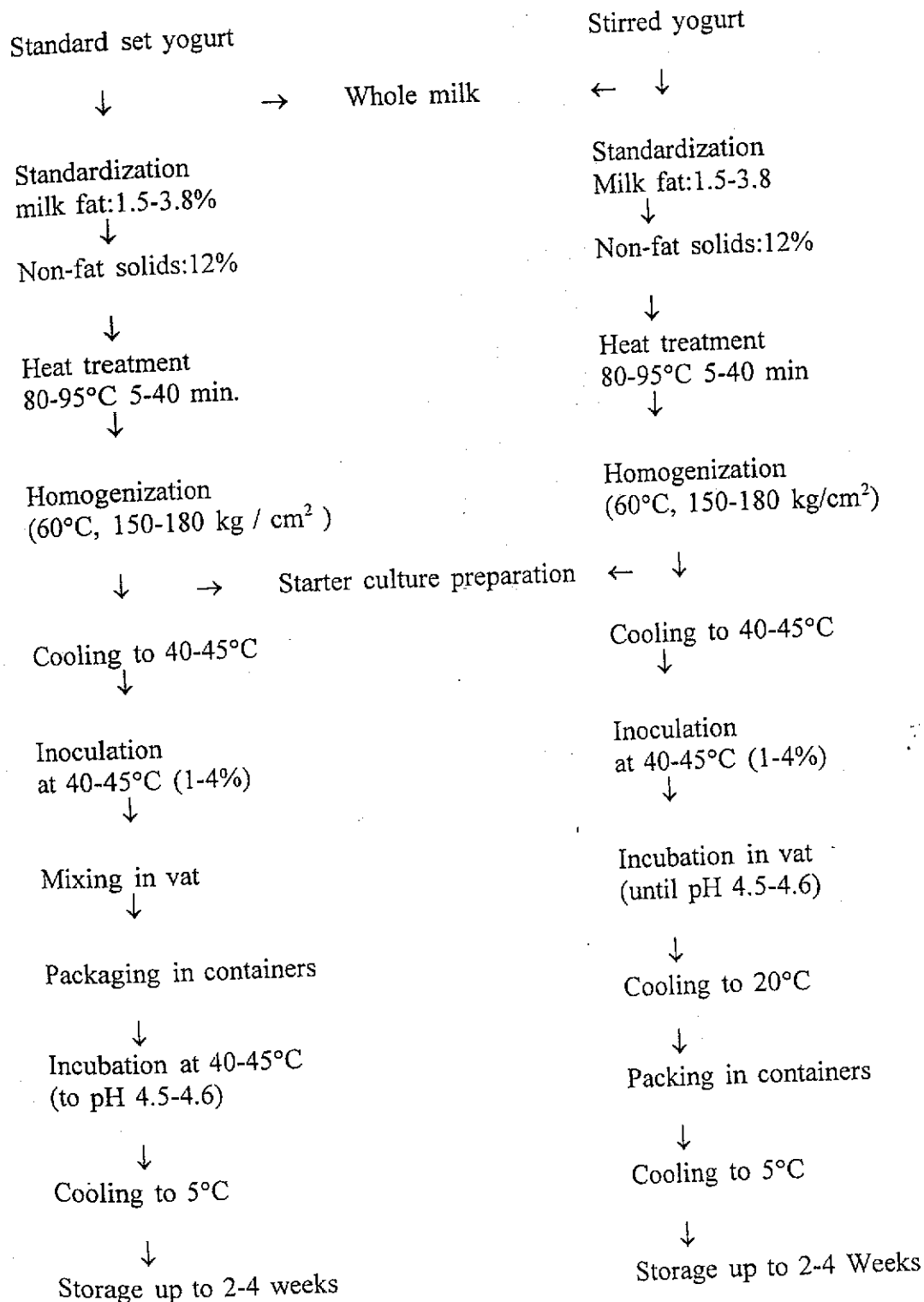
Yogurt is a fermented dairy product which is made by fermentation of milk with a mixed starter that consists of *S. thermophilus* and *L. bulgaricus*. Yogurt is the most consumed, best known and most studied product among the fermented dairy products. There are many published papers, reviews and text books about different aspects of yogurt (Humphreys and Plunkett, 1969; Rasic and Kurmann, 1978; Tamime and Robinson, 1985; Akin, 1987; Roginski, 1988; Vedamuthu, 1991; Chandan and Shahani, 1993).

2.6.1 Standard natural yogurt and its chemical and physical properties

The popularity of yogurt has now spread to most countries in the world. Existing and proposed legal standards for yogurt are well known both for the type of starter culture used and for the criteria of biological composition. In the criteria of chemical composition, yogurt is universally categorised into three types on the basis of fat content ; *i.e.* full-fat, half-fat, low-fat (FAO, 1984). However, according to the Turkish yogurt standard 1330 (Anon, 1989), it is categorized into four types on the basis of fat content *i.e.* full-fat (at least 3.8 %), fat (at least 3 %), half-fat (at least 1.5%), non-fat (less than 1.5 %). Additionally, its other properties required are non-milk solids contents not less than 12 %, titratable acidity not less than 0.8 % and not more than 1.6 % (expressed as lactic acid), and maximum mould and yeast counts 100 *cfu/g*, with no *E. coli*. The product labelled as yogurt should satisfy all the aforementioned criteria. The starter cultures consist of mixed cultures of only *S. thermophilus* and *L. bulgaricus*. In the above standard the presence of "live" starter organisms in the end product is stipulated.

This type of yogurt can be produced in either the set or stirred form. However , the set type of yogurt is more popular in the Turkish food market. Basic production methods for set style yogurt can differ widely. The flow diagrams of the procedures for set and stirred type of yogurt are given in Fig (2.2). The pre-treated milk (clarified, standardized of total solids of fat and non fat milk solids content) is heated in a batch tank or plate heat exchanger at 80 - 95°C for 5 - 40 min. Then it is cooled to 40 - 45°C, inoculated with 1 - 4 % yogurt starter culture (*S.thermophilus* and *L.bulgaricus*) and then filled into retail containers. Batch incubation for stirred yogurt is at 40 - 45°C for 3 - 5 hrs. (pH of 4.5 - 4.6). It is then cooled to 5°C and stored at 5°C. Its shelf life is about 2-4 weeks depending on production conditions.

Figure 2.2 The flow diagram outline for the production of set and stirred type natural yogurts in Turkey.



Cow's milk yogurt is made on a commercial scale in all over the world, but ewe's milk yogurt is made on a commercial scale in limited countries (e.g. Turkey, Germany). Therefore, the properties of ewe's milk yogurt do not seem to have been sufficiently investigated.

Manus (1974) summarised common problems in yogurt production as following:

- i) Syneresis will occur with low whey protein denaturation. This allows minimum water holding capacity of the coagulated proteins.
- ii) When inoculation temperature is high fermentation will be affected due to injuries to the cultures.
- iii) Producing high pH yogurt has inadequate protein binding characteristics which causes to syneresis. Rough handling can also cause syneresis.

The effect of several processing factors on yogurt quality was studied in a series of experiments by Dolezalek and Vokacova (1981). In the temperature range studied (70 - 90°C), heating milk to 80 - 90°C with a 10 min holding time and 2 % starter cultures inoculum at 42°C for 3 hrs incubation was optimum to maximise flavour and firmness.

Chemical and microbiological changes occurring during the storage of yogurt were investigated by Ottogalli et al (1972). Samples of plain yogurt (full-fat or 1 % fat) were stored for 60 days at either 4 or 20°C. At 4°C storage, the plain yogurt showed a gradual reduction in lactic acid bacteria and an increase in total free amino acid (from 36.31 to 38.67 and from 35.39 to 36.39 mg / 100 ml ultrafiltrate). At 20°C storage, the lactic acid bacteria disappeared after about 30 days and there were marked increases in free amino acids. It was concluded that all the yogurt retained their organoleptic characteristics unchanged for $\geq 30 - 45$ days.

The chemical and microbiological properties of plain non fat yogurt were also determined by Layer et al (1993) after 2, 6 and 12 days storage in a refrigerator. The results reported that the viable starter culture bacteria ranged from 14×10^7 -

80×10^8 cfu/g, and the ratio of *L. bulgaricus* to *S. thermophilus* ranged from 0.18 to 15.4. pH ranged from 4.5 - 4.2 during the 12 days storage period and the titratable acidity ranged from 0.9 to 1.3 % lactic acid. At the end of the storage period lactic acid increased slightly to 1.09 - 1.40. As major flavour compounds, acetaldehyde, diacetyl, acetoin were determined.

The bacteriological and biochemical characteristics of various type of commercial yogurt and laboratory scale yogurt prepared from cow's and ewe's milk were investigated by Kehagias et al (1984). The results were reported as the following: The viable counts of *S. thermophilus* and *L. bulgaricus* in most instances exceeded 10^8 cfu/ml. During storage at 5 - 7°C for 2 weeks, the average viable counts decreased in both types of yogurt samples. The average lactose activity declined only in the commercial yogurts during cold storage. Yogurt made from ewe's milk had double the lactose activity of cow's milk yogurt.

Kehagias et al (1986) studied the physico-chemical properties of set type yogurt made from cow's, ewe's and goat's milk. It was heated to 82 - 85°C for 30 min, before it was made into yogurt. All the yogurt samples were stored at 5 - 7°C for up to 10 days. The chemical composition and physical structure of the yogurts were determined. The result were reported as the following:

- i) ewe's milk yogurt had the highest firmness and viscosity and least amount of whey separation when compared with the other types of yogurts (cow's and goat's milk yogurt).
- ii) ewe's milk yogurt had highest total solids level and fat content because of raw milk composition.

Kehagias et al (1987) reported that milk from cow's, ewe's and goats' was inoculated at 2.5 % with three different commercial yogurts culture (Wiesby V, CH-1 and B3 -Rediset). It was then incubated at 42°C for 6 h and stored for 15 days at 4 - 7°C. The increase in titratable acidity and decrease in pH during incubation period was greatest in ewe's milk and least in cow's milk. During the storage

period, acidity of yogurt increased from 0.8 - 0.9 % lactic acid for cow's and goat's milk and 1.0 - 1.1 % lactic acid for ewe's milk yogurt to 0.8 - 1.0 % and 1.3-1.45 % lactic acid. The final pH was about 4 in all cases. Overall, acetaldehyde tended to decrease with goat's and cow's milk yogurt and remain the same or rise slightly with ewe's milk. Acetaldehyde concentration tended to be the highest with cow's milk. Firmness was greatest with ewe's milk yogurt and did not change during storage time.

Yogurt often contains undesirable organisms that cause mould, "off- flavours", short shelf-life and poor texture in the final product. Failure of the bacteria culture to carry out the complete fermentation process and to produce the desirable acidity occurs frequently because the fermentation reaction depends solely upon living organisms and any deficiency in the environment or in the necessary factors for adequate growth may destroy effectiveness of the bacterial culture.

Yogurts are a selective environment for the growth of yeast because of their low pH (Davis, 1975). The spoilage of yogurt by yeasts is generally recognised by the development of yeast off-flavours, loss of the texture quality due to gas production and the swelling and eventual blowing off the product container (Davis, 1975; Kroger, 1976). When yogurts are produced by "good manufacturing practice" they should contain no greater than 1 *yeast cell* / g. If it is correctly stored under refrigerator (+5°C), a product shelf life of 3-4 weeks may be expected. However, examination of yogurts randomly purchased at retail outlets in the UK has shown that 25 % of samples contain greater than 10^3 *yeast cell* / g while some samples exhibited yeast counts as high as 10^5 *yeast cell* / g (Davis, 1974; Davis, 1975). Similar results were observed by Suriyarachia and Fiecht (1981).

Dannenberg and Kessler (1988 a) investigated the effect of the denaturation of β -lactoglobulin (β -Lg) on whey drainage of set-style non fat yogurt. Skimmed milk was heated and held for various periods of time at temperatures ranging from 70-130°C. The results showed that the susceptibility to syneresis decreased with an

increasing degree of denaturation of the β -Lg. Comparable amounts of drained whey were determined at equal denaturation rate. The results proved to be independent of the temperature of heating. The close correlation between the consistency of the yogurt gel and the degree of denaturation of β -Lg became especially evident. It was concluded that when denaturation and complex formation reaches a maximum, a further increase in the severity of the heat treatment of the milk does not improve the water holding capacity of the yogurt gel. Firmness was generally higher the greater the degree of denaturation while the water holding capacity could further be improved at a degree of denaturation of 99 %, the firmness of the gel was found to decrease slightly. An increase in the holding time of the milk to 2.5 times that necessary for a degree of denaturation of β -Lg of 99 % even reduced the firmness of the yogurt gel (Dannenberg and Kessler, 1988 b).

The manufacture of high quality yogurt relies on the production of acid and flavour components at a time compatible with the manufacturing procedure. The acid produced must be accompanied by proper flavour development. Yogurt flavour depends on the acetaldehyde to lactic acid ratio. Too much acetaldehyde results in an undesirable green apple flavour, too much acid results in a sharp and unappetizing flavour (Moon and Reinbold, 1974). The ratio of these two compounds in yogurt is rather one-sided, with the concentration of acetic acid roughly one million times higher than the concentration of acetaldehyde.

Stefanova and G'oshova (1984) reported formation of volatile flavour compounds in Bulgarian yogurt. The yogurt was made from ewe's milk. pH decreased from 4.45 to 4.0 after 15 days at 4°C storage. During storage period flavour became weaker. Acetaldehyde, butanone and diacetyl appeared, in that order, when titratable acidity was in the range 32 - 48°T. The levels of all these compounds, except butanone, decreased during storage.

Hamdan et al (1971) investigated acetaldehyde production by using combined yogurt cultures. Acetaldehyde produced by three commercial yogurt cultures was

determined during seven hours of incubation at 45°C. Samples were stored at 4°C for 4 weeks. The results showed that all three cultures produced maximum acetaldehyde of 22 to 26 ppm at the fifth hour and declined thereafter. Acetaldehyde was reduced during storage by cultures RI (Hansen's) and 405 (Mosoley's) whereas acetaldehyde from culture 403 (Mosoley's) remained constant during two weeks of storage. When a single strain of *L. bulgaricus* and *S. thermophilus* was used, acetaldehyde was greatly reduced compared to that produced by combined commercial cultures.

The volatile flavour compounds of Egyptian yogurt was analyzed by Gaffar (1992) over a two week period at 8°C using a simple head space gas chromatography technique in order to study the changes and relate them to flavour acceptability. The results were reported that volatile compounds present were acetaldehyde, diacetyl, acetoin, acetone, butanone and acetic acid. Acetone and butanone disappeared within the first week of storage, whereas acetaldehyde, diacetyl and acetoin declined steadily but were still present after two weeks. Acetic acid increased to about twice its original level after 10 days of storage. The decreases in acetaldehyde, diacetyl, acetoin and the increase of acetic acid were closely related to the rapid decrease in product acceptability after 8-10 days storage.

The free amino acids content of yogurt made from cow's and ewe's milk is given in Table (2.4). During the manufacture of yogurt, a considerable increase occurs in the free amino acids content. An especially high increase are established in proline, followed by serin, alanine, valine, leucine and histidine. A small increase occurs in the free amino acids content during cold storage (Miller and Kandler, 1966). A study of free amino acids in yogurt made from different kinds of milk (cow's, goat's and ewe's milk) showed that the amino acid spectrum differs significantly among various yogurts (Rasic et al, 1971). This study is summarised as follows; the highest content of free amino acids is established in yogurt made from goat's milk followed by that made from cow's and ewe's milk, amounts were 33.19, 23.61 and 18.23 mg / 100 ml respectively.

Table (2.4) The amount of free amino acids in milk and yogurt (mg/100 g)

Amino acids	Milks		Yogurts			
	Cow *	Ewe *	Ewe *	Cow * **		UF*** Concentrate
Aspartic acid		0.2	1.8	7.8	7.5	10.0
Threonine	0.3	0.1	0.6	3.3	2.4	4.1
Serin	1.4	0.2	2.0	7.8	15.0	9.8
Glutamic acid	1.5	1.1	4.1	17.4	61.0	22.7
Proline	0.1	0.1	4.3	15.1	70.5	21.5
Glycine	0.3	0.2	0.3	5.4	2.8	7.6
Alanine	0.6	0.6	1.3	7.0	11.7	8.5
Valine	0.1	0.2	0.9	8.1	3.1	10.4
Methionine	0.1	0.1	0.2	2.8	0.8	3.7
Isoleucine	0.1	0.1	0.3	6.5	1.5	8.7
Leucine	0.3	0.2	0.5	9.6	5.8	13.1
Tyrosine	0.1	0.2	0.2	1.7	6.1	2.5
Phenylalanine	0.1	0.1	0.2	4.8	1.1	6.6
Lysine	0.3	0.2	0.7	10.1	7.2	14.2
Histidine	0.1	0.1	0.5	1.6	17.0	2.8
Arginine	0.2	0.3	0.9	0.8	-	1.7
Tryptophan	-	-	-	0.3	-	0.9
Total amino acids	5.4	4.0	18.5	110.1	213.5	148.8

Adapted from :

* Tamime and Deeth, (1980)

** Groux, (1972)

*** Nakazawa et al, (1991)

The concentration of the majority of amino acids significantly increased during yogurt making. The highest rate of increase was shown in yogurt made from ewe's milk, followed by yogurt made from cow's milk.

Luca (1974) studied improvement of yogurt quality. Batches of yogurt were prepared using 2-3 % of starter composed of *L. bulgaricus* and *S. thermophilus* (in 1:1, 1:2 and 2:1 ratios) and incubated at 36 - 37°C or 42°C. The best results, as judged by extent of amino acid liberation and acid formation, were obtained at both temperatures with the 1:1 ratio starter cultures. Total free amino acids contents were 70, 41 and 50 mg / 100 g on days 1, 3 and 5 respectively.

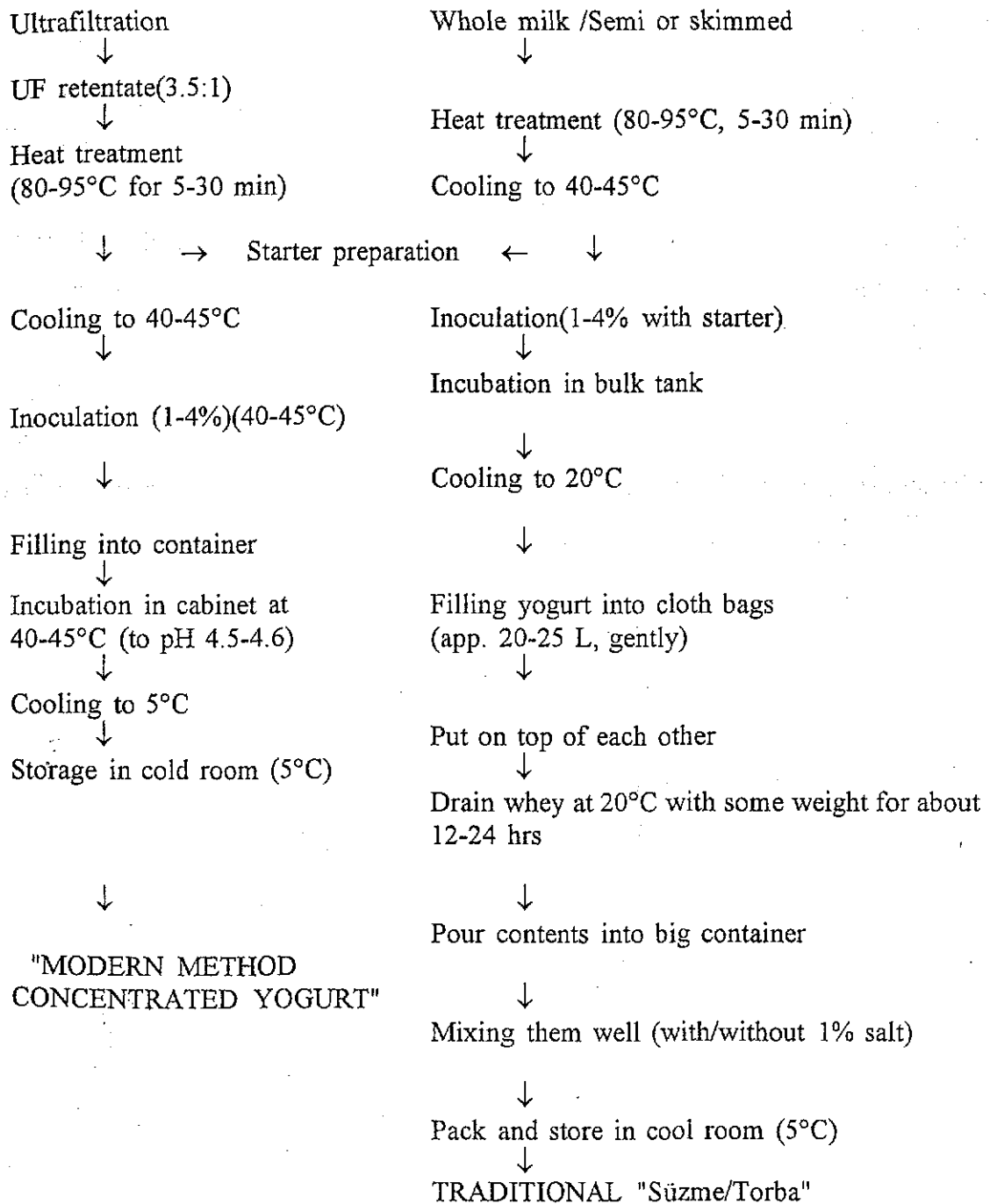
2.6.2 Thick yogurt, production methods of thick yogurt and its chemical and physical properties

Traditionally, cow's, ewe's, goat's and buffalo's milks have been used for the production of concentrated (or high total solids) yogurt in Turkey and the Middle East countries. Although cow's milk is widely used in Turkey, ewe's and goat's milk are very important for the production of concentrated yogurt in countries such as Lebanon and Syria where the climatic conditions are not favourable for the keeping of cattle (Juarez and Ramos, 1984).

Concentrated yogurt can be produced in various ways (Hofi, 1988; Tamime et al., 1987 a,b; 1991 a; Salji, 1991). Schema for concentrated yogurt manufactured by various procedure is shown in Fig (2.3). It is produced mainly by concentrating plain yogurt *i.e.* yogurt which is not sweetened or fortified.

The traditional concentrated yogurt is produced by using a cloth bag for straining the cold or warm yogurt in cold or normal room temperature. More recently the manufacturing method for concentrated yogurt has been replaced by mechanized processes where the warm yogurt is concentrated directly after the fermentation

Figure (2.3) Flow diagram outlining the production of concentrated Turkish "Süzme / Torba" yogurt



period to the desired level of solids by using a mechanical separator (Kharrazi, 1984; Dagher and Ali-Ghariebeh, 1985) or by ultrafiltration / reverse - osmosis (Veinoglou, 1978; Mogansen, 1980; Abrahamsen and Holman, 1980; Tamime et al, 1984). Concentrated yogurt can also be produced using reconstituted milk (Tamime and Robinson, 1978; Gilles, 1978).

2.6.2.1 Concentrated yogurt (" Süzme / Torba " yogurt)

Süzme yogurt is a high total solids concentrated yogurt with characteristics between those of stirred yogurt and fresh cheese. In recent times, the preparation of this type of yogurt has disappeared in many countries. Today it is still mainly produced in Turkey, the Middle East and in some European countries. This yogurt product is made under different name in different countries, e.g " Süzme / Torba " in Turkey; leben / laban / lebneh in the Middle East countries; " lactofil " in Sweden and " super yogurt " in Poland (Tamime and Robinson, 1985). The concentration is attained through traditional draining of yogurt in a cloth bag or, in industrial production, by the centrifugation of yogurt, or via membrane filtration (UF / RO) of milk or yogurt. The flow diagram of production of concentrated yogurt is given in Figure (2.3).

According to the Turkish Food Act of 1980 the general composition and properties of this yogurt must be as follows:

Water	70%
Total solids	30%
Fat	5% full-fat, 2.5% half-fat
Lactic acid	max. 2.25%
Salt	max.1.5% or without salt
Mould and yeast count	max.50 cfu/g
Pathogenic bacteria	None

2.6.2.2 Production methods of thick yogurt and its chemical and physical properties

2.6.2.2.1 Concentration by traditional method

Tamime (1977) studied the influence of certain factors e.g the level of total solids in the original milk, using different strains of the yogurt starter culture and the relationship between the level of total solids in original milk and the rate of whey drainage during the process of concentration of yogurt using the tradition cloth bag method. It was concluded that milk with 16 % total solids was optimum as a starting mix for the production of labneh (ie. 22-23 % total solids and 1.6-1.7 % lactic acid). Labneh was made with three different strains of starter culture (CH-1, Boll-3 and RR) and the drainage times to achieve 24 % total solids were 14, 18 and 47 h respectively. Labneh made with code CH-1 starter culture had the firmest consistency as indicated by the penetrometer reading.

Rao et al (1987) studied mineral composition and microbiological qualities of labneh made from cow's milk by using traditional method. Mineral composition of labneh was 550, 1100 and 2860 mg/L for K, Ca and P respectively. The high level of these minerals was due to concentration effect of products even though there were loses of some minerals in whey during the draining periods. Lactic acid bacteria and counts of yeast and moulds in labneh decreased by 2 to 4 log₁₀ numbers during 6 months of storage

Hamad and Al-Sheikh (1989) investigated the effect of added solids-non fat to milk for concentrated yogurt preparation, the effect of draining temperature on the yield and quality of the finished product. The fresh milk containing 12.5 % total solids was used as a control. Skimmed milk powder was used to fortify the solids non fat content of the fresh milk to 14.5 %, 16.5 % or 18.5 % total solids. It was concluded that an increase in yield was obtained when the labneh was drained at 5°C than at

25°C. There was a slight elevation in total acidity when labneh was drained at 25°C. Labneh made from fortified milk was characterised by having a smooth texture and continuous body with a good spreadability as compared with the control which had a thick and lumpy texture.

Kehagias et al (1992) studied effect of pH on the yield and solids recovery of strained yogurt from goats and cow's milk. The strained yogurt was prepared in a laboratory scale production from cow's milk and milk from indigenous goat breeds by a procedure resembling the industrial method used in large dairy plant. After fermentation period, the pH of set type yogurt was measured. It ranged from 4.1 to 4.9. The result indicated significant variation of the yield of strained yogurt, total recovery of milk solids in it and to the losses of nitrogen in serum. The yield and recovery of milk solids were higher at a lower pH range of 4.3 to 4.1. Goat milk was richer than cow's milk in all constituents. However, the Instron penetration force of the yogurt gel from goat milk was lower compared with that from cow's milk (this was measured just after fermentation). The yield and the recovery of milk solids in strained yogurt from goat milk was higher than the cow's milk.

2.6.2.2.2 Concentration by mechanical separator

Different mechanical separator are used for the production of concentrated yogurt. According to Tamime (1978 a) a nozzle separator has been used successfully for the production of concentrated yogurt with physical properties similar to traditional concentrated yogurt.

According to manufacturer of these separators when using this method, it is essential that skimmed milk yogurt is used rather than full fat milk yogurt because the presence of fat can block the nozzle opening. After fermentation natural skimmed yogurt is stirred vigorously, warmed up to 50-60°C, cooled to 32-42°C and

concentrated to 18% total solids by using the nozzle separator. The concentrated product is cooled to $<10^{\circ}\text{C}$, blended with cream and packaged (Robinson and Tamime, 1986; Tamime and Robinson, 1988). A similar process has been reported by Salji (1983) for the production of labneh in the Middle East. Ordinary mechanical separators have been used for the production of labneh (Kharrazi, 1984; Dagher and Ali-Ghariebeh, 1985), but the process has not been used commercially.

Other fermented dairy products, which have been concentrated by using mechanical separators are Skyr (in Iceland) and Shirkhand (in India). During the manufacture of the former product, the nozzle separators have been employed while a basket centrifuge has been used for the production of shirkhand (Tamime and Robinson, 1988).

2.6.2.2.3 Concentration by membrane filtration method.

There are several ways of applying UF in production of yogurt.

- Pre-concentration of milk (whole or skimmed).

- Pre-concentration of reconstituted milk.

- Addition of UF concentrated liquid (retentate) whole or skimmed milk to yogurt milk.

- Addition of powdered milk protein concentrates or whey protein concentrates in liquid or powdered form.

The manufacture of concentrated yogurt by using the UF technique was first reported by Veinoglou et al (1978). In this study, the milk was concentrated by UF to different levels (e.g. 6.5 to 9.5 % protein and 7.5 to 13.5 % fat). Best results of concentrated yogurt were obtained from UF milk retentate containing 8.5 % protein and 9.5 % fat. This was similar to the traditional yogurt in the overall chemical composition and organoleptic qualities. Therefore, good quality strained yogurt could

be produced from UF milk that has been standardised. Consequently, the method of processing could be mechanised.

The effect of ultrafiltration on lactic acid fermentation of milk was investigated by Tomita et al (1985). It was reported that whole milk was concentrated up to three times by using UF unit. Retentate was homogenized, heated to 90°C for 30 min, inoculated with commercial yogurt starter (CH-1) and incubated at 40°C for 6 h. Growth rates of both *S. thermophilus* and *L. bulgaricus* increased as the degree of concentration of retentate increased. The highest ratio of *S. thermophilus* and *L. bulgaricus* was obtained after 2h growth in retentate verses 3 h in milk. During growth, the increases in the acidity (lactic acid %) were greater in retentate than milk, becoming greater as concentration increased. Retentate from milk concentrated two fold or three fold formed much harder curd than unheated milk. However, the overall results showed that UF retentate are promising materials for yogurt manufacture.

In yogurt made from milk concentrated 1.4 fold (relative to protein) by UF, the lactic acid content is increased from 0.77 % in the control samples up to 0.95 %. The greater lactic acid formation is not caused by a higher lactose content but has to be attributed to the increased buffer capacity due to the increased concentration of protein and minerals (Renner and Eiselt-Lomb, 1985). The subsequent amount of lactic acid during the first week of storage was more pronounced in UF yogurt than in samples where potassium caseinate had been added to the milk (change of pH in UF yogurt from 4.5 to 3.9, in the other samples from 4.55 to 4.35).

Emaldi at al (1974) observed in yogurt, which was prepared from milk concentrated by UF and diafiltration, a high fermentation activity combined with high count of *S. thermophilus* and *L. bulgaricus*, but a lower proteolysis. Yogurt with a reduced activity could be made from milk by means of diafiltration but the lactose content was drastically reduced.

Aroma formation in yogurt manufactured by different techniques was investigated by Estevez et al (1988). Yogurt was made from pasteurized milk, the milk fortified with 5 % milk powder, and the milk ultrafiltered to half of its volume. The milk was inoculated by 1.5 % *L. bulgaricus* and 3 % *S. thermophilus*, and incubated at 45°C. Acetaldehyde production began earlier and reached higher levels in the UF milk (24.98 ppm) than in the other samples (14.76 and 22.8 ppm). The aroma was superior after storage for two weeks (20.59 ppm vs 13.06 and 16.47 ppm for natural and milk powdered fortified).

Yogurt cheese called labneh was manufactured by El-Samragy and Zall (1988) from UF cow's milk. The retentate was heated to 85°C for 20 min and cooled immediately to 45°C. Salt and yogurt starter culture were added at a rate of 0.3 and 3 % respectively, mixed and incubated at 42°C for 2-3 h. At the end of incubation time the concentrated yogurt was placed in cold store. The results showed that the product free from coliform, yeasts, mould and the total counts of lactic acid bacteria was around 4.6×10^7 cfu/g. Fresh and refrigerated samples were evaluated weekly for appearance, flavour, sourness, consistency and overall acceptability by an American and Egyptian test panel. There were no significant differences between the two groups until the fourth week on the base of appearance and sourness. The flavour was acceptable to most panellists, some remarked that the flavour of the sample was rancid.

Hofi (1988) produced concentrated yogurt from the UF cow's milk retentate. The UF retentate (19 % total solids) was produced from pasteurized cow's skimmed milk by using UF unit at 50°C. Cream containing 36 % fat was then added to retentate to attain a mixture containing 10 % milk fat. The yogurt was then produced from this mixture which had 26 % total solids using standard yogurt making procedure. It was concluded that this method seemed to produce high yield, the increases being about 11 %, a shorter production time, about 25 % less than traditional method. Finally avoiding wheying problems, the UF results was

characterised by a smoother body, texture and better palatable taste.

Ultrafiltration carried out at elevated temperature ($> 45^{\circ}\text{C}$) increases the fouling rate of the UF membranes (Attia et al, 1991 a, b) which may effect processing conditions in large scale operations where the equipment requires to be washed more frequently. However, the UF of yogurt at such high temperatures was successfully used to produce labneh within the shortest processing time of all the production methods. The firmness of product was similar to the traditional labneh.

Rasic et al (1992) investigated nutrient of yogurt from "low lactose" milk using a combined lactose UF retentate. The results showed that the removed K, Na, Ca, P from ultrafiltrated milk was approximately equal to that in whole milk yogurt but was about 39% lower than in whole milk yogurt fortified with 4 % skimmed milk powder. Concentration of calcium in low lactose yogurt was 30 % higher compared to whole milk yogurt and was approximately equal to milk yogurt fortified with 4% skimmed milk powder. Phosphate in low lactose yogurt was about 5 % higher compared to whole milk yogurt. Sodium was also greatly reduced in low lactose yogurt. Setting time for coagulation of yogurt was i) for control 3 h, ii) for reconstituted retentate 3.55 hrs.

The rheology of strained yogurt made from cow's milk by three different methods was reported by Tamime et al (1989 a, b) as following: total solids contents of the products were within the range of 21 to 24.2 %, 6.8 to 8.2 % protein and 9.2 to 10.5% fat. Rheological properties such as consistency of traditional labneh and UF labneh was very firm ^{and no syneresis was observed after breaking the coagulum} its texture was crumbly. Syneresis was noticeable after the coagulum had been broken. The best sensory attributes were found with the UF labneh followed by the traditional labneh whereas the retentate labneh appeared not to be satisfactory.

* The coagulum of the UF retentate labneh was very firm and

The effect of processing temperatures on the quality of labneh made by ultrafiltration was reported by Tamime et al (1991 c). In summary twelve batches of labneh were manufactured by using the traditional method and also by UF of yogurt at temperatures 35-55°C. The labneh samples contained 21.3 - 26.4 % total solids (including 6.6 - 9.3 % protein, 9.8 - 12.6 % fat, minerals and unmetabolised lactose). The highest flux rate during ultrafiltration was observed at temperatures $\geq 50^{\circ}\text{C}$ but the total viable counts of starter bacteria were lower when compared with labneh concentrated at $\leq 45^{\circ}\text{C}$ for industrial scale operations.

Two types of labneh (high total solids yogurt) were investigated by Tamime et al (1991 a and b). Yogurts were made from cow's, ewe's and goat milk by using traditional and ultrafiltration methods. Traditional labneh was produced by straining yogurt in a cloth bag and UF labneh was made by UF of warm yogurt. All yogurt samples were smoothed by a passage through a lactic curd homogeniser. The labneh samples contained 20-22.5 % total solids, 6.7-8.2 % protein, 7.8-8.9 % fat, minerals and unmetabolised lactose. Homogenization markedly decreased firmness of the goats and ewe's labnehs and is therefore not recommended in industrial production. Firmness of labneh made from cow's milk was affected by homogenization to a lesser degree. Homogenisation increased the pore size of goat's and ewe's milk labneh. There was an apparent relationship between pore size and firmness. It was concluded that there was marked differences in the firmness of goat's, ewe's and cow's milk labnehs in spite of their similar total solids contents. The uniformly structured cow's milk labneh made either by the traditional method or by UF was firmest, whereas the goat's labneh contained large void spaces and was the softest, also irrespective of the method of manufacture. Homogenization of the goat's and ewe's milk labnehs produced a smoother texture but markedly decreased the firmness.

Manufacture of yogurt from milk concentrated through UF was investigated by Nakazawa et al (1991). The total solids of UF concentrate and control yogurt was

18.38 % and 11.75 % respectively. Their results showed that UF concentrate had more starter activity at 45°C for 6 h incubation period (1.2×10^{10} cfu/ml) than the control yogurt (8.1×10^8 cfu/ml). Starter culture produced more lactic acid for UF concentrate than control yogurt (1.48 and 1.2 % lactic acid respectively). Total free amino acids were 160.4 mg % UF concentrate and 117.8 mg % for control yogurt.

2.6.2.2.4 Production method of concentrated yogurt from reconstituted milk

Beyer and Mair-Woldburg (1974) reported that a yogurt with 20 % total solids resulted in good quality labneh. Interesting comment was also made that although the consistency of the initial yogurt depended to a very large extent on the dried milk used, labneh could be produced from any yogurt as long as this was not made from milk powder containing off-flavours.

In New Zealand a labneh producing procedure has been developed by Gilles, (1978). This involves the initial development of a high-heat whole milk powder with a reduced lactose content. The powder could be reconstituted to a total solids level up to 30 % with a lactose level in the finished product comparable to traditional labneh (2-4 % lactose). In contrast, standard whole milk powder could not be reconstituted to same total solids levels while the higher lactose level (10-12 %) imparted too sweet a flavour to the finished product.

Abd-El-Salam and El-Alamy (1982) produced yogurt and concentrated yogurt from UF recombined skimmed milk powder. The skim milk powder was reconstituted to 10 % total solids and concentrated, by using UF, to four fold. Anhydrous milk fat was added to the retentate and the mixture was homogenised. The milk retentate containing 25 - 26 % total solids and 8 % fat was diluted with the permeate at different proportions (*i.e.* 16-24 % total solids) to study the effect on the quality of

concentrated yogurt. Each blend was heated to 90°C for 2 min, cooled to 40°C and inoculated with a yogurt starter cultures at rate of 2 %. Incubation was carried out at 41°C for 3 hrs and cooling took place in a refrigerator. Acid development, consistency and wheying off were measured and the organoleptic properties of the end products were evaluated. It was concluded that a good quality natural yogurt could be produced from UF retentate containing 16 % total solids, and concentrated yogurt 24 % total solids had a tough texture and lacked the desirable "elasticity" of traditional concentrated yogurt.

2.6.3 Microbiological properties of yogurt

Milk is the normal habitat of a number of lactic acid bacteria which cause spontaneous souring of milk held at bacterial growth temperatures for an appropriate length of time. Depending on the type of lactic acid bacteria gaining entry from the environmental sources (air, utensils, milking equipment *etc.*), the sour milk attains uncontrollable flavour and texture characteristics. Modern industrial processes utilise defined lactic acid bacteria as a starter for yogurt production.

A starter consists of food grade microorganisms that on culturing in milk produce predictable attributes characterizing yogurt. The composition of yogurt starter cultures is shown in Table (2.5). Also shown are the additional organisms found in yogurt or yogurt like products marketed in various parts of the world. Most of the yogurt in the world is fermented with *S. thermophilus* and *L. bulgaricus*. In Europe and Japan, optional bacteria, especially those of intestinal origin, are incorporated in the starter or the product. *S. thermophilus* and *L. bulgaricus* are fairly compatible as well as symbiotic for growth in milk medium. However, the optional organisms do not necessarily exhibit compatibility with *S. thermophilus* and *L. bulgaricus*. Nevertheless, product characteristics, especially flavour, are significantly altered from traditional yogurt when yogurt culture is supplemented with optional bacteria.

Table (2.5) Required and optional bacteria of yogurt starter cultures

Required by IDF standard for yogurt	Optional additional bacteria used or suggested
<i>S. thermophilus</i>	<i>L. acidophilus</i> , <i>L. casei</i> , <i>L. lactis</i>
<i>L. bulgaricus</i>	<i>L. helveticus</i> , <i>Bifidobacterium bifidum</i> , <i>L. jugurty</i> , <i>B. longug</i> , <i>B. infantis</i>

2.6.3.1 Taxonomy of yogurt starter cultures

L. bulgaricus was first described by Orla-Jensen (1919) as *Thermobacterium bulgaricum*. It is presently considered as one of the subspecies of *L. delbrueckii*. *S. thermophilus* was also first described by Orlo-Janson (1919). *S. salivarius subsp thermophilus* is the new name proposed to designate *S. thermophilus* which stands apart from the other streptococci and especially lactic streptococci, at present designated as lactococci. It is exclusively, isolated from the dairy environment, ferment only few carbohydrates, ie lactose, sucrose, glucose and is characterized by its thermoresistance and rather high growth temperature which may reach 50-52 °C.

Thermophilic lactic starters generally contain one or several lactobacilli (*L. bulgaricus*, *L. lactis*, *L. helveticus*) and one streptococcus which is generally *S. thermophilus*. The yogurt starter cultures involved in the production of yogurt are distributed in different phenotypic and genotypic groups. These groups are characterized by different nutritional requirements, metabolic, cultivational and technological properties. The main distinctive characteristics of these lactic acid bacteria are given in Table (2.6)

In the present study, the former names of yogurt bacteria, i.e. *S. thermophilus* and *L. bulgaricus* will be used for purposes of convenience.

2.6.3.2 Biological aspect of the association between *S. thermophilus* and *L. bulgaricus*

The symbiotic relationship between the two bacteria *S. thermophilus* and *L. bulgaricus* is extremely important to the yogurt process and was first observed over half a century ago by Orla-Jenson (1919). The growth association between the two organisms of the yogurt starter cultures (*S. thermophilus* and *L. bulgaricus*) is called a symbiosis, and this relationship has been reported by many researcher.

Oner and Erickson, (1986) indicated that interaction between yogurt bacteria are very complex and greatly depend on strains involved. The schema of symbiosis of yogurt bacteria is given in Fig (2.4). A positive interaction is observed between *S. thermophilus* and *L. bulgaricus* in mixed culture, leading to the stimulation of growth and acid production of both bacteria compared to their single strain cultures (Pette and Lolkema, 1950 a; Accolas et al, 1977). Mixed yogurt starter cultures may also stimulate the production of some metabolites such as acetaldehyde (Hamdan et al, 1971; Bottazzi et al, 1973) and influence carbohydrate utilization.

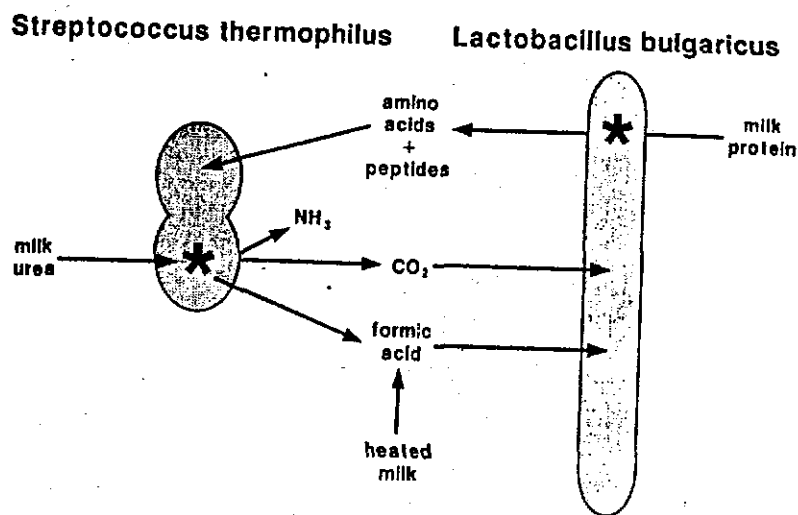
Lactic acid bacteria need a lot of essential amino acids for growth because of their inability to synthesize them from a more simple source of nitrogen. *S. thermophilus* does not possess substantial extra cellular proteolytic activity and the amino acids and free peptide content of milk is not enough to promote its full growth. Milk has only 25 - 35 % of the leucine needs of *S. thermophilus* (Thomas and Mills, 1981). *L. bulgaricus* proteases break down casein and supply the *S. thermophilus* with amino acid and peptides (Pette and Lolkema, 1950 b; Bautista et al, 1966; Shankar and Davies, 1978; Radke-Mitchell and Sandine, 1984).

Table (2.6) The main taxonomic properties of *S. thermophilus* and *L. bulgaricus*

Characteristics	<i>S. thermophilus</i>	<i>L. bulgaricus</i>
(G+C) content of DNA	Close to 40 %	Close to 50 %
Morphology	Spherical or ovoid cell in pairs or long chains; Polymorphism in old cul- tures 0.7-0.9µm diameter	Variable with age, medium, strain. Metachromatic granules in old cultures slender rod, 0.8-1µm wide, 4-6µm long
Ecology	Milk and dairy product only	(-)
Growth at 45°C	(+)	(+)
at 10°C	(-)	(-)
in 2% NaCl	(+)	(-)
in 4% NaCl	(-)	(-)
Resistance of tem.(°C)	at 65°C for 30 min (+)	(-)
Fermentation	Homofermentative	Homofermentative
Lactic acid isomer	L-(+)	L-(+),D-(-),DL
Mucopolysaccharide	(+)	(-)
Catalase	(-)	(-)
Granules	(-)	(+)
Acid in milk	0.7-1.0% lactic acids	generally ≤1.8 %
Acid from:		
Glucose	(+)	(+)
Galactose	(+)	(-)
Lactose	(+)	(+)
Sucrose	(+)	(-)
Maltose	(±)	(-)

Adapted from: Bergey's Manual of Determinative Bacteriology 9th edition (1986)

Fig (2.4) The schema of symbiosis of yogurt bacteria



The stimulative effect of amino acids results in a shortened generation time of *streptococci* cell and increased numbers. As a result, the streptococci grow faster in the early part of incubation, outnumbering the *L. bulgaricus* 3-4 to 1 after the first hour. In the later stage, the growth of *S. thermophilus* is slowed down due to the adverse effect of lactic acid so that the number of *L. bulgaricus* gradually approximate to those of *S. thermophilus*

The growth of *L. bulgaricus* is stimulated by a compound produced by *S. thermophilus* which seems to be formic acid (Gallesloot et al, 1968; Shankar and Davies, 1978). The production of formic acid by *S. thermophilus* was confirmed by Veringa et al (1968). Higashio et al (1978) reported a combined stimulating effect of formic and pyruvic acids. Bottazzi et al (1971) demonstrated that the presence of formic acid in the milk increases the ratio of rods to cocci.

Driessen et al (1982) have shown CO_2 has a stimulatory action on *L. bulgaricus*. CO_2 might be involved in the synthesis of aspartic acid and decarboxilation of urea contained in milk through urease.

* *S. thermophilus* produces CO_2 which is necessary for growth of *L. bulgaricus* (Fig 2.4)

for growth of *L. bulgaricus* (Tinson et al, 1982). In this way, it can stimulate growth of lactobacilli. The quantity of CO₂ dissolved in milk decreases after heat treatment and so the remaining CO₂ is too low to meet the requirement of the *L. bulgaricus* (Driessen et al, 1982) .

2.6.4 Some important metabolic activity of yogurt starter cultures

2.6.4.1 Carbohydrate metabolism

Energy is required by microorganisms to maintain their life cycle. Such energy can be provided to the bacterial cell via different systems (Lavrence et al, 1976). The energy must be supplied mainly by fermentation of carbohydrate. As a carbohydrate, lactose is the only available carbohydrate in milk for this purpose.

Lactic acid bacteria, both rod and cocci, follow different pathways of carbohydrate fermentation. They achieve end products which are at least 90 and 50 % lactate for homofermentative and heterofermentative species, respectively (Kandler, 1983). *S. thermophilus* and *L. bulgaricus* are both homofermentative. The main products of fermentation is L-lactate for *S. thermophilus* and D-, L- or DL- lactate isomers for *L. bulgaricus*.

One of the most important problems for lactic acid bacteria growing in milk is the uptake of lactose. Most of the lactobacilli take up lactose by a specific permease and split it subsequently by β -galactosidas (β -gal) into glucose and galactose.

Lactose coming into contact with the cell membrane of bacteria is phosphorylated by the localized enzyme action of the so-called phosphoenol pyruvate-dependent phosphotransferase system (PEP-PTS).

It is then taken into cell, where lactose-P is hydrolysed by phospho- β -galactosidase (*P- β -gal*) into glucose and galactose-6-phosphate which are then metabolised into lactic acid (Hickey et al, 1986; Thomson, 1987).

Glucose is metabolised via the Embden-Meyerhof-Parnas (EMP) pathway (Fig 2.5) (Kandler 1983) to pyruvic acid, which in turn is converted to lactic acid via the action of lactate dehydrogenase enzyme present in lactic acid bacteria (Garvie, 1978). The mechanism of glucose metabolism by the lactic acid bacteria is now well understood, but the mechanisms of galactose metabolism is not yet clear. The galactose is converted to glucose by "Leloir pathway" and together with the glucose is fermented by glycolysis (Hickey et al, 1986). In any event, galactose accumulates in yogurt (Goodenough and Keleyn, 1976) and is not catabolized to any great extent. This may be because galactose is not metabolised in the presence of the readily fermentable glucose, which may suppress the Leloir and D-Tagatose-6P pathways. The majority of streptococci take up lactose and galactose by the action of the PEP. This is a phosphotransferase dependent system (Mc Kay et al, 1970; Thomson, 1980).

2.6.4.2 Protein metabolism

Fresh milk contains only small amounts of free amino acids and peptides. The total amount and the type of free amino acids present in yogurt and milk depend on several variables such as the strain of starter bacteria used, the age of product, the ratio between *S. thermophilus* / *L. bulgaricus* , the type of milk (cow's, ewe's milk etc.) and the production process employed (Mills and Thomas, 1980).

Lactic acid bacteria require several amino acids for growth (Law et al, 1976; Mills and Thomas, 1981). The proteolytic systems in lactic acid bacteria are important for making protein and peptide-N in the milk available for growth.

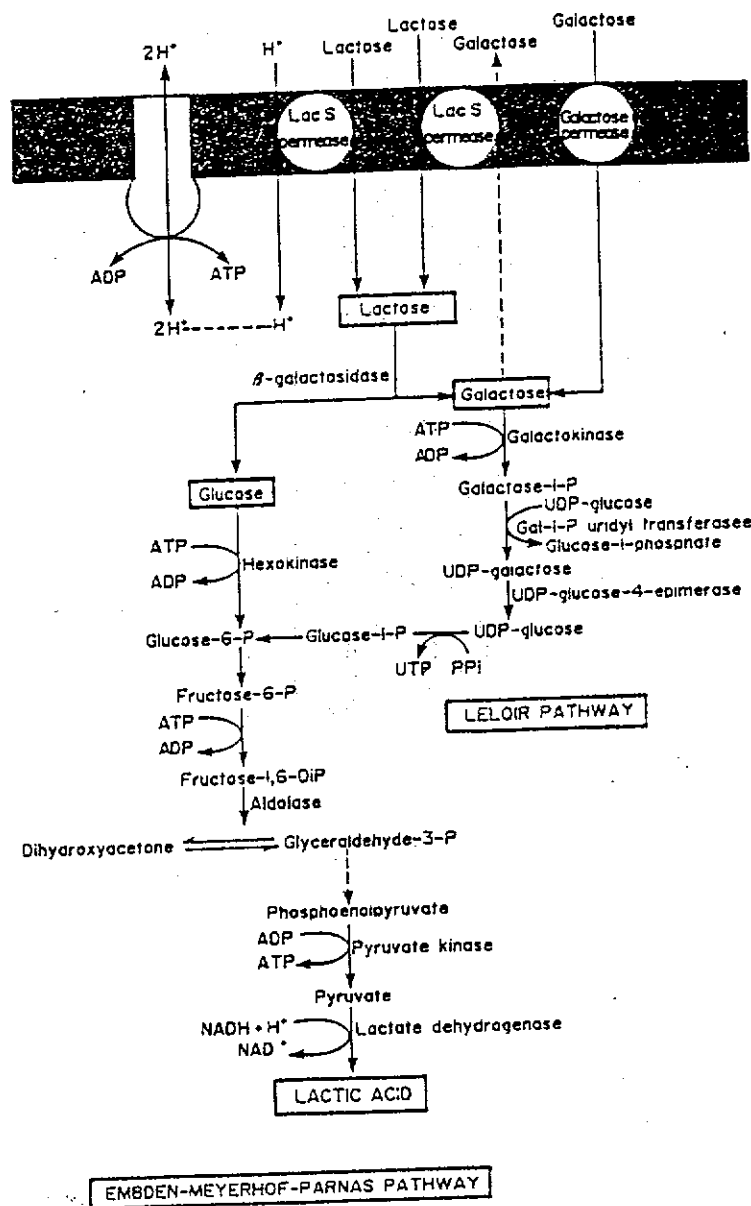


Fig (2.5) Scheme of lactose utilization by homofermentative bacteria.

Adapted from Zourari et al, (1992)

A detailed description of the proteolytic system of lactic acid bacteria have been published (Castberg and Morris, 1976; Law and Kolstad, 1983; Ono et al, 1992).

Lactic acid bacteria are weakly proteolytic compared with many other groups of bacteria (e.g. *Bacillus*) (Dellaglio, 1988). However, their proteinase systems are composed of a number of different proteinases and peptidases located in the cell wall, cell membrane and cytoplasm. The peptides are transported into the cell by specific systems and further broken down to amino acids inside the cell (Law, 1977 and 1978; Rice et al, 1978). Large peptidase are hydrolysed by endo- and exopeptidase associated with the cell membrane into smaller peptidase which are more easily transported into cell (Exterkate, 1975).

S. thermophilus and *L. bulgaricus* cause a significant degree of proteolysis during the fermentation which may be important for the following reasons:

- i) They lead to changes in the physical structure of the product.
- ii) Contribute to the production of flavour compounds (e.g. they act as precursors for several of enzymatic and chemical reactions which produce the flavour compounds).
- iii) The liberation of amino acids into the milk is important for the growth of *S. thermophilus* and probably improves the symbiotic relationship between *S. thermophilus* and *L. bulgaricus*.

The activity of peptide hydrolyses of the yogurt starters is affected by many factors such as:

- i) The ratio of *S. thermophilus* to *L. bulgaricus* which influences the amount of proteolysis
- ii) Their production is at a maximum during the logarithmic growth phase, and declines slowly after the stationary phase.

- iii) When the amount of cocci is greater than the rods, the hydrolysis of whey proteins in milk yields lower levels of non protein nitrogen.
- iv) Bitterness in yogurt is associated with the proteolytic activity of *L. bulgaricus* and such undesirable characteristics could be reduced if the fermentation is carried out at 38°C instead of 44°C
- v) High proteolytic activity is observed during the production of yogurt from lactose hydrolysed milk.

2.6.4.3 Formation of flavour compounds of yogurt

2.6.4.3.1 Flavour compounds of yogurt

The typical aroma of fermented milks mainly comes from a mixture of lactic acid and carbonyl compounds like acetaldehyde, acetone, acetoin, and diacetyl (Bottazzi and Dellaglio, 1967, Bottazzi and Vescovo, 1969), produced by *S. thermophilus* and *L. bulgaricus*. The production of carbonyl compounds by starter bacteria is given in Table (2.7) and the pathway leading to production of flavour compounds are outlined in Fig (2.6). In addition to carbonyl compounds, many volatile compounds have also been identified in yogurt, such as volatile fatty acid (Turcic et al, 1969) and several compounds derived from the thermal degradation of lipids, lactose and proteins during heat treatment of milk before yogurt manufacture eg. aldehydes, ketones, alcohols, lactone, sulphur compounds (Tamime and Death 1980).

Acetaldehyde is the main compound recognised as responsible for the flavour intensity of yogurt (Pete and Lolkema, 1950; Law, 1981). The amount of acetaldehyde required for the development of a characteristic flavour varies widely between dairy products.

Table (2.7) Production of carbonyl compounds (ppm) by yogurt starter cultures (Tamime and Robinson, 1985)

Organisms	Acetaldehyde	Acetone	Diacetyl
<i>S.thermophilus</i>	1.0 - 8.3	0.3 - 5.2	0.1 - 13
<i>L. bulgaricus</i>	1.4 - 12	0.3 - 3.2	0.5 - 13
Mixed cultures	2.0 - 41	1.3 - 4.0	0.4 - 0.9

There is no agreement on the acetaldehyde values to be obtained for optimum flavour. For some the optimum lies in the range 23 - 41 ppm (Marshall, 1982) while, for others, good flavour quality of yogurt contains 13 - 16 ppm, and still lower value can be encountered in commercial products (Badings and Necter, 1980; Green and Manning, 1982).

Diacetyl contributes to the delicate, full flavour of yogurt and seems to be important when the acetaldehyde content is low (Groux, 1973). According to Rasic and Kurmann, (1978) *S. thermophilus* is the only bacterium responsible for diacetyl and acetone production in yogurt. Diacetyl and acetone are very low, rarely reaching 0.5 ppm (Marshall, 1982). Acetone is considered to be of limited importance in the flavour of yogurt and usually originated from the milk itself.

2.6.4.3.2 Formation of acetaldehyde from glucose

The EMP- pathway generates pyruvate. An α_2 -carboxyles catalyzes the formation of acetaldehyde from pyruvate. An aldehyde dehydrogenase may also generate acetaldehyde from acetyl CoA which is formed from pyruvate by the action of a pyruvate dehydrogenase.

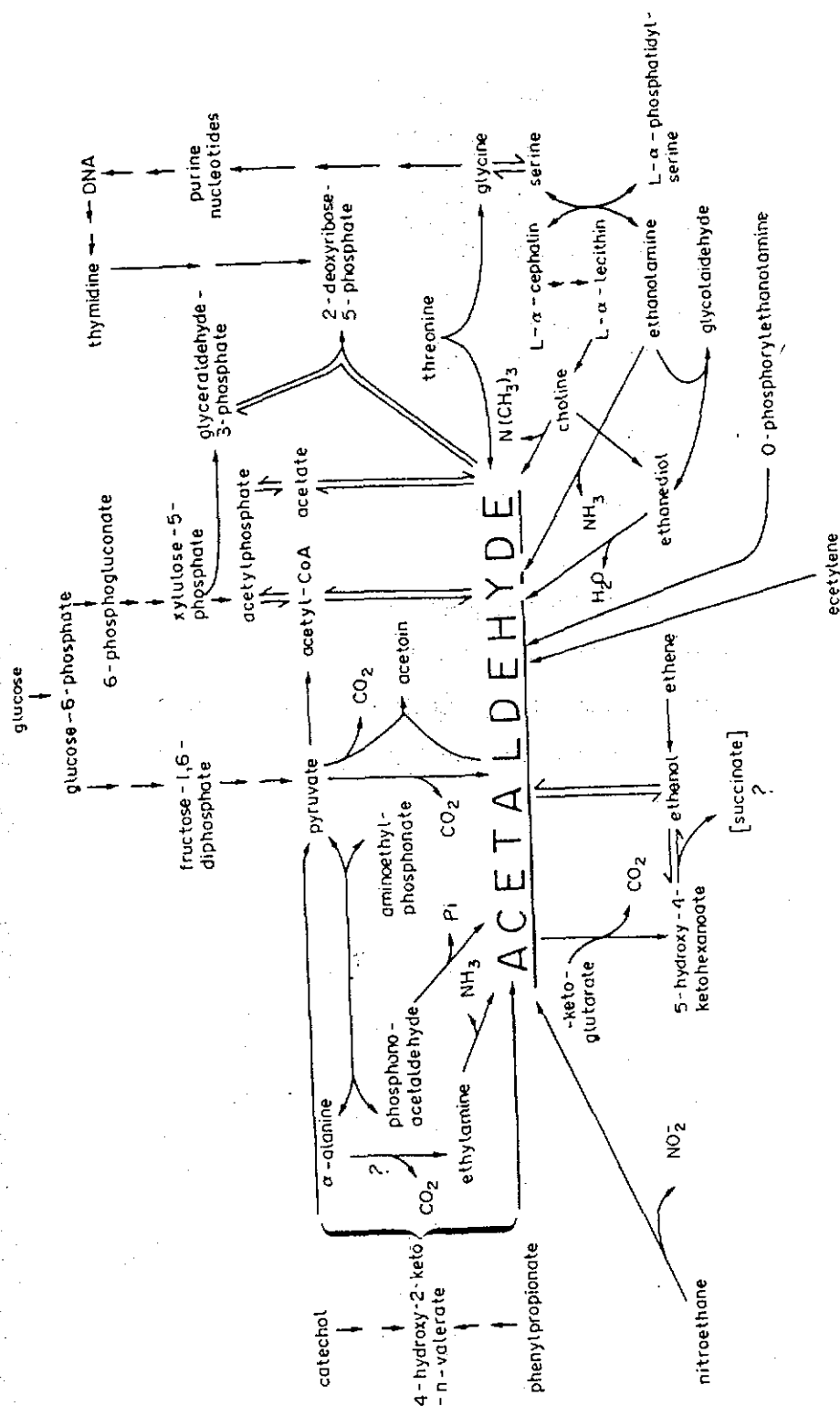


Fig (2.6) Diagrammatic representation of the known reactions involving acetaldehyde. Adapted from Lees and Jago (1978)

Reya et al (1986) have not detected significant α -carboxylase and aldehyde dehydrogenase activity from two strains of *S. thermophilus* and two strains of *L. bulgaricus*. However, an aldehyde dehydrogenase activity from four strains of each *S. thermophilus* and *L. bulgaricus* species had been found by Lees and Jago (1976).

2.6.4.3.3 Formation of acetaldehyde from protein

Pyruvate is an end product of the catabolism of many amino acids such as serin, cystine and alanine (Lees and Jago, 1976). Threonine aldolase is another acetaldehyde forming enzyme which has been detected by Raye et al (1986) in two strains of *L. bulgaricus* investigated. However, this enzyme was detected in both species by Lees and Jago, (1976) and Wilkins et al (1986).

The addition of threonine (0.6 %) or glycine (2 %) to the growth medium does not significantly affect the specific activity of threonine aldolase in the cell extract (Lees and Jago, 1976 b). High threonine concentration combined with low glycine concentrations in the growth medium increase the biosynthesis of the enzyme. When this ratio is inverted a decrease is observed. However, the threonine concentration during growth may have a greater effect on increasing threonine aldolase activity than the glycine concentration may have on inhibiting it.

2.6.4.3.4 Formation of acetaldehyde from nucleic acids

Two strains of *S. thermophilus* and one *L. bulgaricus* strain studied by Raye et al (1986) possess a deoxyriboaldolase which catalyzed the synthesis of acetaldehyde from 2-deoxyribose-5-phosphate. However, the synthesis of acetaldehyde via this enzyme is probably limited. This enzyme could be involved in the degradation rather than synthesis of DNA.

2.6.5 Nutritional and Therapeutic properties of yogurt

Yogurt consist of all components and nutrients among metabolites produced by fermentation by lactic acid bacteria. The composition of yogurt varies depending on the type of yogurt, source of milk (*i.e.* whole, skimmed, cow's, ewe's milk), season, fermentation period and the production conditions (heat treatment).

Yogurt is an exceptional source of high quality protein, Ca, P and K in the human diet. In addition, the starter culture of yogurt has appreciable lactose activity and thereby contribute to the digestion of a lactose intolerant person (Goodenough and Leys, 1976; Kilara and Sahahani, 1976).

In this section, the nutritional and physiological properties of yogurt will be briefly discussed.

2.6.5.1 Protein

Yogurt is more digestible than normal milk (Rasic et al, 1971 a,b. ; Stojslaljevic et al, 1971; Alm, 1982). The increased digestibility of yogurt protein compared with milk protein in milk has been attributed to several factors.

(i) Protein in yogurt is partly hydrolysed, they differ in configuration from the protein in milk. The protein in milk shows a complete micellar structure but in yogurt the micellar structures are broken down and the proteins are in amorphous form. Due to this, the form of small particles present a large surface to the digestive enzymes.

(ii) During the fermentation period and storage time the microorganisms break down a part of the protein into peptide and free amino acids causing, what might be called, predigestion of protein (Rasic and Kurmann, 1978).

(ii) contd.

Raw milk needs twice as much time as yogurt to reach the same degree of digestion (Alm, 1981; Breslaw and Kleyn, 1977).

The most important function of yogurt as protein food is to supply the amino acids necessary for the synthesis of tissue proteins so its nutritional and physiological value is decided by a good content of the amino acids essential for human and efficient digestion and absorption in the body (Yukuchi et al., 1992).

Whereas the free amino acids content of cow's milk is less than 10 mg / 100 ml, it is said to be 24 - 70 mg / 100 ml in yogurt (Yukuchi et al, 1992). The free amino acids in yogurt are used to some extent in the multiplication of the lactic starter bacteria; however, yogurt contains large amounts of the non-essential amino acids (eg, glutamic acids and proline in free form) because these are not used by bacteria. These are only accumulated in yogurt.

Good quality protein, particularly animal protein, is especially important in the diet of humans. Consumption of five hundred grams of yogurt a day will provide the recommended intake of essential amino acids for humans with the exception of methionine and cystine. In recent studies with rats, yogurt had the highest values for NPU (Net Protein Utilization) and PER (Protein Efficiency Ratio) at different levels of protein content, with maximum utilization at the 10 % protein level (Balasubamanya et al., 1984). The type of protein in yogurt which is responsible for this beneficial effect (Balanc, 1984).

The spectacular increase of non-protein nitrogen and of free amino acids in yogurt reflects proteolysis as a whole. The intermediary compounds are certain to have their importance on the nutritional level even though this has not yet been specifically demonstrated.

2.6.5.2 Lactose

The most important carbohydrate in yogurt is lactose. Lactose levels generally decrease in yogurt compared with milk. The decrease is 20 - 50 %, which is very important with regard to problems of lactose intolerance. Lactose is hydrolysed by lactic acid bacteria during fermentation. It is first hydrolysed to glucose and galactose but the glucose is then converted to lactic acid and very little remains in yogurt. On the other hand the majority of the galactose remains unchanged in yogurt.

Glucose, produced by the digestion of lactose in the body, is metabolised in the liver and part of it is stored as glycogen to be converted back to glucose and in the form of blood sugar to be used as an important energy source for tissues and cells in all parts of the body as necessary. Part of the glucose is utilized as a constituent sugar for organs like the liver, muscular tissue and nerve tissue. Galactose is the sugar best absorbed in the intestinal tract and is changed by metabolism to glucose in the liver. Part of the galactose contributes, as galactose, to the synthesis of brain cerebroside and nerve tissue.

The literature on the nutritional and physiological value of lactose and lactic acid in yogurt can be summarised as follows (Rasic and Kurmann, 1987; Blanc, 1986; Gilliland, 1991; Yukuchi et al, 1992; Fernandez et al, 1992).

- i) Lactose, glucose, galactose are an important energy source (4 kcal/g) for tissues and cell in all parts of the body.
- ii) They are incorporated as constituent sugars in organ, muscle and nerve tissue. Galactose, in particular, contributes to the synthesis of brain and nerve tissue in the early years of life.
- iii) Galactose increases the capacity to utilize fats.

- iv) Lactic acid in yogurt is a natural preservative, it makes yogurt biologically safer.
- v) Improving the digestibility of milk proteins by precipitating them in finer curd particles.
- vi) Lactose increase the capacity of the body to utilize phosphorus, calcium and iron.
- vii) Lactose stimulates gastrointestinal activity
- viii) Lactose promotes the production of lactic acid, acetic acid and other inhibitor agent by lactic acid bacteria, which in turn suppress the growth of putrefactive bacteria in the intestinal tract.

2.6.5.3 Fat

Milk fat is made up of triglyceride 97 - 98 %, phospholipid 0.2 - 1 %, cholesterol 0.2-0.4% and minute amount of fat-soluble vitamin, pigments and free fatty acids. The lipolytic activity of microbial cultures is generally limited (Alm, 1982). However, during the production of yogurt based on cow's milk, the relative content of many saturated fatty acids increases in comparison with that of initial milk but the total acid content decreases. Concentration of stearic, oleic, linoleic and palmito-oleic acid decrease during fermentation period. The effect of milk, fermented milks and their components on the blood cholesterol level have not been explained conclusively and some authors have put forward some doubts in this regard (Hussi et al., 1981; Massey, 1981). Nevertheless, a reasonable amount of these product in a balanced diet cannot have any undesirable effect on health. Oxidation products of cholesterol that can be formed in course of manufacturing and storage of yogurt could have ill effects on humans (Peng and Taylor, 1984).

The nutritional and physiological value of milk fat can be summarized as follows (Rasic and Kurmann, 1978; Alm, 1982).

- i) Milk fat has highest energy value (9 kcal/g).
- ii) A high digestibility of milk fat as compared with other animal fat is due to the low melting point, 31 - 36°C (for cow's milk fat).
- iii) It supplies essential fatty acids and water soluble vitamins (A, D, E and vitamin K).
- iv) Choline, which is a constituent of the phospholipid, promotes the oxidation of lipids in the liver and acts to maintain an equilibrium cholesterol concentration.
- v) Utilization of lipids in the body is considerably increased in the presence of lactose and protein.

2.6.5.4 Minerals

Virtually all of the minerals in yogurt comes from its raw material (milk, milk powder etc.). The contents of the major minerals differ considerably among products, sometimes being higher in yogurt than in milk because of fortification. The contents of minerals are hardly altered at all during the lactic acid fermentation of milk but there are many reports that the utilization of calcium, phosphorus and iron in the body is better than in the case of milk (Rasic and Kurmann, 1978; Gurr, 1987; Rusoft, 1987; IDF, 1991 b). This seems to be because the state of dissociation of Ca, P, Fe ions is changed by the production of lactic acid. However, there are as yet no reliable reproducible data. The utilization of Ca and P in the body is known to be raised in the presence of lactose and vitamin D and for this reason the utilization of Ca and P in milk is high. The minerals in fermented milk are mainly absorbed in the small intestine as soluble low-molecular weight compounds although small amounts are also absorbed from the stomach and the large intestine. They are metabolized in the body and play various roles. In relation to nutritional and physiological value of the minerals, fermented milks are particularly significant as a source of Ca, P. Calcium and P which are the most essential minerals for bones

(Fernandez et al., 1992). Yogurt is excellent source of dietary minerals, particularly Ca, P and Mg (Gurr, 1987; IDF, 1991 b). The bioavailability of Ca from yogurt and other fermented milks is high and it is readily absorbed. Calcium absorption is facilitated in the presence of some food constituents and lactose is one of them (Gurr 1987).

Minerals have the following functions in the body:-

- i) They are components of hard tissues (Ca, P and Mg contribute as major constituents to the formation of bones and teeth).
- ii) They are components of soft tissues (Fe,P, K, Cl and I contribute to the formation of muscle, skin and blood).
- iii) They are regulators of biological functions (contributing, as salt dissolved in body fluids, in sensitivity to nervous stimuli, regulating the pH of digestive fluids and other secretions).

2.6.5.5 Therapeutic properties of yogurt

Nutritional and physiological value of the individual components of fermented milks have been discussed in previous sections. It remains to discuss the therapeutic properties of yogurt.

Scientific explanation of positive effect of fermented milk products was first provided during the first decade of this century by Metchnikoff (1908). He claimed that yogurt bacteria inhibit the growth of anaerobic sporeformers in the large intestine thus preventing putrefactive processes from taking place in alimentary tract and thereby prolonging life. He believed that senility and death are accelerated by poisoning from a putrefactive intestinal microflora. To reduce the effect of the putrefactive process, he firmly believed in the consumption of a large quantity of products capable of replacing the putrefactive microflora by a lactic microflora.

Although today this theory seems exaggerated, the antimicrobial, antitumour, antimutagenic properties of fermented milk products have been well documented by several author (IDF, 1991 b; Gilliland, 1991; Yukuchi et al, 1991; Fernandez, 1992).

The beneficial effect of fermented milk products in human nutrition is dependent upon the content of metabolic products resulting from biochemical activities of lactic acid bacteria during the fermentation period. Lactic acid, the main chemical compound, shows bacteriostatic and in some cases bactericidal effects against putrefactive microorganisms which are sensitive to its action. Particularly sensitive are spore-forming bacilli and coliform. Other organic acids (acetic, propionic, formic), although produced in small quantities, may even exert much stronger antimicrobial effects than lactic acid. Antimicrobial effects of fermented milks can be attributed to organic acids, antibiotic factors, volatile acids, hydrogen peroxide and to other still unidentified substances present in fermented products. Fermented milks provide, also, living cells able to proliferate in human intestine and discourage the growth of harmful organisms. The presence of a large number of living microorganism having a special metabolic function, when fermented milk is consumed regularly, may contribute to the development and maintenance of non-specific immunological reactions in human body.

3.0 MATERIAL AND METHODS

3.1 Material

3.1.1 Raw materials

3.1.1.1 Raw cow's and ewe's milks

In this study, two types of antibiotic-free whole milk namely cow's and ewe's milk were used. Cow's milk was obtained from Outwoods Farm (Mr. Bailey, Outwoods Farm, Forest Road., Loughborough) and ewe's milk was obtained from Wayfield Farm, (Mr. Titcomb., Wayfield Farm, Gospel Lane, Snitterfield. Stratford). It was cooled to about 5°C and then transferred from farm to laboratory. The milk was delivered in 10 L teflon containers which were cleaned before using.

3.1.1.2 Starter cultures

Commercial mixed species yogurt starter cultures (Code CH-1) were obtained from CHR. Hansen's Laboratory Ltd. (Reading, Berks. UK). During the work the freeze dried cultures were maintained in a deep-freezer.

3.1.1.3 Filter medium

The cotton cloth filter medium was obtained from Sümerbank Textile Industry AS, Adana, Turkey. It is a 2/2 twill. The synthetic cloth filter medium (J/SYN/32 Terylene) was obtained from Johnson Progress Ltd Stratford London. It is a multi filament and plain weave. These two type of weave are illustrated in Fig (3.1).

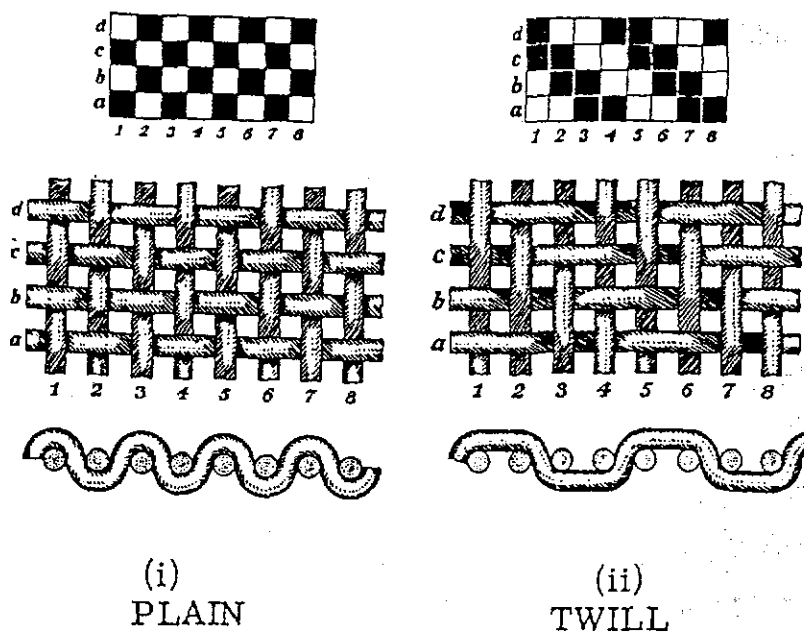


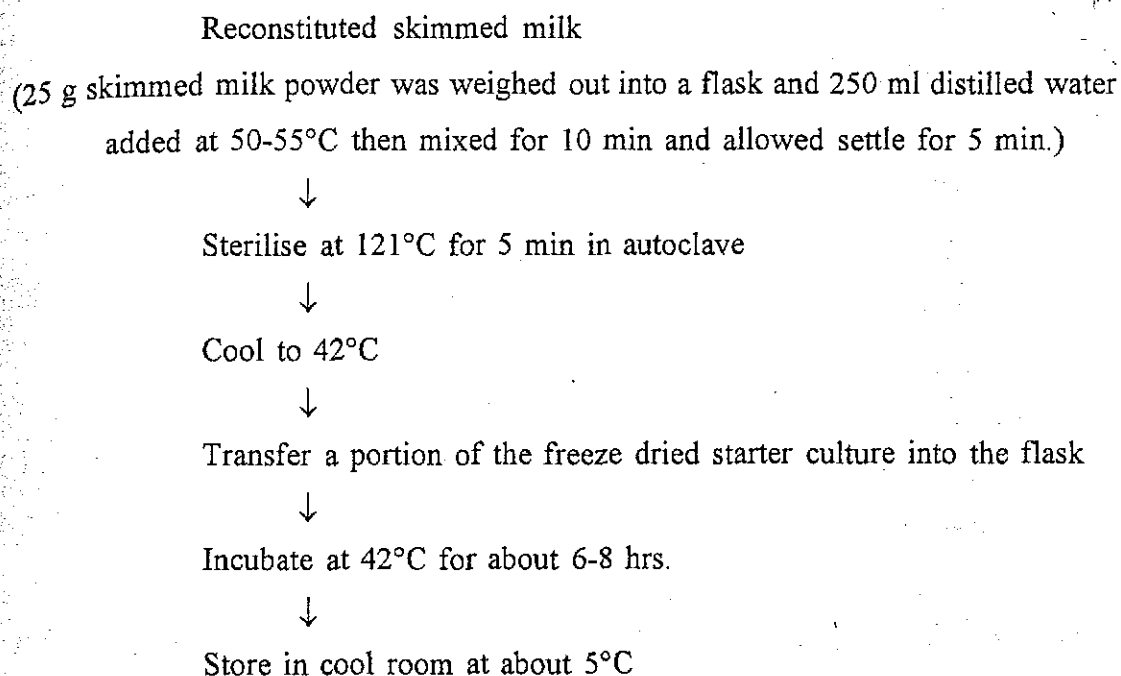
Fig (3.1) The two type of weaves.

3.2 Methods

3.2.1 Starter cultures propagation

A typical flow diagram for the propagation of starter culture is given in Fig (3.2). 250 ml of reconstituted skimmed milk was made from antibiotic-free skimmed milk powder (10 % solids nonfat). Skimmed milk powder was obtained from Oxoid Ltd. (Basingstoke, Hants, UK). Mother cultures were propagated using the supplier's directions. It was then sub-cultured every 5 days.

Fig (3.2) The flow diagram of starter cultures propagation



3.2.2 Concentration method of raw milk

Raw milk samples were concentrated by using an ultrafiltration (UF) unit (DDS 35, 2.25 UF Lab unit supplied by DDS RO-Division DIL-4900 Nakskov, Denmark) with membrane type GR 61PP (Polysulfon membrane with an approximately cut-off value of molecular weight of 20000) until the desired total solids level was reached. The filtration temperature was $40 \pm 2^\circ\text{C}$, the inlet pressure was 5 bar, the outlet pressure was 4 bar and the flow rate was $5 \text{ m}^3/\text{h}$. The filtration time was 20 min. for sheep's milk and 35 min. for cow's milk. The concentration level was 4:1 for cow's milk and 2.8:1 for ewe's milk

3.2.3 Heat treatment

After standardisation of the concentrated whole milk, it was heated to 80°C and held for 15 min in a waterbath (Techne, Tempette Junior TE-8J supplied by Fison

Scientific Equipment plc , Loughborough. UK.). The waterbath had a water circulation system. During heat treatment the milk was agitated by a stirrer (Supplied by Voss Instruments Ltd. Essex). After heat treatment, the milk was quenched in ice water to the temperature of 45°C.

3.2.4 Production methods of high total solids Turkish style thick yogurt

In this present study two different methods were used. Concentration the milk using UF and concentration yogurt using modified traditional method.

3.2.4.1 Production method of stirred style Turkish thick yogurt by using modified traditional method

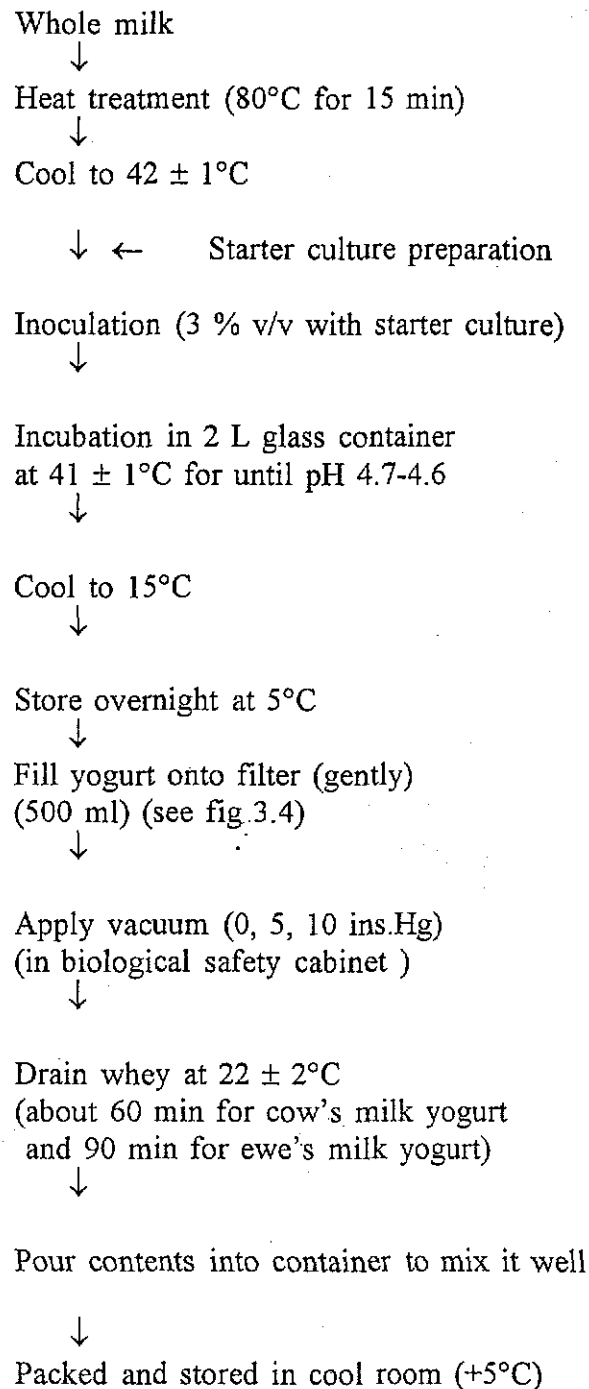
The flow diagram of filtration processes of yogurt is given in Fig (3.3) and a picture of modified traditional filtration apparatus is shown in Fig (3.4). Three different vacuum levels (0, 5 and 10 ins.Hg) and two different filter media, namely cotton and synthetic cloth were used. The specification of the cotton and synthetic cloth filter media are given in section 3.1.1.3. The whole milk was heated to 80°C and held at this temperature for 15 min in a water bath (Thermostatically controlled electrical water bath supplied by Fison Scientific Equipment plc). It was than cooled to 42°C and inoculated with yogurt starter culture (see section 3.1.1.2). The inoculate was incubated in 2 L glass jar (supplied by Fison Scientific Equipment plc) at $41 \pm 1^\circ\text{C}$ until the acidity reached pH 4.7 - 4.6. After incubation the product was stored overnight in refrigerator at 5°C. On the following day 500 ml samples of yogurt was concentrated by using the modified traditional filter (see Fig 3.4) at room temperature using the different vacuum levels. A Buchner funnel was used for the filtration experiments. It was plugged into the top of a measurement cylinder which had been fitted with a side arm through which a vacuum was applied.

The vacuum pump (Speedi Vac^R High Vacuum Pump, Model ISP 30 C) was supplied by Edwards High Vacuum Ltd Sussex UK. The filter cloth, trapped between two concentric rings which sealed on the inside of the funnel, was supported about 5 mm above the inside of the funnel by wire mesh so that all the cloth was draining liquid. The diameter of the cloth, over which filtering was effective, was 115 mm. Filtration time was 60 min for cow's milk yogurt and 90 min for ewe's milk yogurt. Filtration was carried out in a biological safety cabinet supplied by MDH Ltd, (Andover Hampshire UK.) to prevent unexpected contamination.

3.2.4.2 Production method of set style Turkish thick yogurt by using fortified UF milk retentate

The flow diagram of set style thick yogurt using UF retentate is given in Fig (3.5). A picture of the apparatus can be seen in Fig (3.6). The whole raw milk (cow and ewe) was prewarmed to 40 ± 2 °C before concentration of milk by ultrafiltration unit to about 34 % total solids (TS) as noted in section 3.3.2. The retentate was standardised to 24, 26, 28, 30 and 32 % total solids level with fresh permeate, heated to 80°C and maintained for 15 min in a water bath (thermostatically controlled electrical water bath supplied by Fison Scientific Equipment plc). then it was cooled to 42°C and inoculated with yogurt starter culture (see section 3.1.1.2). The inoculate was incubated in 100-250 ml glass jar (supplied by Fison Scientific Equipment plc) at 41 ± 1 °C until the acidity reached pH 4.7-4.6. After incubation the product was stored overnight in refrigerator at 5°C.

Fig (3.3) The flow diagram of concentration of yogurt by using modified traditional method (vacuum batch filtration)



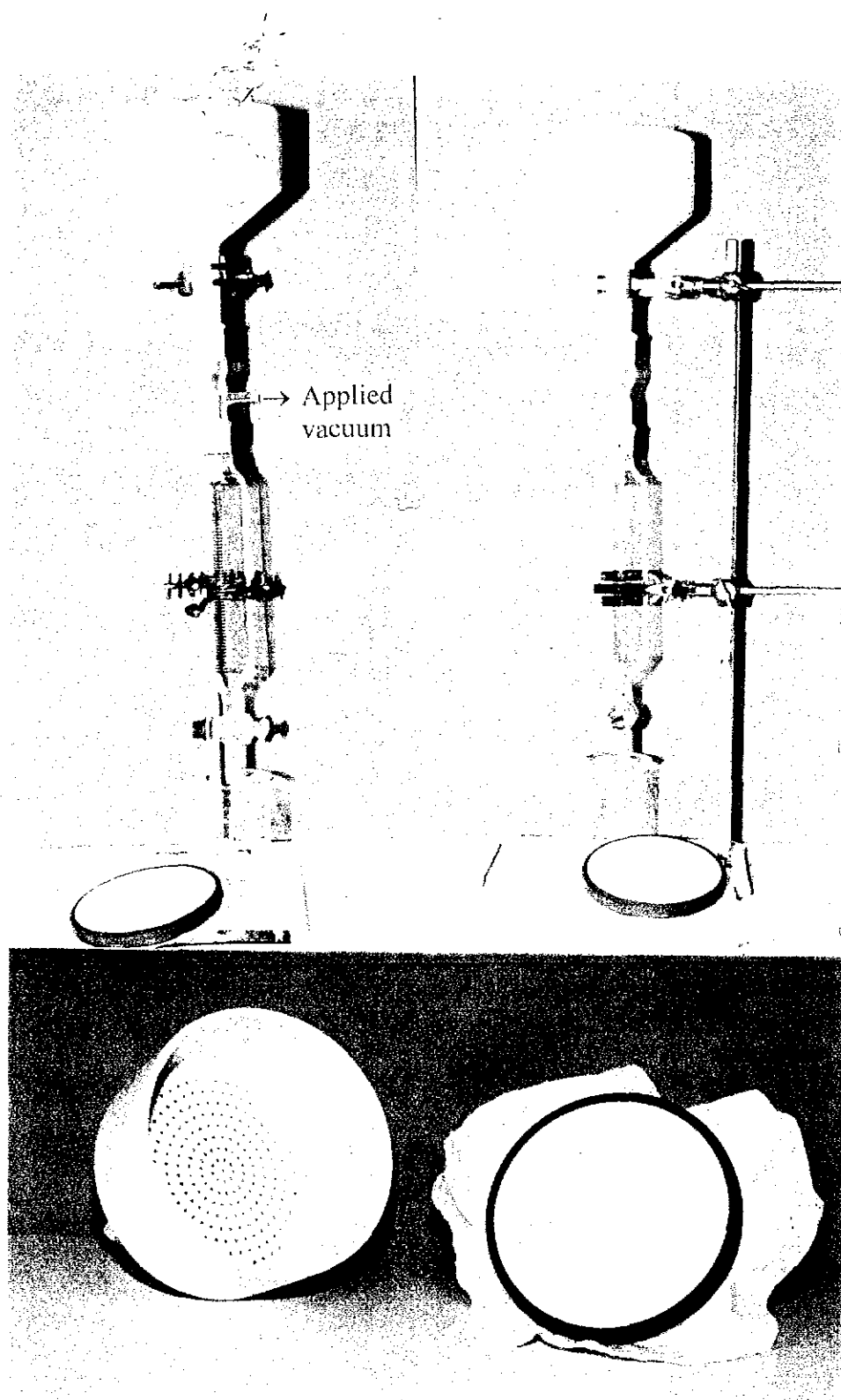


Fig (3.4) Modified traditional filtration apparatus

Fig (3.5) A generalized scheme for production method of set style turkish thick yogurt produced from UF retentate.

Prewarmed whole milk to $40 \pm 2^{\circ}\text{C}$



Concentrate milk to about 34 % total solids level
using an DDS pilot unit (see fig 3.6)



Standardised retentate to 24 to 32 % total solids



Heat to $80 \pm 2^{\circ}\text{C}$ for 15 min



Cool to 45°C



Inoculate with 3 % starter cultures



Incubate at $41 \pm 1^{\circ}\text{C}$
(in 100 and 250 ml glass jar)
until pH reaches 4.7-4.6



Store overnight at 5°C for analysis

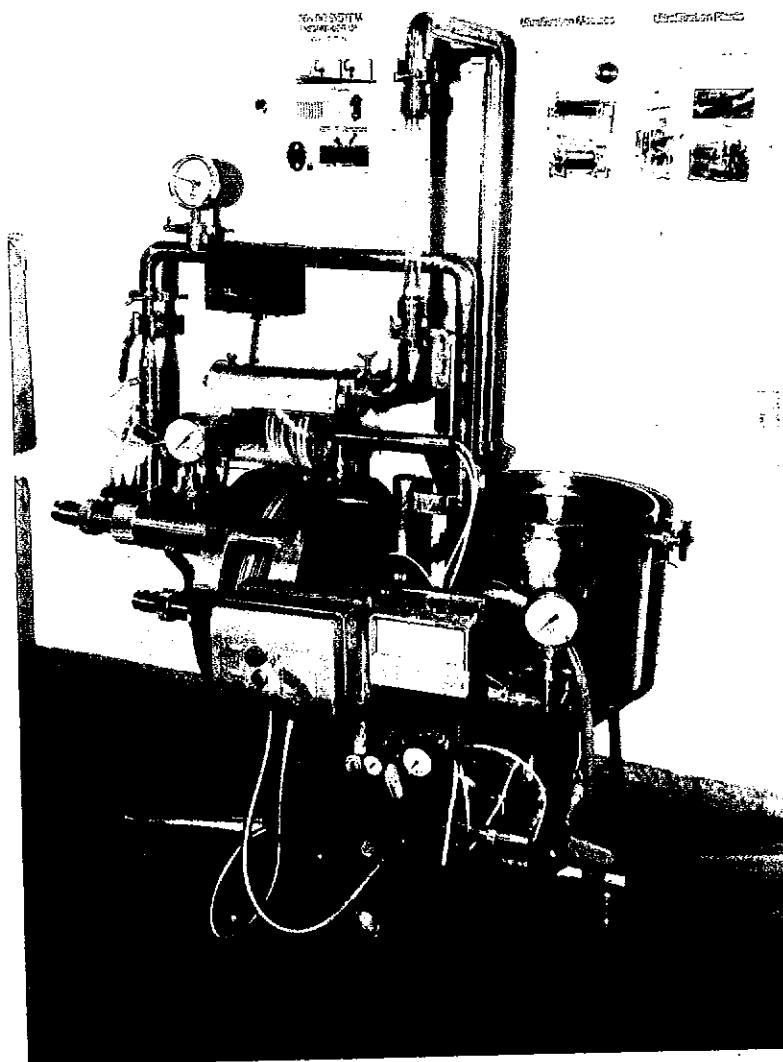


Fig (3.6) Ultrafiltration unit used in experimentation.

3.2.5 Analysis methods

3.2.5.1 Analysis methods of milk

3.2.5.1.1 Determination of pH

The pH value of milk was measured using a pH meter (EIL 7030 model Analytical Supplies Ltd Derby. UK) which was fitted with a standard combined glass electrode. The equipment was adjusted with buffer solutions of pH 7 and 4 (Fison Scientific Equipment plc, Loughborough) taking into consideration the temperature of the buffer solutions and the sample before starting the measurement. The samples were mixed thoroughly and the pH electrode were inserted to a depth of about 3-4 cm. The reading was recorded and pH electrode removed from sample and cleaned with distilled water. One pH reading was carried out with each sample.

3.2.5.1.2 Determination of titratable acidity

Titrateable acidity in milk was determined according to the method described in BS 1741: Part 10:Section 10.1, (1989). 10 ml or 10 g of sample was weighed out and 1 ml of 0.5 % (w/v) solution of phenolphthalein [0.5 g of phenolphthalein was dissolved in 55 ml of 95% (v/v) ethyl alcohol, and then made up to 100 ml with distilled water.] was added as an indicator. The sample with indicator was titrated with 0.1 N NaOH solution until the end point pinkish colour was reached. The volume of NaOH solution used for titration was divided by 10 and the value expressed the acidity as per cent of lactic acid.

3.2.5.1.3 Total solids

The total solids was determined according to method described by BS 1741:Part:2, (1963). About 5 g. of mixed milk sample was weighted out into a pre-weighed

round flat-bottomed metal dish. It was placed on a boiling waterbath (supplied by Fison Scientific Equipment plc) for about 45 min. Then, the bottom of the dish was wiped and the sample transferred into an oven at 105 °C (supplied by Fison Scientific Equipment plc. Loughborough) for 4 hrs until constant weight was reached.

3.2.5.1.4 Protein

The Kjeldhal procedure:

The method was adapted from Kirk and Sawyer (1991)

A) Reagent and apparatus:

i) Reagent:

- a) Sulphuric acid. (sp gr. 1.84 nitrogen free)
- b) Hydrochloric acid, 0.1N standardised.
- c) Boric acid solution (2%)
- d) Sodium hydroxide solution 50% (w/v).
- e) Hydrogen peroxide
- f) Kjeldahl catalyst tablet-selenium. (Each tablet contains 1 g Sodium sulphate and the equivalent of 0.05 g selenium supplied by Fison Scientific Equipment plc. Loughborough. UK.)
- g) Screened methyl red indicator solution [10 ml/L bromcoresol green (100 mg in 100 ml ethyl alcohol) / 7 ml/L methyl red (100 mg in 100 ml ethyl alcohol)]

ii) Apparatus :

- a) Kjeldahl digestion tube (supplied by Fison Scientific Equipment plc. Loughborough. UK.).
- b) Digestion unit: Tecator Digestion system 12 1009 digester (supplied by Tecator ltd. Bristol.)
- c) Distillation unit: Tecator Kjeltec system 1002 (supplied by Tecator ltd. Bristol.)

B) Digestion.

- i) Approx. 0.5-2 g. well mixed sample was weighted out into filter paper and transferred to a digestion tube.
- ii) Kjeltab tablets were added into digestion tube.
- iii) 5 ml concentrated sulphuric acid with 5 ml hydrogen peroxide was added into digestion tube and mixed well.
- iv) The digestion tubes were placed in the Kjeldhal digestion unit (see above).
- v) They were attached to the venting tubes which remove the acid vapours. Then the tubes were heated slowly. Digestion tubes were held in digestion unit until the liquid has become completely clear. The total digestion time was approximately 60 min.
- vi) The digestion tubes were removed from the digestion unit and allowed to cool down for 10 min on the metal rack.

C) Steam distillation:

A volume of 25 ml of boric acid solution (2%) with screened methyl red indicator was transferred in to a titration receiver flask. It was placed on the distillation unit. The tube containing the digested sample was attached to the distillation unit and the start button pressed to effect a metered addition of sodium hydroxide solution [50 % (w/v)]. The distillation unit was turned on and 150 ml distillate collected into a receiving flask. The distillation was stopped and the flask was removed letting the tip of the condenser drain into the flask momentarily. The tip was rinsed with distilled water catching rinsing in the flask and the distillate was titrated with 0.1 N HCl solution until neutral grey colour. *The 96 ml sample was collected.*

3.2.5.1.5 Lactose

The method is described by Kirk and Sawyer, (1991).

A) Apparatus and operating conditions:

Apparatus : HPLC
Column : Zorbax NH₂ (supplied by Jones Chromatography Ltd UK).
Detector : Waters 410 differential refractometer (supplied by Millipore).
Pump : HPLC Pulseless pump.
Recorder : Plotter 800 (Supplied by Kontron Instruments UK).
Flow rate : 1 ml/min.
Injection : 20 μ l

B) Reagents

Mobile phase: acetonitrile/water (80 / 20) (It was filtered through 0.2 μ m membrane filter and degassed with helium before using).

C) Procedure :

- i) Approximately 5 g sample was weighed into 100 ml volumetric flask to give final lactose extract solution strength 0.1 g (w/v), where possible.
- ii) 40 ml water was added into volumetric flask. Then it was shaken with a mechanical shaker (supplied by Infors AG CH-4103 Bottingen) for about 45 min. 40 ml acetonitrile was added into the same flask and shaken for a further 10 min. It was marked as acetonitrile/water (50/50) solution.
- iii) The solution which contained lactose from sample was filtered through Watman-42^R or equivalent.
- iv) Standard solution using pure lactose was prepared with similar procedure.
- v) Standard and samples were injected into HPLC column. The peak area of lactose for each samples was recorded. The concentration was calculated from the peak area corresponding to the amount of lactose present in sample. Results were expressed in terms of a percentage (w/v) in the original milk samples.

3.2.5.1.6 Fat

The fat content of milk was determined according to procedure described by BS 696, Part:1, 2 (1988) for the test together with the test apparatus. A volume 10 ml of sulphuric acid (density 1.82 at 20°C), 10.94 ml of well mixed milk and 1 ml of fat free amyl-alcohol was poured into a butyrometer with a pipette, ensuring that they did not mix with one another. The butyrometer was closed with a stopper, the contents were thoroughly mixed and immediately centrifuged at 1100 rpm for 5 min. At the end of the time the butyrometer was transferred into a waterbath at 65°C for 5 min. and the percentage fat read off directly from the scale.

3.2.5.1.7 Ash

Ash content of milk was determined according to the procedure described by Case et al (1985). A porcelain dish was heated in a muffle furnace to dull redness and cooled down to room temperature in a desiccator until ready to use. 10 g of sample was weighed out into the pre-weighted porcelain dish. Then it was transferred into an oven (Gallenkamp, supplied by Fison Scientific Equipment plc. Loughborough) at 100°C for one hour. Then transfer to a ventlator hood and placed on an iron tripod over a bunsen burner and slowly carbonized. Finally, it was placed in a muffle furnace (Gallenkamp, supplied by Fison Scientific Equipment plc. Loughborough) regulated at 550°C until free from carbon and a light grey ash remained and then cooled in desiccator. It was weighed to calculate amount of ash.

3.2.5.1.8 Buffer capacity

The buffer capacity of milk and ultrafiltered cow's and sheep's milk (24-32% total solids) were determined by a modification of the method of Sutherland and Jameson, (1981). A volume of 15 ml milk sample, containing 0.4 g/L sodium azide (Supplied by Sigma Chemical Co.) was added to each of 9 screw-capped vials. 0 - 1.6 ml of

1.20M lactic acid (Supplied by Fison Scientific Equipment plc) was added in increments of 0.2 ml. Each vial was brought to a volume of 16.6 ml with distilled water. The mixtures were mixed by a shaker (supplied by Infors AG CH-4103 Bottingen) at room temperature (ca 22°C) for 16 h and the pH was determined (see section 3.3.5.1.1). The data were plotted as pH versus lactic acid added per gram of milk medium and the buffer capacity was calculated from the slope of the curve at a pH 5.3 and expressed as the amount of lactic acid (μmoles) required to decrease the pH of 1 g of milk medium by one unit.

3.2.5.1.9 Mineral analysis

A) Apparatus and operation conditions:

Apparatus : Dionex series 4500i.
Column : Dionex Ion-PacTM A/S 4A P/N 37041 for Anions.
Dionex Ion-PacTM CS12 (10-32) P/N 41001 for Cations.
Detector : Dionex Conductivity detector.
Pump : Dionex gradient pump.
Computer : Deel 210.
Printer : Epson FX 850.
Software : AI 450TM
Flow rate : 1.8 ml/min for Anions.
1 ml/min for Cations.
Injection : 50 μl

B) Reagents :

Eluent : 16 mM HCl for Cation
1.7 mM NaHCO₃ for Anion
1.8 mM NaCO₃
Regenerant : 25 mM H₂SO₄ for Cation
15 mM KOH for Anion

C) Procedure :

- i) Approximately 0.05 g dry ash which was obtained from dry ashing method (see section 3.2.5.1.7) was weighed out into 100 ml flask.
- ii) 10 ml diluted HCl (1:1) was added.
- iii) The ash was digested on a boiling waterbath for about 60 min. The flask was covered with watch glass during digestion period.
- iv) The content of flask was transferred to a 250 ml volumetric flask, stirred and then cooled under the tap.
- v) The content of the volumetric flask was diluted nearly to required volume and stirred again. The volume was then made up to 250 ml.
- vi) The instrument was set according to the instructions.
- vii) The reference (deionized water) solution and standard solutions were injected into the column before a set of sample analysis to validate the calibration. Then samples were injected to measure the quantity of minerals present.
- viii) Result were processed with a computer and presented as concentration (mg/L) corresponding to the absorption or emission values of the samples, reference and standard solutions.
- ix) These detected concentration of minerals(Na, K, Mg, Ca, P and S) were converted to the amount of minerals in the original milk sample.

3.2.5.1.10 Microbiological analysis

3.2.5.1.10.1 Total count of initial bacteria in raw milk

The total count of initial bacteria in raw milk was determined according to procedure described by Messer et al (1985) for the examination of dairy products. The growth medium used was plate count agar (Oxoid Ltd., Basingstoke, Hants, UK) and the plates were incubated at $32 \pm 1^\circ\text{C}$ for 48h in " LEEC " incubator (supplied by Laboratory and Electrical Engineering Co. Nottingham). The count was reported as *cfu/g*.

3.2.5.1.10.2 Coliform bacteria count

Coliform test was carried out according to the procedure described by Hartman and La Grange (1985). The growth medium used was Violet Red Bile Agar (Oxoid Ltd., Basingstoke, Hants, UK.). The plates were incubated at $32 \pm 1^\circ\text{C}$ for 24h in "LEEC" incubator (supplied by Laboratory and Electrical Engineering Co. Nottingham). It was reported as coliform bacteria *cfu/g*.

3.2.5.1.10.3 Yeast and Moulds count

Yeast and moulds count were determined according to Frank et al (1985). The growth medium used was Potato Dextrose Agar. The plates were incubated at 25°C for 5 days in incubator. It was reported as the number of yeast and moulds cells per gram of sample.

3.2.5.2 Analysis methods of yogurt

3.2.5.2.1 Determination of pH

The pH in yogurt was determined according to the methods mentioned in section 3.2.5.1.1.

3.2.5.2.2 Determination of titratable acidity

Titratable acidity of yogurt and concentrated yogurts were determined by titration. 5 g of sample was weighed into 100 ml flask and 5 g of water was added and mixed well. This was titrated according method described in section 3.3.5.1.2 and the result was adjusted by a dilution factor.

3.2.5.2.3 Determination of total solids

Total solids in yogurt was determined according to the methods described in section 3.2.5.1.3.

3.2.5.2.4 Determination of protein

Protein in yogurt was determined according to the methods described in section 3.2.5.1.4.

3.2.5.2.5 Determination of lactose

Lactose in yogurt was calculated by using the equation:

$$\text{Lactose} = \text{Total solids} - [\text{protein} + \text{fat} + \text{ash}]$$

3.2.5.2.6 Determination of fat

10 g of concentrated yogurt was mixed with 10 g of warm distilled water (40°C) and the fat content was determined according to Gerber method described in section 3.2.5.1.6 and the final reading was multiplied by the rate of dilution factor.

3.2.5.2.7 Determination of ash

Ash in yogurt was determined according to the methods described in section 3.2.5.1.7.

3.2.2.8 Determination of firmness

A penetrometer was used in this particular study. It was the universal model manufactured by Stanhope-Seta Ltd (Stanhope-Seta Ltd Chertsey, Surrey UK.). The shape and size of cone and plunger had to be devised in relation to both the nature of the product and the size of the sample. The cone and the plunger were made in

the department workshop. The shape of cones varied from completely flat to ones with sides angled at 30° (see Fig 3.7). However, the diameter of cone used was always approximately half the container diameter at the 200-250 ml level; in this way, any distortion of reading by wall pressure would be minimised.

30° cone and light plunger were used because it was found that the standard plunger and cones made from stainless steel were too heavy for standard control yogurt. For this reason, an exact copy of stainless steel plunger and cones was made from aluminium. The firmness of yogurt gels were measured by the penetrometer. The cone was positioned on the surface of the yogurt gels and then released for a fixed time (5 sec). At the end of the time the probe was clamped and the penetration depth was measured using a dial gauge. Three reading were taken at different positions of the surface of the sample to minimize deviations. Measurement was made at $7 \pm 2^\circ\text{C}$. The weight of probe and cone was 61.12 g. The result was expressed as penetration (1/10 mm).

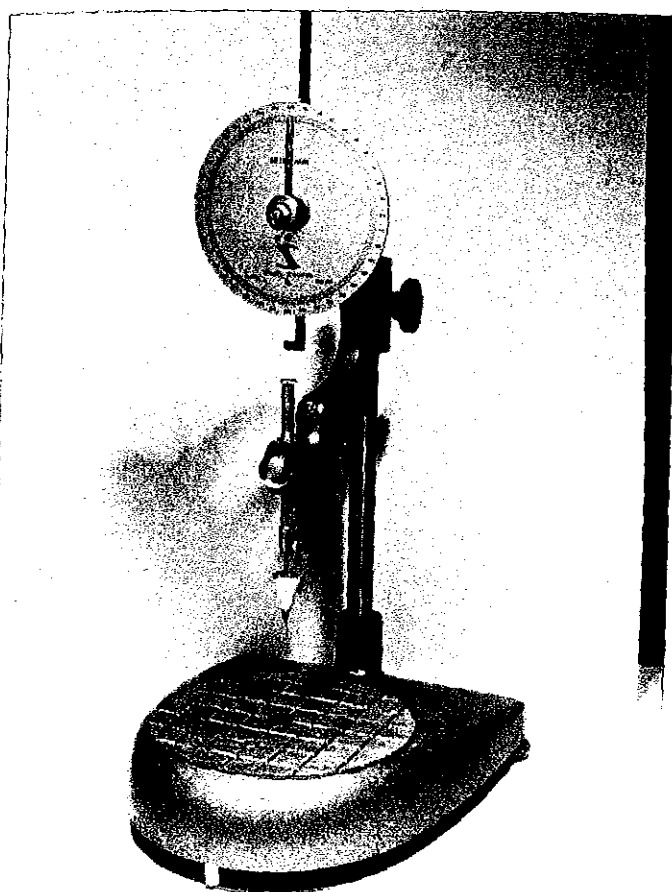


Fig (3.7) Penetrometer apparatus

3.2.5.2.9 Water Holding Capacity

Water holding capacity (WHC) of yogurt gels were determined by the centrifugation procedure suggested by Kaláb et al (1976). Samples were centrifuged at 2500 g for 30 min at 20°C in a centrifuge (MSE Hi-Spin 21, supplied by Fison scientific equipment plc.). They were drained of supernatant for 10 min, and centrifuged was weighed out. The WHC was expressed as percent centrifuged weight relative to the original weight of yogurt. (Parnell-Clunies et al, 1986).

3.2.5.2.10 Volatile aroma compounds

A) Apparatus and operation conditions:

Gas Chromatography	: PYE Unicam Series 304.
Column	:BP1 SGE capillary column (25M x 0.32mm id.).
Detector	:Flame ionisation detector.
Integrator	:PYE unicam PU4820 Computing Integrator.
Syringe	:Microliter syringe 701N. Hamilton Bonaduz AG. PO Box 26-7402. Bonaduz. Switzerland.
Column temperature	:65°C
Injector temperature	:150°C
Detector temperature	:200°C
Carrier gas (Helium)	:8 ml/min
Air	:10 psi
Hydrogen gas	:15 psi
Chart speed	:1 cm/min

B) Steam distillation apparatus:

Quiqfit steam apparatus was set up as shown in Fig (3.8). The apparatus consisted of several major units, a two neck (1000 ml) and three neck (500 ml) round bottom flask, a condenser, a heater and an ice water bath. The steam for the distillation was generated by the steam source. The steam source comprised the two neck flask containing the water, placed in the heater. One neck was connected to the three neck flask and the other provided the pressure release. The volatile components of the sample placed in the three neck flask were evaporated by the steam. The three neck flask was connected to the condenser by a splash head modified with a 19/24 male joint. The condensate was collected in a sample bottle placed in an ice water bath. All joins were sealed with silicon grease.

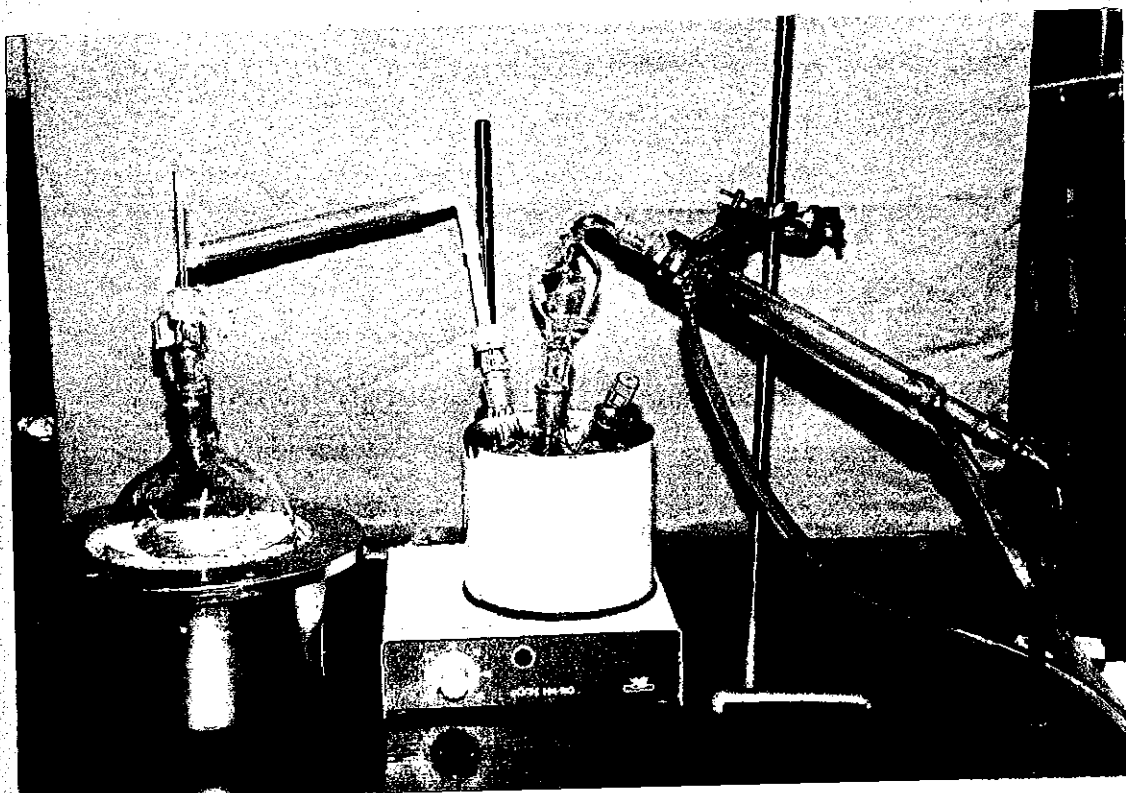


Fig (3.8) Steam distillation apparatus.

C) Reagents and Standards:

i) Chemicals

Acetaldehyde, acetone and diacetyl (supplied by Aldrich Chemical Co. Ltd UK.).

ii) Standard solution and sample preparation:

Standard solution

Aqueous stock solutions, containing 1 g / ml of the test compounds were prepared. Acetaldehyde was chilled in an ice bath and approximately 1 g was weighed into 100 ml volumetric flask containing about 80 ml chilled water. This solution was further diluted with chilled water to give a 8, 12, 14, 16, 20, 24 ppm solution. The stock solutions were stable for at least 2 months when stored at 5°C in glass bottles with sealed caps (Ulberth, 1991). All others standard solutions were prepared in the same way.

D) Sample preparation

A 50 g sample was weighed into round bottom two necks flask then 25 g of chilled water was added and mixed well. It was fitted with steam line and condenser (see Fig 3.8). Distillate was collected in ice-water bath to keep volatile compounds in distillate. About 20 g distillate was collected as a sample to inject.

3.2.5.2.11 Free amino acids

A) Chemicals, Reagents and Buffers:

i) Chemicals :

Chemicals required for reagent and buffers manufacture are listed below;

Ninhydrin	Merck 6762
Hydrindantin (dihydrate)	Merck 4510
Dimethyl sulfoxide	Merck 802912
NaOH	Merck 6498
Na-Citrate	Merck 6448
NaCl	Merck 6404
Citric acid (monohydrate)	Merck 244
Methanol	Merck 6008
HCl	Merck 318
Phenol	Riedel De Haen 16018
Brij-35	Merck 10522
Lithium hydroxide(monohydrate)	Merck 11652
Lithium chloride	Merck 5679

ii) Buffers:

a) Preparation of lithium acetate buffer:

42 g of lithium hydroxide (monohydrate) was added to approximately 600 ml of distilled water while stirring vigorously and 293 ml of glacial acetic acid was carefully added to this solution. It was allowed to cool and topped up to 1000 ml. The pH was adjusted to 5.2 as required. The solution was filtered before use.

b) Buffer -I:

NaOH	6.40 g
Citric acid (monohydrate)	14.70 g
Methanol	80.00 ml
Phenol	1.00 ml

Total volume is 1000 ml and the pH was adjusted to 3.2 with HCl.

c) Buffer-II :

Na-Citrate	19.60 g
NaCl	29.20 g
Phenol	1.00 ml

Total volume is 1000 ml and the pH was adjusted to 3.79 with HCl.

d) Buffer-III:

Na-Citrate	19.60 g
NaCl	81.80 g
Phenol	1.00 ml

Total volume is 1000 ml and the pH was adjusted to 4.3 with HCl.

iii) Preparation of ninhydrin reagent:

Ninhydrin reagent for use with buffer systems described in buffer section can be made up as follows. (The reagent must be manufactured and stored under nitrogen). 650 ml of dimethyl sulfoxide was measured into the reagent bottle and purged with cleaned nitrogen for 10 min.

20 g of ninhydrin was added with 1.3 g of hydrindantin (dihydrate) and allowed to dissolve (the reagent may be stirred or the nitrogen purge used to aid dissolution). Once completely dissolved, stirring was continued for 10 min prior to adding 350 ml of 4 N lithium acetate buffer pH 5.2.

B) Sample preparation

Sample preparation of milk for free amino acids described by Williams et al (1980) was modified for use. Approximately one gram of milk sample was weighed into 10 ml jar. The amount of 5 g of 5 % (w/v) 5-sulphosalisilic acid solution (Aldrich Chemical Co. Ltd) was added. Each sample was thoroughly mixed, stored overnight at 4°C and then centrifuged (MSE micro centaur. Supplied by Fison Scientific Equipment plc.) for 5 min at 13000 rpm. The supernatant was removed and then stored at -20 °C to await amino acid analysis. Equal volume of 0.2 N sodium citrate was added (pH 2.2) to each sample before injection into column.

C) Apparatus: Chromacon 500 amino acids analyzer

(Supplied by Kontron Electrolab UK)

Column	: Ion exchange column which contains AS70™ resin.
Pump system	: Two reciprocated piston pumps with gravity assisted double check valves.
Operating pressure	: 100 bar
Flow rates	: 24 ml/h (for eluent) 12 ml/h (for ninhydrin reagent)
Detector	: Absorption detector
Column temperature	: 50 to 80°C in 0.5°C increment
Sampler	: Injection volume is 100 µL Wash and Dry time is 8 min

Identification and computation: The peak areas on the chromatogram were measured by computing integrator then compared with the peak area of standard amino acids mixture

3.2.5.2.12 Mineral analysis

Mineral content of yogurt was determined according to the methods described in section 3.2.5.1.9.

3.2.5.2.13 Microbiological analysis

3.2.5.2.13.1 Total count

Total count of yogurt was determined according to the methods described in section 3.2.5.1.10.1.

3.2.5.2.13.2 Starter culture count

L-S differential medium as recommended by Eloy and Lacrosse (1976) was used for the enumeration of the yogurt starter culture. Starter culture viable cell count was determined according to procedure described in Oxoid Ltd manual (1982). The growth media used was Lactic- Streptococ Differential Agar (supplied by Oxoid Ltd Hunt). The differentiation between the Lactobacilli and Streptococci is based on colonial morphology which is summarised as follows:

L. bulgaricus: Irregular red, rhizoidal colonies, 1 - 1.5 mm diameter, surrounded by a white opaque zone.

S. thermophilus: Round or oval red colonies 0.2 - 0.5 mm diameter, surrounded by a small clear zone.

3.2.5.2.13.3 Coliform bacteria count

Coliform bacteria count of yogurt was determined according to the methods described in section 3.2.5.1.10.2.

3.2.5.2.13.4 Yeast and Moulds

Yeast and moulds count of yogurt was determined according to the methods described in section 3.2.5.2.10.3.

3.2.5.3 Chemical analysis of the UF permeate and whey from modified traditional filtration.

UF permeate and whey samples were analyzed for pH, titratable acidity, total solids, protein, fat and ash according to methods mentioned in sections 3.2.5.1, 3.2.5.2, 3.2.5.3, 3.2.5.4, 3.2.5.6 and 3.2.5.7.

3.2.5.4 Statistical analysis.

The experiment results were statistically analyzed using analysis of variance and Paired-t Test with the Origin computer package (MicroCal Software, Inc.).

4.0 RESULT AND DISCUSSION.

Different total solids level for UF concentrated milk retentate, modified traditional concentrated yogurt, yogurt whey and permeate from UF are coded with letters in the text as follows in Table (4.1)

Table (4.1) Coding system key for products.

Code	Explanation
A	Whole milk yogurt (control)
B	Approximately 24% total solids yogurt from UF retentate
C	Approximately 26% total solids yogurt from UF retentate
D	Approximately 28% total solids yogurt from UF retentate
E	Approximately 30% total solids yogurt from UF retentate
F	Approximately 32% total solids yogurt from UF retentate
G	Stirred yogurt of 25% total solids made by modified traditional method

If these letters are coded with a "C" at the beginning it means cow's milk (for example CA, CB, CC, CD, CE, CF, CG) "CA" being whole cow's milk yogurt. Similarly, when "E" appears at the beginning this indicates it is ewe's milk (for example EA, EB, EC, ED, EE, EF, EG) "EA" being whole milk yogurt made from ewe's milk. This coding system will be used throughout the remaining part of the thesis

4.1 Raw materials

4.1.1 Chemical composition of milks

Whole cow's and ewe's milk were used for the manufacture of high total solids set and stirred style yogurt, produced using UF and modified traditional filtration methods. Experimentation was carried out at different months of the year to coincide

with the lactation period of these animals. Cow's milk tests were conducted in October -January in 1992-93 and ewe's milk tests were carried out in February- July 1993.

The mean chemical composition of milks used in this study are given in Table (4.2) and raw data are presented in Appendix (I - 1) for cow's milk and Appendix (I - 2) for ewe's milk. There was a slight variation in the composition of cow's milk, especially total solids, during the testing. This variation can be attributed to various factors such as heredity, stage of lactation, season of the year, nutrition of animal, age of animal and environment (Juarez and Ramos, 1984). Variation in the composition of ewe's milk was also evident in the total solids especially fat, protein and lactose contents. These variation are similarly attributed to the factors mentioned above.

Table (4.2) Chemical composition of milk used for this study (% w/w).

Components	Types of milk	
	Cow	Ewe
Total solids	12.18	17.12
Protein	3.42	5.41
Fat	4.02	6.1
Lactose	4.63	5.04
Ash	0.71	0.91
pH	6.7	6.67
Lactic acid %	0.19	0.2

4.1.2 The microbiological quality of raw milk

The microbiological quality of cow's and ewe's milk is illustrated in Table (4.3). The number of bacteria in raw milk must meet $\leq 10^5$ cfu/ml plate count at 30 °C set in the Official Journal of the European Communities (No 268/ 18 - 1992) for raw

cow's and ewe's milk intended for the production of fermented milk. The total count for cow's milk in this study ranged between $18-34 \times 10^3$ cfu/ml which is within the limit of the above standard.

Only limited data is available on the microbiological quality of ewe's milk. The total count for cow's milk in this study ranged between 6.2 to 12×10^3 cfu/ml. The above standard is also applied to ewe's milk it is evident that the quality of such milk is very good.

The psychotrophic count of both cow's and ewe's milk were good. The psychotrophic count in milk may explain the weaker coagulum after fermentation because of the proteolytic activity of these organisms (Cousin and Marth, 1977), or to the different casein fraction present in these milk (Juarez and Ramos, 1986). Coliform bacteria count in 1/10 diluted samples was positive.

Table (4.3) The microbiological quality of raw cow's and ewe's milk used in this study (cfu/ml).

Species	Initial total count	Psychotrophic count	Coliform bacteria count
Cow	18×10^3	25×10^2	Positive
Ewe	26×10^3	6.5×10^3	Positive

4.1.3 Buffer capacity of milk

Whole cow's and ewe's milk was concentrated (4:1) by using a UF unit as previously noted (see section 3.2.2). The whole milk retentate was diluted with fresh permeate to approximately 24, 26, 28, 30 and 32 % total solid levels.

Known amounts of lactic acid were added in the whole milk and the fortified UF

whole milk retentates from cow's and ewe's milk. The method is described in section 3.2.5.1.8. The mean of pH values are shown in Fig (4.1) for cow's milk, Fig (4.2) for ewe's milk and raw data are presented in Appendix (II - 1) for cow's milk and Appendix (II - 2) for ewe's milk. The buffer capacity at pH 5.3 for CA, CB, CC, CD, CE, and CF coded samples were 52, 75, 84, 89, 93 and 99 μmoles lactic acid / pH unit per gram milk medium respectively. The means are significantly different at the 95 % confidence level, between CA and CB, CC, CD, CE, CF coded samples, but the means are not significantly different at the same confidence between CB and CC, CD, CE, CF coded samples. The buffer capacities were 61, 81, 83, 84, 87 and 90 μmoles lactic acid / pH unit per gram milk medium for EA, EB, EC, ED, EE and EF coded samples respectively. The means are significantly different, at the 95 % confidence level, between EA and EB, EC, ED, EE, EF coded samples but the means are not significantly different between EB and EC, ED, EE, EF coded samples. The buffer capacity of cow's and ewe's milk are significantly different, at the 95 % confidence level, for ^{cow's} all samples. This difference can be attributed to total solids, especially protein and minerals content. Some constituents of milk are responsible for such buffer action and according to Jeness and Patton (1959) and Sutherland and Jameson (1981), these are carbon dioxide, protein, phosphates, citrates, lactate and other minor miscellaneous constituents. The buffer capacity of the diluted UF retentate milks are greater than whole milk due to higher total solid but the maximum buffering capacity occurs in the same region of between pH 5.1 - 5.3. Covacevich and Kosikowski (1979) reported an exponential relationship between retentate buffer capacity and total solids level. When milk is ultrafiltered these components are concentrated and cause a concomitant increase in buffer capacity (Brule et al, 1974; Covacevich and Kosikowski 1979). As a result pH reduction becomes difficult despite the presence of many active starter bacteria in growth phase. This is because large amounts of acid are required to overcome the increased buffer capacity of UF retentate. Similar buffer capacity effect may also be due to residual organic acids (More et al, 1973). Hickey et al (1987) reported that the buffering capacities at pH 5.8 for a 39.4 % total solids level cow's milk retentate and 11.2 % whole milk were 118 and 52 μmoles lactic acid / pH unit per gram milk medium respectively.

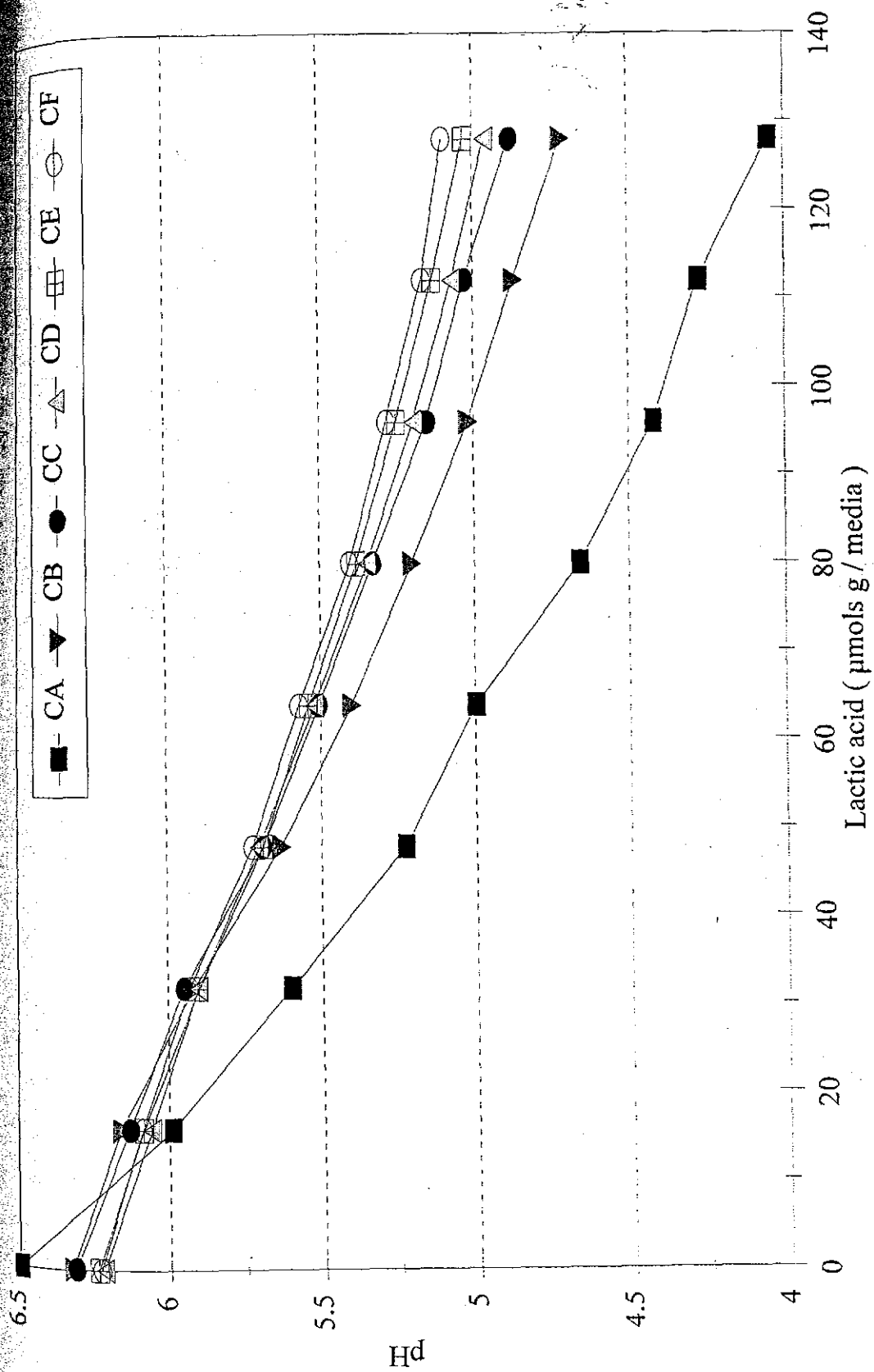


Fig (4.1) Buffer capacity of fortified UF retentate for cow's milk

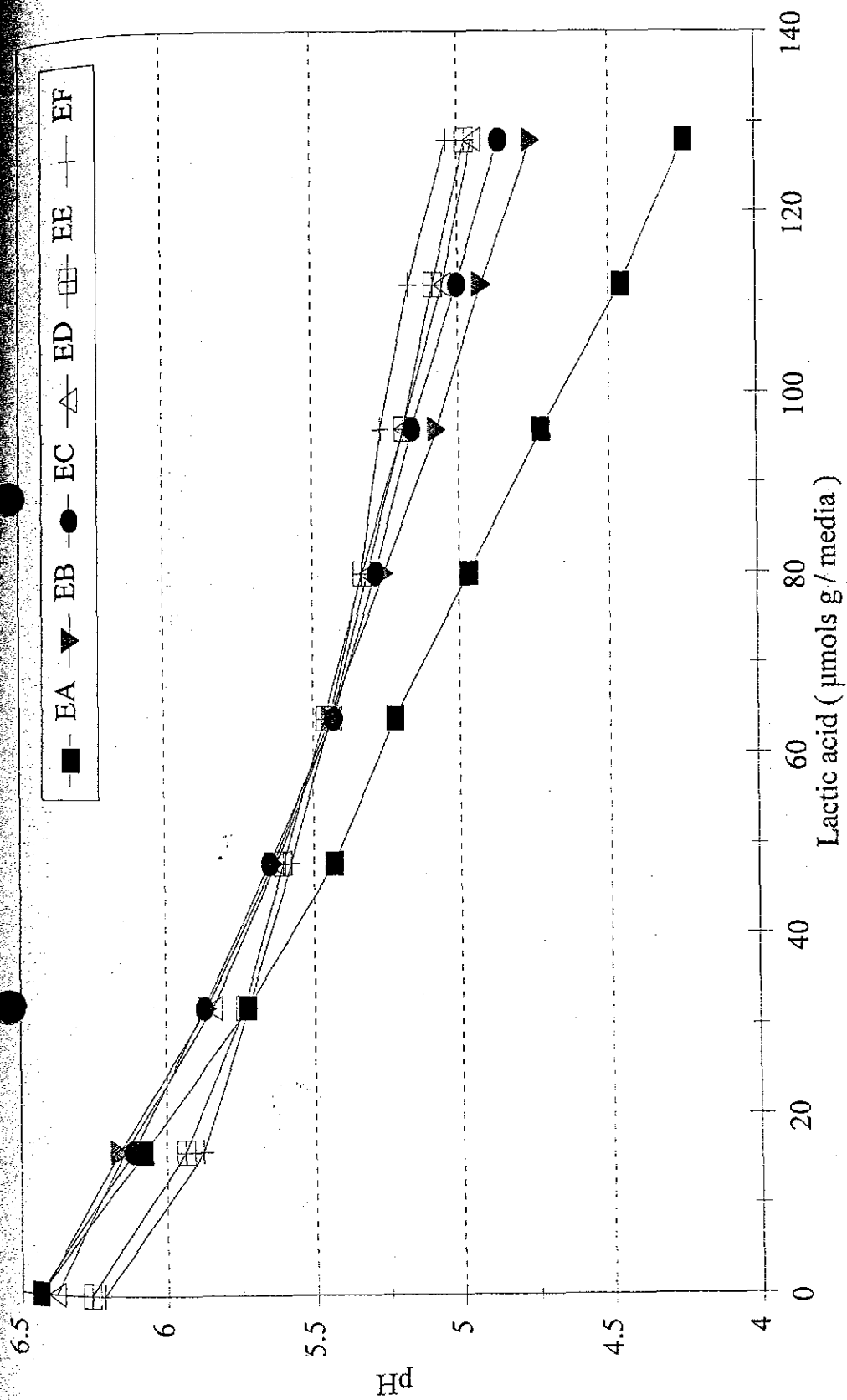


Fig (4.2) Buffer capacity of fortified UF retentate for ewe's milk

4.2 Changes of titratable acidity and pH of whole milk and fortified UF retentate during incubation period

Fermentation of whole milk and fortified UF retentate milk of different total solids levels were carried out using 3 per cent commercial starter culture (coded CH-1) and incubated for 180 min for CA, EA coded samples and 270 min for CB-CF and EB-EF coded milk samples at 41 ± 1 °C.

The comparative rate of acid development and pH changes for whole and fortified UF milk retentate are presented in Fig (4.3) for cow's milk and Fig (4.4) for ewe's milk. The raw data are also presented in Appendix (III - 1) for cow's milk and Appendix (III-3) for ewe's milk. The acid production in milks at the end of the incubation time reached 0.91, 1.39, 1.60, 1.63, 1.65 and 1.68 % lactic acid for CA - CF coded samples and 0.92, 1.39, 1.53, 1.56, 1.63 and 1.65 % lactic acid for EA-EF coded samples respectively. These results show the final titratable acidity level from different total solids milk samples. However, the level of titratable acidity given above, in fact, include the natural acidity of the milk. Although the developed acidity of yogurt increased from 0.7 % (CA) to 1.1 % lactic acid (CF) and 0.72 % (EA) to 1.01 % lactic acid (EF) coded samples the significance of the increases is probably marginal and may simple reflect a slight increase in the starter activity with the increases of the total solids in the milk. The data indicate that the rate of acid development and titratable acidity changes in both cow's and ewe's milk were more or less similar at same total solids levels.

It is clearly evident from Fig (4.3) and (4.4) that the increase of total solids level (especially protein and minerals) in milk affected the acid production and the final acidity. The production times were extended by up to 120 min when the fortified milk retentate was used. Similar observation has been reported in literature (Hickey et al 1983, Mistry and Kosikowski 1986). This may be due to the alteration of milk composition as a result of the ultrafiltration process thereby influencing the growth and acid production of starter bacteria.

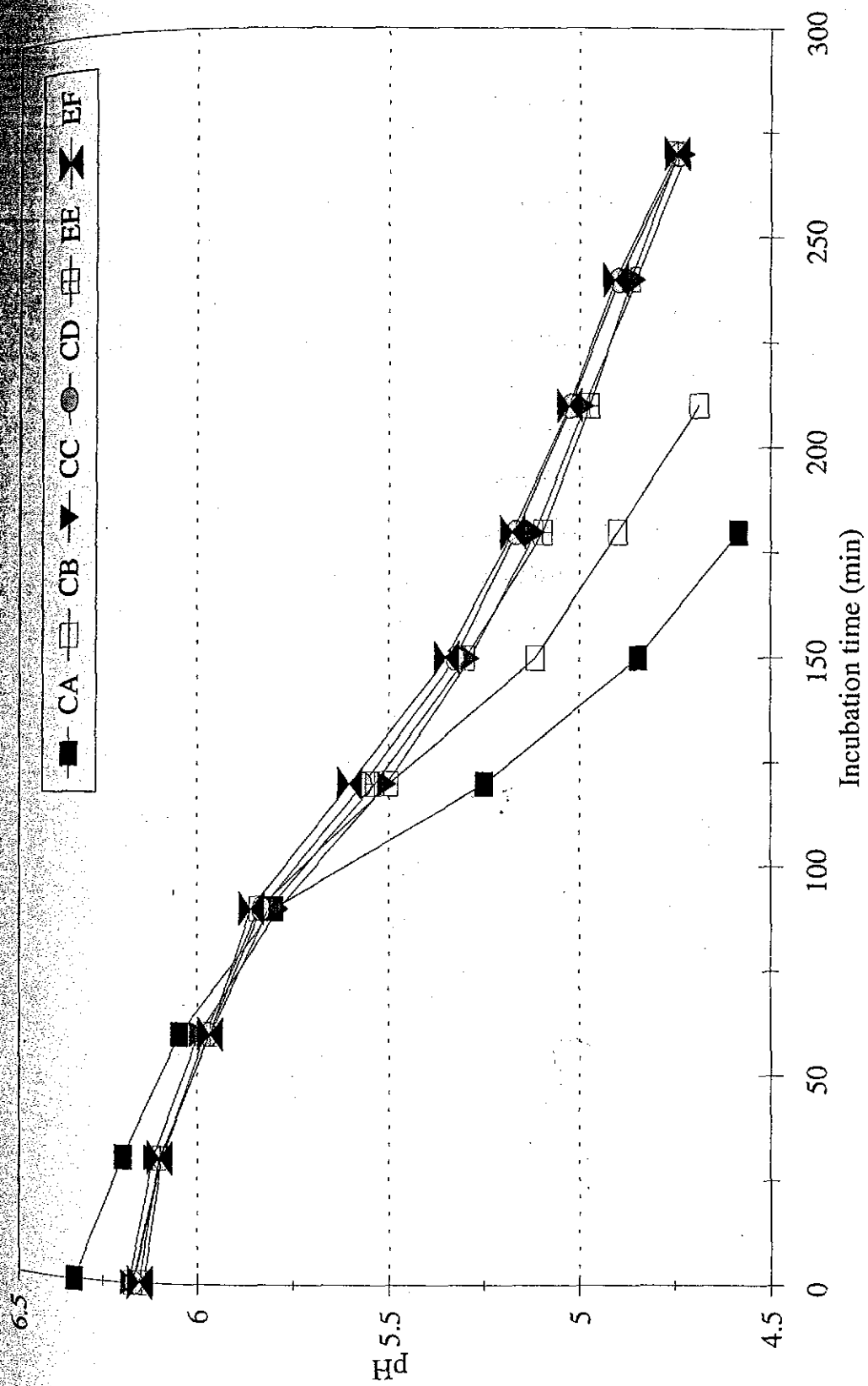


Fig (4.3) Changes of pH from different total solids UF cow's milk retentate during incubation period

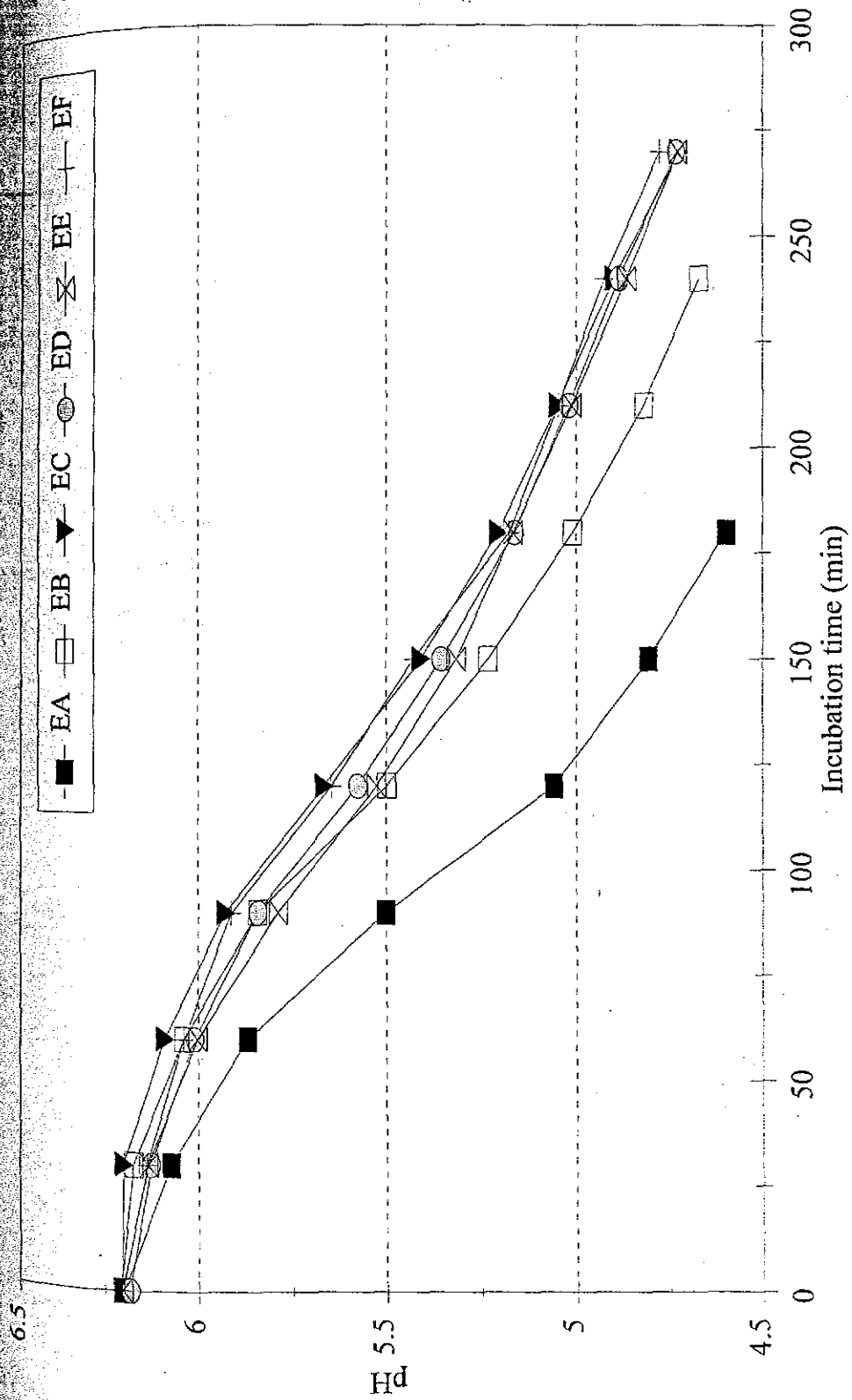


Fig (4.4) Changes of pH from different total solids UF ewe's milk retentate during incubation period

The acid development was monitored by pH and the results were plotted as pH versus incubation time. These are presented in Fig (4.5) for cow's milk and Fig (4.6) for ewe's milk. The raw data are presented in Appendix (III-3) for cow's milk and in Appendix (III-4) for ewe's milk. The pH changes were affected by total solids. The pH changes are similar between species (cow and ewe) at the same total solids levels. This can be observed from the data given in Fig (4.5) and (4.6). It can be attributed to the following reasons: - Ultrafiltration process increases the content of protein and others milk compounds (Ca, P) in UF milk retentate and its buffer capacity is increased (Covachevich and Kosikowski, 1979; Sutherland and Joneson, 1981). As a result, the amount of lactic acid that yogurt starter bacteria must produce to cause a unit changes in pH is increased considerably over that needed when UF milk retentate is used (Mistry and Kosikowski, 1985, 1986).

Hoier (1984) obtained a pH of 4.63 with a yogurt culture at the end of an unspecified time at 40 °C in retentate containing 14 % protein. Growth of starter organisms was unaffected by the high content of total solids or ash in retentates. However, the results obtained by Patel et al, (1986) did not agree with results was noted by Hoier (1984). In the present study, the results confirmed the results obtained by Hoier (1984). The determination of pH is valuable, for the increase in acidity or lowering of pH causes a change in physico-chemical characteristics of the milk. Thus, although visible coagulation started after 1.5-3 hours of incubation at $41 \pm 1^\circ\text{C}$, the transformation of the milk from a liquid state to a junket form could be clearly observed when the pH was between 4.5 - 5.2.

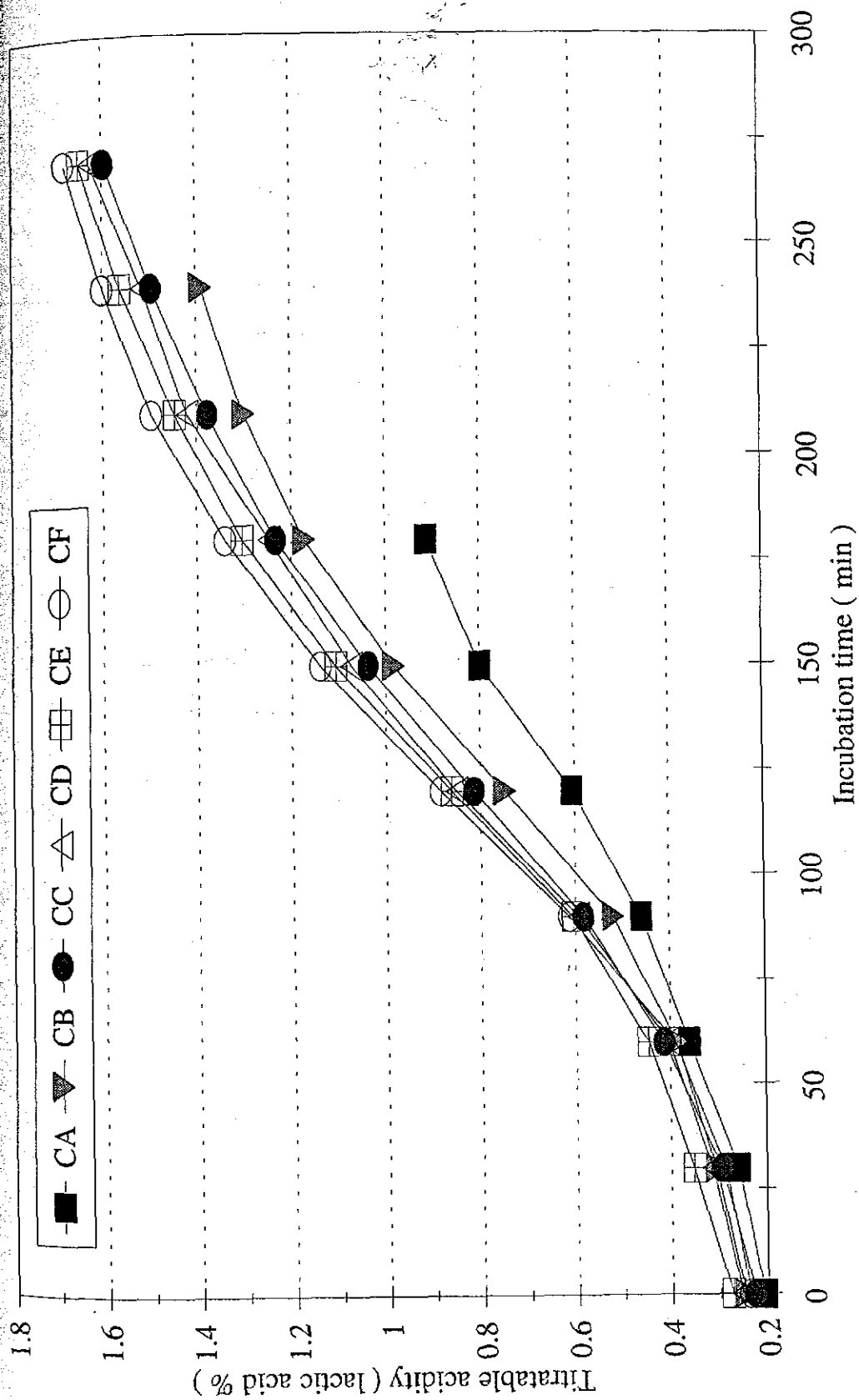


Fig (4.5) Titratable acidity measurement during incubation time from cow's milk

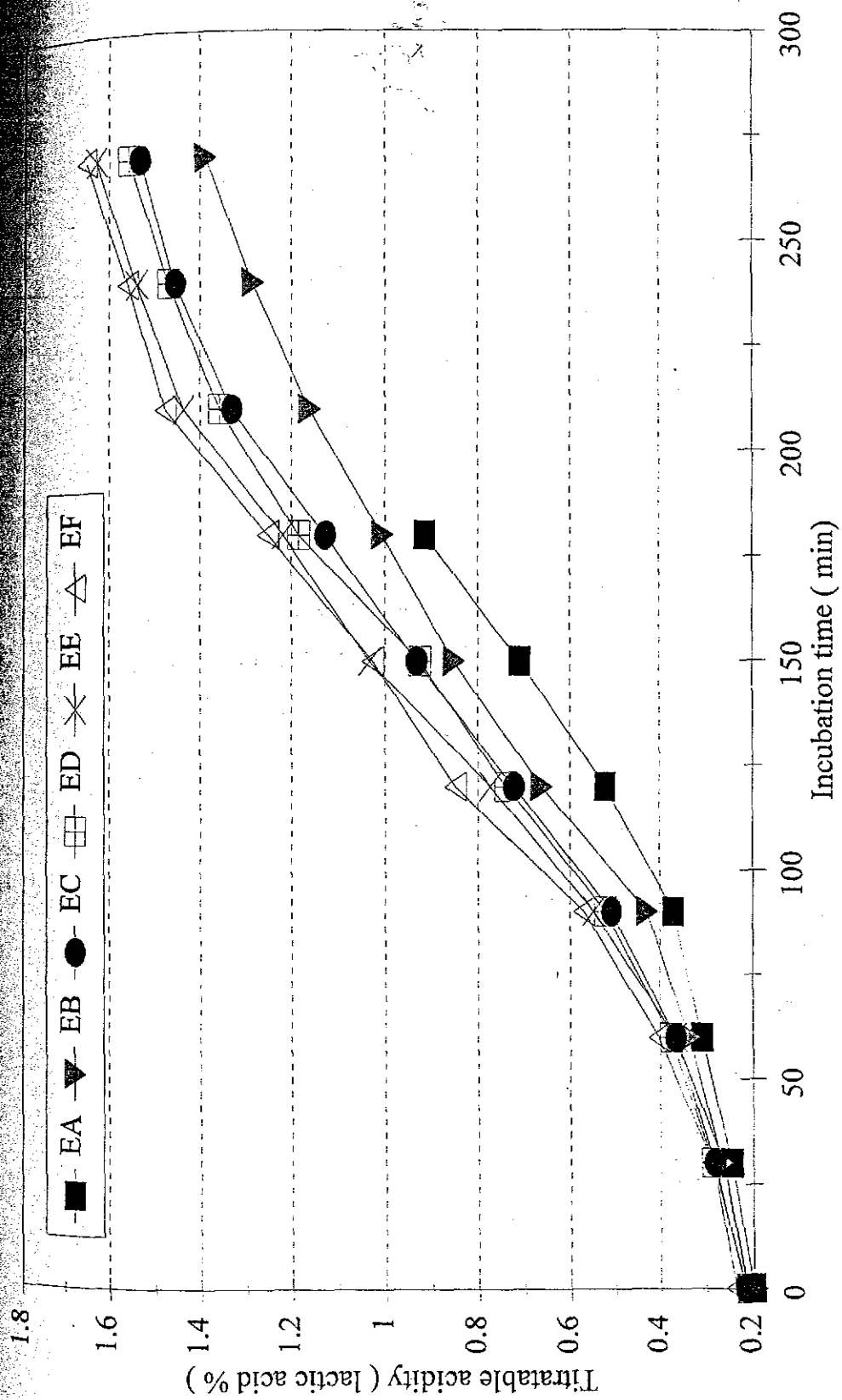


Fig (4.6) Titratable acidity measurement during incubation time from ewe's milk

4.3 Some chemical, physical and microbiological properties of thick yogurt produced from cow's and ewe's milk using different production processes.

4.3.1 The production methods of set and stirred style thick yogurt.

Stirred style thick yogurt (süzme or torba yogurt) is a fermented dairy product which is produced in Turkey and some other countries. The traditional method of production consists of straining natural yogurt using a cloth bag. The quality of stirred style thick yogurt using the cloth bag may be affected by factors such as the level of total solids in the original milk, the type of starter culture and the temperature of whey drainage. Some yogurt starter cultures producing hydrocolloid polysaccharides are not suitable to produce thick yogurt by using cloth bag method because the whey drainage will take a long time. The draining temperature may have an effect on the yield and quality of the product. Hamad and Al-Sheikeh (1989) concluded that the removal of the whey from yogurt at 25°C resulted in lower yields when compared with yogurt concentrated at 5°C. Also the final product can have a higher acidity than the initial yogurt.

An alternative method for the manufacture of thick yogurt is the UF method (Veinoglou et al, 1978; El-Samragy and Zall, 1988). The fermentation of UF milk retentate for the production of thick yogurt can be defined as a set style thick yogurt because fermentation stage is carried out in the retail container. The yogurt produced by traditional or modified traditional method can be defined similarly with stirred style yogurt because the yogurt gel is broken during filtration stage. It is evident that set style thick yogurt made from fortified UF retentate and stirred style thick yogurt made from natural yogurt by using traditional or modified traditional method may not have same characteristics.

4.3.1.1 Transfer rate of total solid during the concentration

The transfer rate of components from whole fresh milk to the final product were compared by the two methods of UF and modified traditional filtration. The differences were calculated on the total solids basis. The result of transfer ratio of the total solids for UF method ranged from 67 % for 32 % total solid level from fortified retentate to 73 % for 24 % total solids level from fortified retentate. The transfer ratio of total solids for modified traditional method using cotton filter medium was 65 % and 62 % for synthetic filter medium with 5 ins.Hg vacuum level.

The total solids transfer rate for thick yogurt produced using modified traditional method was 65 % compared to 73 % in yogurt produced from UF retentate which means the increases in transfer rate was up to 8 % depending on final total solids level in product. This amount caused an increase in thick yogurt yield by around 10%. This increases can be attributed to higher protein, fat and ash transfer ratio. Some of the minerals are bonded with protein. Due to this ash content of retentate is high.

It can be concluded that the results reflect economical and industrial advantages. These are the saving of milk by up to 10 % required for a given amount of thick yogurt which is turn means that a more profitable utilisation of the milk and a reduction in the amount of some materials used for yogurt making; such as starter cultures (up to 80 %). The UF method reduces the production time of thick yogurt. This condition is related to membrane area. A more hygienic product can be produced with UF method.

4.3.2 Chemical composition of yogurt and permeate

4.3.2.1 The chemical composition of fortified set-style thick yogurt made from different milk.

The thick yogurts were made in October - March 1992 -93 for cow's milk and February - July 1993 for ewe's milk. Yogurt made from fortified UF milk retentate was carried out using five different total solids levels. The original whole cow's milk contained 12.18 % total solids, 3.42 % protein, 4.02 % fat, 4.63 % lactose and 0.71 % ash based on the average of three samples. The original whole ewe's milk contained 17.12 % total solids, 5.42 % protein, 6.1 % fat, 5.04 % lactose and 0.91 % ash.

The mean chemical composition of set style thick yogurts, made from fortified UF milk retentate, are summarised in Table (4.4) and each trial illustrated in Appendix (II-1) for cow's milk yogurt and in Appendix (II-2) for ewe's milk yogurt.

It is evident from the data shown in Table (4.4) that the total solids, including protein, fat and ash contents, have been proportionally increased in all set-style thick yogurt samples coded CB - CF and EB - EF compared to the milk where the lactose content was reduced in all set-style yogurts to a level that ranged between 4.7 - 4.01 % and 5.07 - 4.65 % for cow's and ewe's milk yogurts respectively. The concentration factor of protein, fat and ash in set-style thick yogurts were in range of 2.35 - 3.53:1, 2.51 - 3.65:1 and 1.53 - 1.99:1 for cow's milk respectively and 1.56 - 2.35:1, 1.43 - 2.25:1 and 1.35 - 1.64:1 for ewe's milk respectively.

It can be concluded that concentration factors were high for fat and protein and low for minerals. During the concentration of milk by using the UF unit, some minerals passed through the membrane. Although concentration factor is low for ewe's milk yogurts, protein levels are slightly higher than cow's milk yogurt. This originated from the protein level of the initial milk. Lactose in yogurt slightly decreased compared to the initial milk with high concentration level. This was an expected

result because lactose can pass through the membrane.

4.3.2.2 Chemical composition of yogurt made using the modified traditional method

a) The modified traditional filtration experiment was carried out using three different vacuum levels (0, 5 and 10 ins. Hg) and two different filter media namely cotton and synthetic cloth. Their characteristics are described in section 3.1.1.3. These were aimed at providing total solid levels between 26 - 28 % for subsequent production of yogurt. The chemical composition of yogurt and stirred style thick yogurt is given in Table (4.4) and raw data in Appendix (II-3) and (II-4) for cow's and ewe's milk yogurt, both for cotton filter medium and synthetic filter medium.

The total solids, protein, fat lactose and ash contents of cow's milk yogurt for the different vacuum levels ranged between 15- 26 %, 5- 10 %, 5- 9.45 %, 4.46 -4.55% and 0.73 - 0.71 % for cotton filter medium and 15 - 25 %, 4.25 - 9.76 %, 5 -11.5%, 4.5 - 4.17 % and 0.74 - 0.73 % for synthetic filter medium respectively. It can be observed that the fat content in the stirred style thick yogurt, produced using modified traditional method with cotton filter cloth as filter media, was higher than that of a parallel sample made by the synthetic fibre filter media. This was mainly due to the efficiency of recovery of fat when using the cotton fibre filter medium. This level of total solid for the different types of concentrated thick yogurt were similar to the data previously reported by Tamime et al (1989) and Tamime and Robinson (1988). There are some differences between the different processed products. Such differences can be attributed to process technique. It was difficult to control the rate of whey drainage when using the traditional production method (natural filtration), for this reason the processing method was modified by applying constant vacuum to be able to control whey drainage.

The average of the total solids content of stirred style thick yogurt, produced using the modified traditional filtration method and with different types of milk ranged

from 25 - 27 %. Such levels meet the existing Saudi Arabian and Lebanese standard, but they are well below the Turkish Food Act. Nevertheless, the composition standard of all type of stirred style thick yogurt made from cow's milk were similar to those reported by Hofi (1988).

Table (4.4) Chemical composition of whole milk, set and stirred style thick yogurts produced using different production methods.

Product codes	Total solids % (w/w)	Protein % (w/w)	Fat % (w/w)	Lactose % (w/w)	Ash % (w/w)
CA	12.18 ± 0.09	3.42 ± 0.05	4.02 ± 0.06	4.63 ± 0.10	0.71 ± 0.01
CB	24.03 ± 0.10	8.07 ± 0.04	10.10 ± 0.05	4.70 ± 0.15	1.09 ± 0.03
CC	26.03 ± 0.20	8.64 ± 0.06	11.70 ± 0.50	4.53 ± 0.03	1.16 ± 0.20
CD	28.08 ± 0.20	9.67 ± 0.05	12.60 ± 0.20	4.42 ± 0.01	1.21 ± 0.45
CE	29.93 ± 0.16	10.73 ± 0.10	13.67 ± 0.10	4.23 ± 0.07	1.28 ± 0.10
CF	31.99 ± 0.04	12.10 ± 0.20	14.70 ± 0.15	3.91 ± 0.05	1.41 ± 0.02
CG	26.57 ± 0.21	10.36 ± 0.15	10.47 ± 0.21	4.55 ± 0.15	0.72 ± 0.01
EA	17.12 ± 0.16	5.42 ± 0.08	6.10 ± 0.36	5.04 ± 0.11	0.91 ± 0.03
EB	24.17 ± 0.29	8.47 ± 0.10	8.77 ± 0.20	5.02 ± 0.06	1.23 ± 0.05
EC	26.25 ± 0.33	9.06 ± 0.21	10.47 ± 0.37	5.01 ± 0.18	1.33 ± 0.07
ED	27.95 ± 0.26	10.28 ± 0.17	10.73 ± 0.25	5.07 ± 0.40	1.40 ± 0.05
EE	30.08 ± 0.22	11.70 ± 0.15	11.83 ± 0.25	4.86 ± 0.22	1.50 ± 0.05
EF	31.98 ± 0.14	12.77 ± 0.15	13.77 ± 0.10	4.65 ± 0.60	1.50 ± 0.11
EG	26.51 ± 0.35	9.92 ± 0.25	11.80 ± 0.30	4.23 ± 0.07	0.88 ± 0.05

b) Some commercial traditional thick (süzme) yogurt from Turkey, produced using natural filtration, was also analyzed to compare with stirred style thick yogurt produced using the modified traditional method. These samples were collected from different firms in Turkey. The results are presented in Table (4.5). The chemical composition of commercial traditional thick yogurts varied over a large range. This variation can be attributed to lack of homogeneity after concentration.

Table (4.5) Chemical composition of commercial süzme yogurt produced in Turkey.

Components	Minimum	Maximum
Total solids %	20.87	30.65
Fat %	6.8	10.3
Protein %	9.8	15.2
Titrateable acidity (% La)	2.08	3.2
pH	3.46	3.7

4.3.2.3 Chemical composition of permeate from UF and modified traditional filtration

The mean chemical composition of permeate obtained from cow's and ewe's milk yogurt by using vacuum filtration for different vacuum level with filter media and for ultrafiltration of milk are given in Table (4.6) and raw data for permeate or yogurt whey, obtained from modified traditional filtration, in Appendix (II-3) and (II-4). The total solids, protein, fat, lactose and ash contents of permeate for cow's milk yogurt ranged between 5.28 - 6.4 %, 0.21 - 1.10 %, 0.1 - 0.90 %, 4.23 - 4.47 % and 0.67 - 0.72 % for cotton filter medium and 5.54 - 8.01 %, 0.45 - 1.16 %, 0.2-1.10 %, 4.3 - 4.36 % and 0.7 - 0.73 % for synthetic filter medium respectively.

Permeate for ewe's milk yogurt range between 5.76 - 7.21 %, 0.22 - 0.9 %, 0.2 - 0.3 %, 4.6 - 4.69 % and 0.81 - 0.89 % for cotton filter medium and 5.86 - 10.1 %, 0.3 - 2.2 %, 0.2 - 1.10 %, 4.71 - 4.68% and 0.85 - 0.88 % synthetic filter medium respectively. The total solids, protein, lactose and ash content of permeate from ultrafiltration of milk were 5.41 %, 0.19%, 4.74 % and 0.49 % for cow's milk and 5.3 %, 0.23 %, 4.63 % and 0.44 % for ewe's milk respectively

Total solids level of yogurt whey provided by synthetic filter medium is higher than cotton filter medium. Such differences can be attributed to the different pore size of filter medium because all others parameter were similar. Total solids and protein level in permeates are slightly lower than result reported by Tamime et al (1991 a). This can be attributed to process conditions. The total solids losses were also related to applied vacuum. The total solids losses in the yogurt whey consisted mainly of lactose, mineral and some organic acid.

Permeate were obtained from different process (UF of milk and modified traditional filtration of yogurt). Protein losses were observed in both permeates from UF and modified traditional filtration (see Table 4.6). Protein losses were in permeate from UF process is most likely to be soluble nitrogen compounds but protein in permeate from modified traditional filtration may be protein and some soluble nitrogen compounds. However, higher protein losses were found in the permeate from modified traditional filtration than UF permeate. As it can be seen from the results high protein losses were observed in the modified traditional method. Protein losses with permeate varied with filter media (cotton and synthetic) and applied vacuum levels. The highest protein losses were observed with synthetic filter media and using 10 ins.Hg vacuum than with other conditions. Lowest protein losses were observed when cotton filter media and no vacuum applied. This variation originates from the properties of the material which were used as filter media. The result obtained from UF process confirmed the data reported by Yan et al (1979) and were slightly lower than the results reported by Covachevich and Kosikowski (1979)

Table (4.6) Chemical composition of permeate obtained from UF and modified traditional filtration

Product Codes	Filter media	Total solids % (w/w)	Protein % (w/w)	Fat % (w/w)	Lactose % (w/w)	Ash % (w/w)
UF-CP		5.42	0.19	non	4.74	0.49
UF-EP		5.30	0.23	non	4.63	0.44
T-C. No Vacuum	Cotton	5.28	0.21	0.10	4.47	0.67
T-C. 5 ins.Hg Vac.	Cotton	5.91	0.85	0.15	4.21	0.72
T-C. 10 ins.Hg Vac.	Cotton	6.32	1.10	0.90	4.23	0.71
T-C. No Vacuum	Synthetic	5.54	0.45	0.20	4.30	0.70
T-C. 5 ins.Hg Vac.	Synthetic	5.99	0.92	0.25	4.36	0.73
T-C. 10 ins.Hg Vac.	Synthetic	8.00	1.10	1.10	4.30	0.73
T-E. No Vacuum	Cotton	5.76	0.22	0.20	4.60	0.81
T-E. 5 ins.Hg Vac.	Cotton	6.03	0.64	0.10	4.35	0.84
T-E. 10 ins.Hg Vac.	Cotton	7.21	0.90	0.30	4.90	0.89
T-E. No Vacuum	Synthetic	5.86	0.30	0.20	4.70	0.85
T-E. 5 ins.Hg Vac.	Synthetic	7.32	1.22	0.20	4.68	0.90
T-E. 10 ins.Hg Vac.	Synthetic	10.10	2.20	1.10	4.68	0.88

UF - CP : Permeate from cow's milk

UF - EP : Permeate from ewe's milk

T-C : Permeate from traditional filtration for cow's milk

T-E : Permeate from traditional filtration for ewe's milk

4.3.2.4 Minerals content in yogurts

Mineral contents of set-style thick yogurts are given in Fig (4.7) and (4.8) and raw data are presented in Appendix (II-5) and (II-6). It is difficult to say definite things about these results because increases or decreases were not regular. However, mineral contents of thick yogurts made from fortified UF milk retentate were generally greater compared to control milk. The mineral contents of yogurt, produced by using modified traditional method were smaller compared to initial milk. This difference can be attributed to production methods. Thick yogurt, produced by using modified traditional method, was concentrated after fermentation. Some of the mineral dissolved into the water phase during the fermentation. These minerals can pass through the filter. However set-style thick yogurt, made from fortified UF retentate, was produced after concentration of milk. Some minerals are partly bound to protein, namely, calcium, magnesium and phosphate. Ca, Mg and P will be retained with protein in retentate during ultrafiltration process, but some of the minerals are free and they may pass through the membrane. The high content of minerals in UF retentate is a consequent of the association of calcium and phosphorus with the casein micella complex. Similar trends in retentate composition and permeation rates were reported previously by Covacevich and Kosikowski (1979) and Glover (1986).

4.3.3 Titratable acidity and pH of thick yogurts

After the incubation period, yogurt samples were transferred to refrigerator and held at 4 °C. The total titratable acidity and pH were measured after 24 hours storage. The method is described in section 3.2.5.2.2. The mean values of triplicated runs are presented in Table (4.7) and the raw data are also presented in Appendix (III-5) and (III-6) for all yogurt samples.

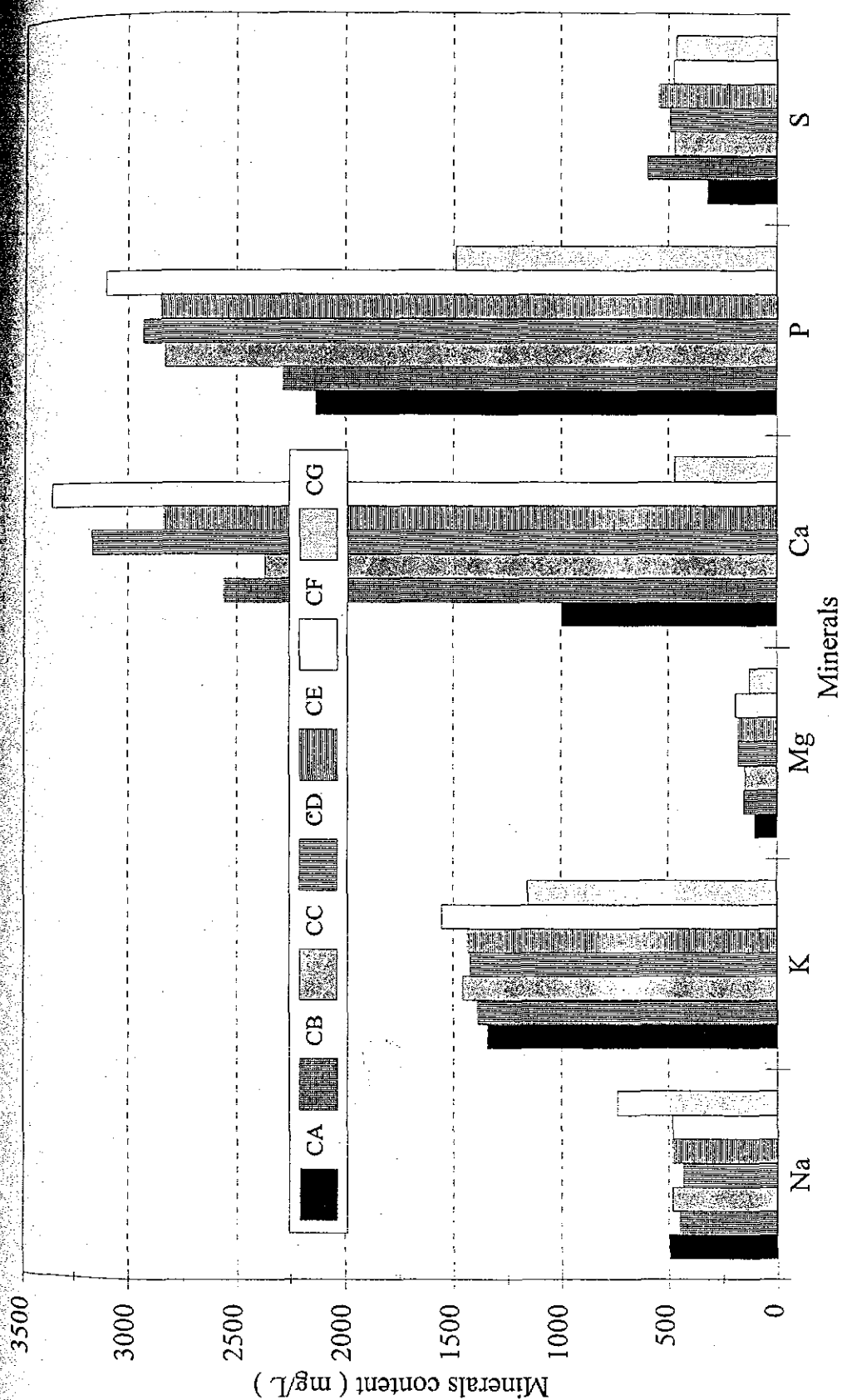


Fig (4.7) Mineral content of set and stirred style yogurt produced from cow's milk using different production methods.

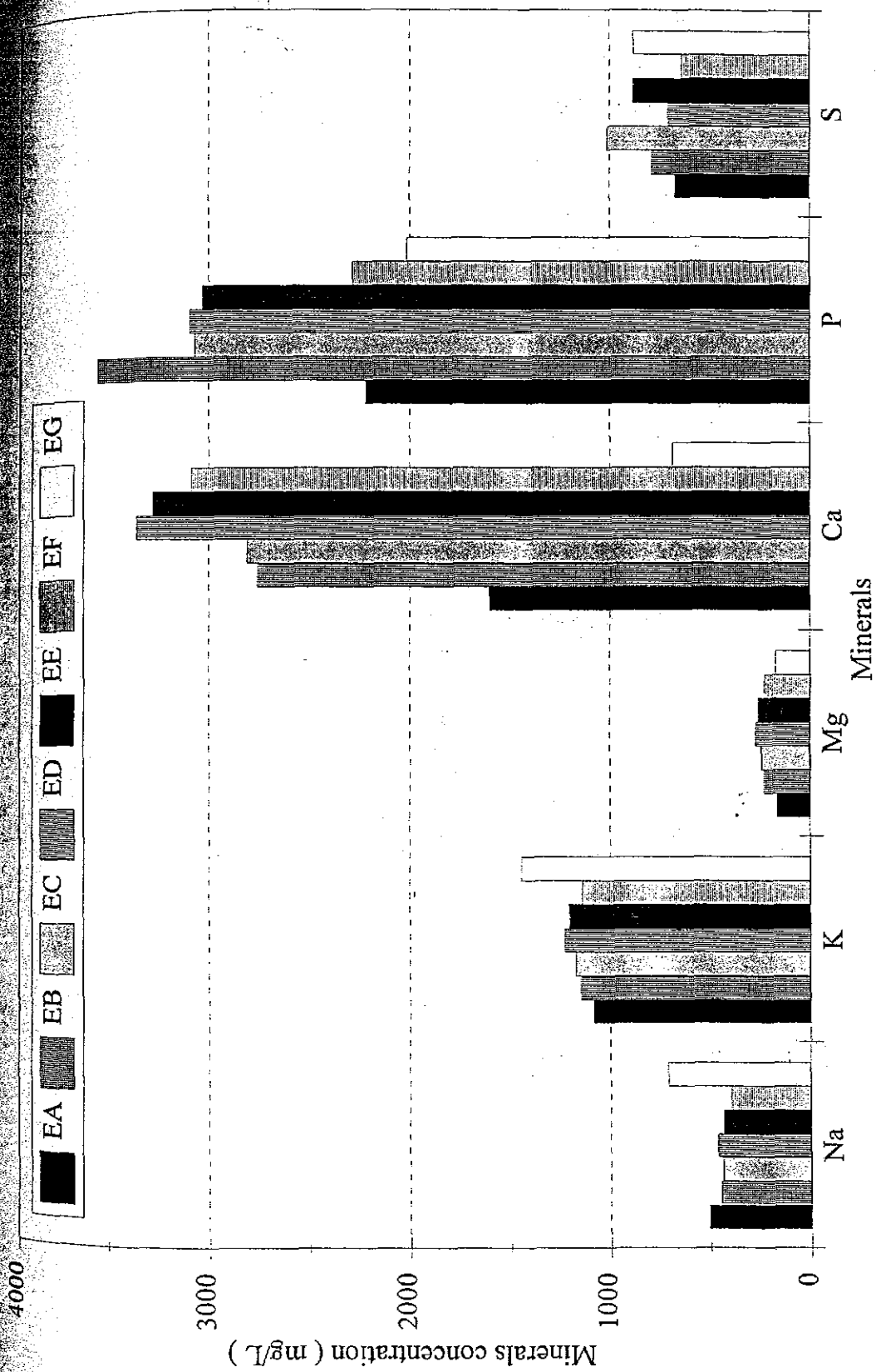


Fig (4.8) Minerals content of set and stirred style yogurt produced from ewe's milk using different production methods.

Table (4.7) Titratable acidity and pH in different types of yogurt after 24 hours storage at 4 °C in refrigerator.

Product codes	Titratable acidity (% La)	pH	Products codes	Titratable acidity (% La)	pH
CA	1.08	4.38	EA	1.18	4.44
CB	1.45	4.51	EB	1.44	4.49
CC	1.69	4.53	EC	1.56	4.51
CD	1.72	4.56	ED	1.60	4.52
CE	1.74	4.67	EE	1.65	4.65
CF	1.77	4.72	EF	1.69	4.69
CG	1.18	4.27	EG	1.04	4.24

24 hrs after the incubation period, the total titratable acidity increased 18.7 % (CA), 29.7 % (CG) and 5 % (CB - CF) coded sample and 28 % (EA), 12.8 % (EG) and 2 % (EB - EF) coded samples respectively because of the cooling down and 24 hours storage time. It can be seen from the data presented in Table (4.7) that the increases of the total titratable acidity were small in thick yogurts (CB - CF and EB - EF coded samples). The production of lactic acid was affected by total solids level of milks. The means values of total titratable acidity were significantly different at the 99 % confidence level between controls (CA, EA) and CB - CF, EB-EF coded samples. The rate of acid development of yogurt starter culture in milk was uniform and all the thick yogurt samples using the modified traditional method had similar acidity. These are 1.18, 1.04 % lactic acid for CG and EG respectively. The pH level was 4.25, 4.27 for CG and EG on the refrigerated product respectively. The mean values of pH were 4.38 for control (CA) and between 4.15 - 4.72 for CB - CF coded sample and 4.44 for control EA and between 4.49 - 4.69 for EB - EF coded sample respectively. The results indicated that the mean values of the pH for CE, CF, EE and EF coded samples were higher than other thick yogurt samples. This can be attributed to high buffer capacity of concentrated milk. These result confirm previously reported data by Tamime et al (1989 a, b) and Tamime et al (1991 a).

However, it was not significantly different between set and stirred style thick yogurt (CB-CG and EB-EG coded) for both species (cow and ewe). These acidity levels were in range of Turkish Food Act, which has a maximum 2.25 % lactic acid. The titratable acidity level of all thick yogurt samples were lower than commercial traditional Turkish süzme yogurt. This differences may originate from process condition. The production time of commercial traditional thick yogurt is long. During the drainage period in room temperature acidity might increase. Over-acidification is observed most frequently with natural yogurts. This property contributes to other defects such as syneresis and increased reduction of viable count in product. It is mainly due to the uncontrollable growth of strains of *L. bulgaricus* at low pH values and refrigerated temperatures. It can be improved to a limited extend by applying good manufacturing practice and by using starter cultures with reduced over-acidification behaviour. The significance of the increase may simple reflect a slight increase in the starter activity with the increases of total solid in the milk. This may be due to the alteration of milk components or release of stimulating factors in milk as a result of the UF process thereby influencing the acid production of starter bacteria.

It can be concluded that in this study, the results of the acidity level of CB-CG and EB-EG coded sample, produced by different production methods, indicated that thick yogurt with low pH and titratable acidity can be produced using fortified UF retentate and the modified traditional filtration method.

4.3.4 Water Holding Capacity of yogurt

The water holding capacity (WHC) of yogurt is shown in Fig (4.9) and raw data in Appendix (IV-1) and (IV-2) for cow's and ewe's milk yogurt samples respectively. Lowest WHC was demonstrated by control yogurt (CA) and (EA). The average mean value was 70.78 % for (CA) and 55.09 % for (EA) coded samples. The highest WHC was demonstrated by CF coded yogurt. The average WHC value of CF coded sample was 86.4 %. The WHC of set style thick yogurts, coded as CB,

CC, CD, CE and CF₂ made from fortified UF retentate, are 23.01 %, 21.52 %, 19.44 %, 16.16 %, and 11.6 % respectively. The WHC of EB, EC, ED, EE and EF coded samples are 24.94%, 22.29 %, 20.75 %, 17.42 % and 13.81 % respectively. The water holding capacity of stirred style thick yogurts coded CG and EG, produced using modified traditional method, were 16.78 % and 15.93 % respectively. As it can be seen from the data there is a sharp increase between CA and CB - CF and slight increases between CB, CC, CD, CE and CF. There is a sharp increase between EA and EB - EF and slight increases between EB, EC, ED, EE and EF coded sample. The means of water holding capacity are very significant between CA and CB, CC, CD, CE, CF but the means of CB - CF coded samples are not significantly different at the 99 % confidence level. Also, the means are significantly different in yogurt made by using modified traditional method (CG coded sample) and its equivalent (CC or CD coded sample) set style thick yogurt, made from ^{Concentrated} fortified UF retentate at the 99 % confidence level. In this period, the WHC of set style thick yogurt, made from fortified UF retentates, are slightly decreased; this ^{increase} differences can be attributed to acidity development during storage period of the samples. Similar observation were observed in ewe's milk samples (EA - EF). Parnell-Clunies et al (1986) reported that the storage period did not significantly effect WHC. In this study the results agree with the results reported by Parnell-Clunies et al (1986).

Literature reports on the effect of total solids on WHC have cited significant positive effects of total solids especially protein (Becker and Puhan 1989). Changes in the structure of the casein gel can occur even after the network has been completely formed because the gel contracts. Under quiescent conditions, the gel may lose two-thirds of its volume, and up to nine-tenths (or even more) if external pressure is applied (Walstra et al, 1985). Rearrangements within the network produced by attractive forces between individual casein particles or clustered micelles can lead to additional inter-molecular bonds and, therefore, to a contraction of the gel and the expulsion of water. This phenomenon, called syneresis or whey separation, is thus caused by spontaneous release of water from the gel accompanied by a reduction in volume and is caused by changes in temperature and pH value and by mechanical

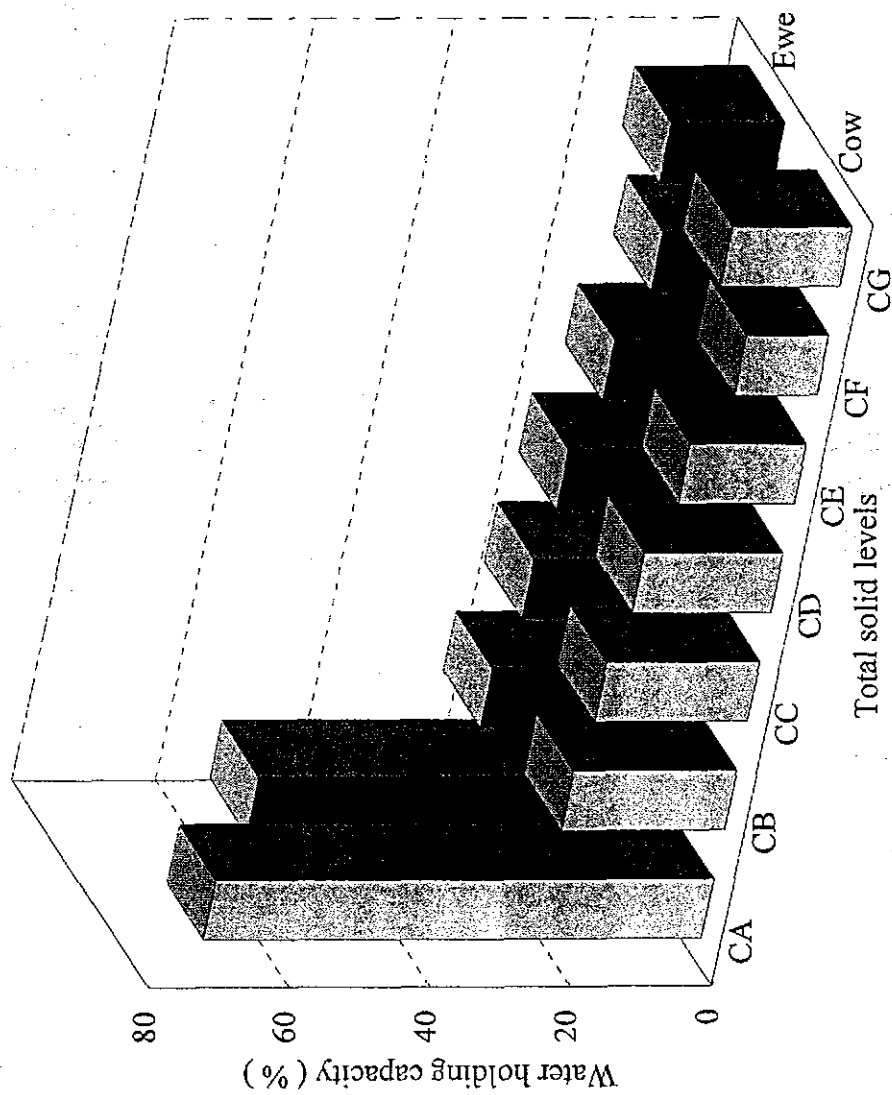


Fig (4.9) Water Holding Capacity of cow's and ewe's milk yogurt produced by using different production method

factors such as vibration (Dannenberg and Kessler, 1988). The factors that effect the whey separation from literature can be summarized as follows:

- i) the total solids content of milk, particularly protein content (Modler and Kaláb 1983; Modler et al, 1983; Mistry and Hassan, 1992)
- ii) heat treatment (Dannenberg and Kessler, 1988)
- iii) acidity level of coagulum (Rasic and Kurmann, 1978), iv) the content of salt in milk (Pette and Lolkema, 1951)
- v) dimension of the components in the protein matrix of yogurt.

Water is distributed within the protein gel in three different ways. These are namely capillary, bound and free or bulk water. The free water represents the greatest portion. It occurs in the casein network as invisible droplets. In contrast to the capillary and bound types of water, free water can be released from the gel by external influences.

Yogurt with increased total solids by adding high milk protein powder displayed minimal free whey, whereas the control yogurt showed whey separation. Microstructure examination of yogurt indicated that the control yogurt which was made from whole milk, had the most porous structure, whereas yogurts with high protein milk powder had lower porosity and the protein was partially fused together to a greater extent. Yogurts made with added high protein milk powder had the least whey separation. It has been shown in other studies that yogurts prepared with casein-supplemented milk have low whey separation because of water binding by casein (Modler and Kaláb 1983; Modler et al, 1983).

Susceptibility of whey separation is also affected by total solids content of milk. The increase in total solids content can be achieved in several ways (see section 2.5.2.2). By maintaining the same casein to non casein ratio, there are a high number of casein micelles per unit volume at a high total solids content which results in smaller compartments occupied by smaller compartments, a high number of casein micelles and a large amount of whey proteins per unit volume bind a large proportion of water. This was demonstrated by centrifuging the yogurts at different

centrifugal forces for 10 min (Harwalker and Kaláb, 1983). Only a part of the aqueous phase in yogurt may be separated from the protein matrix by centrifugation because the other part is firmly bound by protein. As the total solids content in the yogurt is increased, the volume of aqueous phase which may be separated is decreased. The matrix of low solids yogurt are subjected to noticeable compaction whereas matrices of high solids yogurts are compacted to a considerably lesser extent or not compacted at all.

As a conclusion, the results as presented in this section show a clear relation between the total solids level and the whey separation of the end product

4.3.5 Firmness of yogurt

The firmness of yogurt was measured using the penetrometer as illustrated in Fig (3.9). It must be emphasised at this stage that the depth of penetration (1/10 mm) is a function of the firmness of the yogurt (Kessler, 1981). The higher the depth of penetration the softer the coagulum or the lower the depth of penetration the harder the coagulum.

Firmness of the different samples of thick or concentrated yogurt as measured by the penetrometer are given in Table (4.8). Controls (CA and EA) and thick yogurts (CG and EG) gave higher penetration (lower firmness) as compared with CB - CF and EB - EF coded yogurt samples. According to Tamime et al (1989 b) unsmoothed UF retentate thick yogurt was the firmest despite its lowest total solids and protein contents because the yogurt made from UF retentate would resemble a set style yogurt whereas the other two yogurt samples would be similar to the stirred style thick yogurt. It is well-established fact that the structural damage to the coagulum cause low firmness.

Therefore, it can be observed that there is a relationship between the total solids level of milk and firmness. The firmness improved gradually with total solids level.

However, in this study, the change between (CB-CF coded samples) 24.32 % total

Table (4.8) Penetration results of different total solid thick-yogurt made from fortified UF milk retentate and by using modified traditional method.^(*)

Product codes	Penetration (1/10 mm)	Product codes	Penetration (1/10 mm)
CA	<400	EA	382
CB	210	EB	201
CC	180	EC	173
CD	166	ED	167
CE	146	EE	155
CF	143	EF	149
CG	215.5	EG	241.5

(*) Results are the average of three trials of cow's and ewe's milk. Each data from three different areas of the samples. Temperature was 7 ± 2 °C during the measurement. The measurement was made after overnight storage at 4°C

solid level was less pronounced. The changes of firmness between CA and CB - CF are significantly different. The changes of firmness between CB, CC and CD are also significantly different. The changes of firmness between CE and CF coded samples are significantly different at the level of 99 % confidence level. This differences may be attributed to compositional differences, origin of milk and protein matrix formation, including the casein chains and clusters. The effect of production methods (fortified UF milk retentate and modified traditional method)

on firmness is also significantly different. This differences may originate from type of yogurt, for instance, CG coded yogurt is a stirred yogurt. Coagulum was broken during the process for yogurt made using the modified traditional method but coagulum of yogurt, made by fortified UF milk retentate, was not broken. A similar observation was reported by Tamime et al (1989 b). Only limited data are available on the measurement of the curd tension of thick yogurt using a penetrometer. Stirred style thick yogurt was produced using traditional method with total solids level was between 22-24 %. A penetrometer has been used to measure the firmness of the samples by Tamime and Robinson (1978 b).

It was also used by Kessler (1981). The penetration reading from these samples ranged between 110 - 140 (1 / 10 mm penetration of the probe). A texture analyzer was also used on the different type of thick yogurt by Tamime et al (1989 a, b). The result was reported as following :

- i) Homogenized sample gave lower reading as compared with unhomogenised samples.
- ii) The results of homogenised sample were dependent on the level of total solid in sample. Labneh made from unhomogenised UF retentate showed the highest results. But after homogenization of the product the firmness was greatly reduced. This was due, primarily, to the formation and destruction of the coagulum.

The firmness of süzme (thick) yogurt made by modified traditional method was not uniform. Because the sample was not homogeneous. The layer of yogurt near to the filter medium was considerably thicker than the yogurt in the centre of the batch also it was lumpy. Consequently, a wide range of penetration results were obtained when sampled for analysis. However, reproducible results were obtained after the stirred style thick yogurt had been smoothed. Becker and Puhani (1989) reported that the yogurt made from UF milk retentate is a firmer body than yogurt made from evaporated milk and reconstituted milk. The influence of fat was also significant. It was concluded that the fat had a greater influence on the firmness than did the increase of solid non fat.

It can be concluded from the data in Table (4.8) that the high total solid thick yogurt (CB - CF), made from fortified UF milk retentate as set yogurt, had a higher firmness than the control yogurt (CA) and thick yogurt (CG) made using modified traditional method. However CG coded yogurt had a higher firmness than CA coded yogurt. Thick yogurt made from cow's milk was slightly different as compared to thick yogurt made from ewe's milk for all total solid levels and processing. The differences in firmness between the various thick yogurts cannot be explained merely by their total solid contents, especially protein contents. There is a probability that the differences in casein fractions may play an important role.

4.3.6 Volatile aroma compounds of yogurt

4.3.6.1 Analysis method of some volatile aroma compounds

Although more than 100 individual chemical compounds have been isolated from these type of fermented milk products (Badings and Neeter, 1980), only acetaldehyde, acetoin, diacetyl have great impact on the desired product flavour (Tamime and Robinson, 1985). Moreover, they are present in amounts detectable by common laboratory techniques. In these assay, no single method will provide a flavour profile truly representative of the food. The flavour constituents, typically, are released from the product by steam distillation. The volatile compounds must then be isolated from aqueous solution. This was done by using gas chromatography. As pointed out by De Haast et al (1978), the quantification of individual components using gas chromatography is the most specific method for accurate assessment of aroma components from fermented milk products. Steam distillation is a very popular method for flavour compounds isolation. The method is very efficient and artifacts may be minimized with adequate care. Capillary column, which provide very reproducible retention times, has become the standard for work in flavour area (Heath and Reineccious, 1986).

4.3.6.2 Volatile aroma compounds in set and stirred style thick yogurt

The aroma compounds of yogurt are mainly produced by carbonyl compounds including acetaldehyde, acetone and diacetyl. Some of the aroma compounds of the set and stirred style thick yogurts were determined according to the method as described in section 3.2.5.2.10. The samples were analyzed after overnight storage at $4 \pm 1^\circ\text{C}$. The level of individual volatile aroma components in yogurts were calculated from the chromatogram.

The volatile aroma compounds content of various products of set and stirred style thick yogurts, which have different total solids levels, are summarized in Table (4.9). Acetaldehyde in yogurt, produced by using modified traditional method, was lower than control and the others. This difference can be attributed to the production method because acetaldehyde is very volatile compound. Its amount was also lower in set style thick cow's milk yogurt than ewe's milk yogurt. The result of the volatile aroma compounds of set and stirred style thick yogurts at various total solid levels were statistically analyzed as follows:

a) Acetaldehyde

The means of acetaldehyde are significantly different between CA and CB-CF coded sample at the 99 % confidence level. Similar results were obtained for ewe's milk yogurt samples (EA-EF). However, the means of acetaldehyde are not significantly different between set style thick yogurt samples which were coded CB-CF and EB-EF. The means of the amounts of acetaldehyde are significantly different between species (cow and ewe) at the 99 % confidence level.

Table (4.9) Some volatile aroma compounds of yogurts after 24 hours storage at 4 °C in refrigerator

Products codes	Acetaldehyde (ppm)	Acetone (ppm)	Diacetyl (ppm)
CA	10.96	2.73	0.70
CB	15.37	3.57	0.80
CC	15.59	3.61	0.84
CD	16.31	3.59	0.87
CE	17.05	3.61	0.89
CF	17.89	3.65	0.91
CG	9.64	2.71	0.52
EA	14.03	2.73	0.66
EB	19.79	3.39	0.76
EC	19.81	3.43	0.79
ED	21.43	3.51	0.85
EE	21.39	3.65	0.86
EF	22.57	3.65	0.90
EG	10.68	2.74	0.62

In addition to these, the amounts of acetaldehyde of stirred style thick yogurt samples (CG and EG coded) are significantly different than set style thick yogurt for all thick yogurt samples (CB-CF and EB-EF coded). This difference can be attributed to production methods.

The acetaldehyde content between 23 - 41 ppm was considered the optimum yogurt flavour by Bottazzi et al (1973). However, Badings and Neeter (1980) reported that good flavour quality yogurt contained 13-16 ppm acetaldehyde. In this study, acetaldehyde concentration never rose above approximately 23 ppm which was for EF coded yogurt sample. It was 18 ppm for (CF), 14 ppm (EA) and 11 ppm (CA) coded sample. Although acetaldehyde is the main volatile, a distinct classification based only on this component seems to be inadequate, e.g. a product with acetaldehyde concentration as low as 8.4 ppm was described as "medium" flavoured.

This fact suggests, in accordance with Groux (1975) and Hamdan et al (1971), that some yogurts low in acetaldehyde still can have a typical aroma and that acetaldehyde is an important aroma compound but not identical with the overall yogurt flavour. Therefore, it can be assumed that the relatively high diacetyl content of this product may have substituted in part the lacking acetaldehyde.

Different acetaldehyde concentrations of yogurts can be found in the literature. Data reported ranged from less than 10 ppm (Bottazzi and Vescovo, 1969) up to 40 ppm (Robinson et al, 1977 ; Bottazzi et al, 1973). Discrepancies may be due to factors such as the use of different starters, synergistic effects of the microflora itself (Bottazzi et al, 1973; Groux, 1973) fermentation conditions (Green and Manning, 1982), proteins used for increasing the total solid content (Estevez et al, 1988; Mistry and Hassan 1992), using UF milk retentate (Biliaderis et al, 1992) and milk from different mammalian species (Yaygin and Mehanna, 1988). Furthermore, the analytical method applied may also be a source of divergent acetaldehyde concentrations (De Haast et al, 1978). Biliaderis et al (1992) reported that yogurt made from UF retentate had slightly higher acetaldehyde than skimmilk yogurt. The most important components required for acetaldehyde production in yogurt are lactose and the amino acids threonine and methionine (Tamime and Robinson 1985). Thus, despite the lower lactose content of UF retentates, acetaldehyde production by starter bacteria was not impaired because acetaldehyde and other carbonyl flavour compounds can be produced by more than one metabolic pathway and various precursors.

b) Acetone

The means of acetone are significantly different at 95 % confidence level between CA and CB-CF coded samples. Similar results were observed for ewe's milk yogurt samples. The means of acetone are significantly different between cow's and ewe's milk yogurt samples at 95 % confidence level. The means of acetone in thick yogurt produced from cow's and ewe's milk by using modified traditional method are not significantly different but significantly different between yogurt produced from

fortified UF retentate and modified-traditional method for all samples at 95 % confidence level. This difference can be attributed to production methods.

The use of concentration ratio of distinct aroma compounds was proposed by Bottazzi and Vescovo (1969) as an aid in differentiating between low and intensively flavoured yogurts. They reported acetaldehyde:acetone ratios of 2.8:1 for products with strong yogurt flavour. This value was exceeded in all samples in this study. This difference is obviously due to the fact that Bottazzi and Vescovo (1969) examined only single strains of lactobacilli, whereas mixed yogurt starters were used in this study. It is well documented that symbiotic effects of the yogurt microflora enhance the formation of typical flavour compounds (Bottazzi 1983; Zourari et al, 1992; Radke-Michell and Sandine, 1984).

c) Diacetyl

Diacetyl was detected in all yogurt samples after overnight storage. Its concentration was between 0.52-0.91 ppm. The lowest value occurred in CG coded sample. The highest value occurred in CF coded sample. There were slight differences between set style thick yogurt (CB-CF and EB-EF coded yogurt). However, these differences are not significantly different at the 99 % confidence level. Different production conditions affected diacetyl amounts in yogurt.

Most investigations on diacetyl production by yogurt starter cultures have been performed with single strains (Tamime and Deeth 1980). It has been suggested that although *S. thermophilus* is the only specie able to produce a significant amounts of diacetyl (Rasic and Kurmann, 1978), Dutta et al (1973) reported high levels produced by *L. bulgaricus*. The presence of diacetyl is thought to contribute to the delicate, full flavour and aroma of yogurt. It is especially important if acetaldehyde is low because it can enhance the yogurt flavour (Groux, 1973).

According to Marshall (1982), the content of diacetyl in yogurt rarely reaches 0.5 ppm. In this study, the results were higher than the results reported by Marshall

(1982). However, even higher levels were reported by Brannoa (1980) and Ulberth and Kneifel (1992) who found 2-3 ppm diacetyl by gas chromatography using head space technique. Bottazzi et al (1971) reported similar amounts of diacetyl (0.4-0.9 ppm). The results obtained in this study are similar to the results reported by Bottazzi et al (1971). Production of diacetyl is conditioned by the seasonal characteristic of milk. In spring, there is often a variation in the manganese content of milk, which is accompanied by a low production of the diacetyl. Bottazzi (1983) reported that the addition of 10 $\mu\text{g/L}$ manganese in milk is advantageous because the starter bacteria are more effectively metabolized. In addition to these, the diacetyl reductase is almost inactive at low pH. The accumulation of diacetyl is slight when the pH is above 5.5 (Bottazzi 1983).

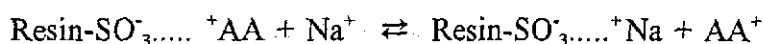
4.3.7 Free amino acids analysis in yogurt

4.3.7.1 The principal of free amino acids analysis

The separation takes place in a column of sulphonated cross-linked polystyrene resin, which is a strong cation exchanger. The matrix of the resin is strongly anionic in nature (SO_3^-) and at the low pH used for separation the positively charged amino acids will be attracted to these negatively charged sulphate groups.

As the pH of the buffer passing through the column is raised, the amino acids will be differentially eluted as their net positive charge is diminished and they are less strongly attracted to the sulphonate site on the resin matrix. They will emerge from the column in a sequence according to their individual pI values, so that at the corresponding pH extra acidic group in their side chain will have very little affinity for the resin and will be the first to emerge from the column, whereas at the same pH value those amino acids whose side chain contains an extra ionizable group are capable of holding on to the resin and will be eluted from the column only as the pH is raised substantially and their net positive charge reduced.

However, it is not only the pH of the eluting buffer which determines the relative elution position of the amino acids but also the cation concentration of the buffer. Na-Citrate buffer solutions are commonly used and the positive sodium ions compete with the positively charged amino acids for the sulphonic acid sites on the resin.



Although the amino acids have a considerable affinity for the resin, the sodium ions are constantly present in a much higher concentration and, as a result, the equilibrium of the above equation is shifted to the right and the amino acids are displaced from the resin. Thus, the molarity of the eluting buffer affects elution and when the ionic concentration of the buffer is increased, the amino acids are eluted more rapidly from the column.

Non-ionic interaction between the amino acids and the resin also influence the elution sequence allowing amino acids with identical pI values to be eluted separately. The polarity of the amino acids is important in this respect and those with a non-polar, hydrophobic R group interact with the strongly hydrophobic resin matrix. The degree of cross-linking also affects separation because it influences the rate of diffusion of the charged species to the exchange sites. Thus an accurate prediction of the position of the amino acids on a chromatogram is difficult and may vary considerably from that determined in practice.

The quantitative resolution of amino acids mixtures is achieved by varying the composition of the buffer flowing through the column by either increasing the pH and maintaining a constant molarity (cation concentration) or by keeping the pH constant but varying the molarity, or by using a combination of both. Other factors such as column temperature, buffer flow rate and type of buffer cation play important, but less significant, parts in the quality of the amino acids separation.

4.3.7.2 Free amino acids content of set and stirred style thick yogurts

The total free amino acids content of yogurt made from different types of milk and different total solids levels showed that the free amino acids spectrum differs significantly among the various yogurts. The total free amino acids content of CA, CB, CC, CD, CE, CF and CG coded samples were 33.08, 62.99, 81.87, 83.95, 82.07, 82.53 and 59.21 mg / 100 g yogurt respectively. The total free amino acids content of 48.26, 69.54, 68.42, 70.03, 86.09, 80.84 and 58.43 mg / 100 g yogurt were found for EA, EB, EC, ED, EE, EF and EG coded samples respectively.

It can be seen from these results that the highest content of total free amino acids was established in yogurt made from whole ewe's milk as compared with whole cow's milk. However, yogurt made from fortified UF milk retentate for both species had similar amount of total free amino acids for equal total solids levels. The increases ranged from 90 % to 154 % for CB-CF coded samples and 78 % for CG coded sample as compared with control yogurt (CA). The ranges were between 41% to 78 % for EB-EF coded samples and 20 % for EG coded sample as compared with control yogurt (EA). The increase of total free amino acids in cow's milk yogurt was two times higher than ewe's milk yogurt as compared with control sample.

The result indicated that the amount of total free amino acids can be affected by the total solid level of the milk, the type of milk, type of starter bacteria and production method of yogurt. When the total solid level was increased in yogurt the amount of total free amino acids was also increased. Similar observation can be seen from the result reported by Nakazawa et al (1991). The amount of total free amino acids was 160.4 mg % for UF concentrate and 117.8 mg % for control yogurt but in this study the amount of total free amino acids is lower than the results were reported by Nakazawa et al (1991), Groux (1972), Luca (1974), Tamime and Deeth (1980) and higher than the results reported by Rasic et al (1971).

Individual free amino acid content of yogurts are given in Fig (4.10), (4.11) Appendix (VI-1-14). It can be seen from the results given for cow's milk yogurt samples that the highest free amino acids were glutamic acid, proline, serin, and aspartic acid, exceeding between 6.29 and 15.49, 4.92 and 12.97, 2.33 and 9.37, 2.23 and 8.84 mg / 100g yogurt respectively and the lowest amount was between 0.147 and 1.57 mg / 100 g yogurt for tryptophan.

It can be observed that the higher amounts of glutamic acid, proline, aspartic acid, serin, lysin and threonine were found in all ewe's milk yogurt samples (EA-EG), at between 9 and 20, 8 and 17, 4 and 11, 2.15 and 7.7, 3 and 6, 2 and 6.7 mg/100 g yogurt respectively. Lower amount of methionine and tryptophan were obtained at between 0.59 and 1.42, 0.119 and 0.9 mg / 100 g yogurt respectively. Yogurt made from ewe's milk contained the highest amounts of glutamic acid, proline and aspartic acid, similar amount of arginine, threonine, serin, lysin and lower amounts of methionine, glycine as compared with yogurt made from cow's milk for all samples (CA-CG and EA-EG coded samples). The amounts of some individual free amino acids are similar to the results reported by Rasic et al (1971) and Tamime and Deeth (1980). The results were lower than the results reported by Groux (1972) and Nakazawa et al (1991). These differences can be attributed raw material, analysis method and starter cultures.

It can be concluded that with the yogurt made from fortified UF milk retentate the total free amino acids increased as compared with whole milk. The total free amino acids content is also different in different types of milks. The total free amino acids ratio is about 1 % of total protein in yogurt. The stirred style thick yogurt (CG and EG coded samples) has lower total free amino acids than its equivalent total solids levels in set style thick yogurt. It can be attributed to the filtration process. During the filtration process 46.3 % and 51.57 % of total free amino acids passed through the filter medium with yogurt whey for cow's and ewe's milk yogurt respectively. Glutamic acid and proline stand out as the two most prominent free amino acids in yogurt from these results.

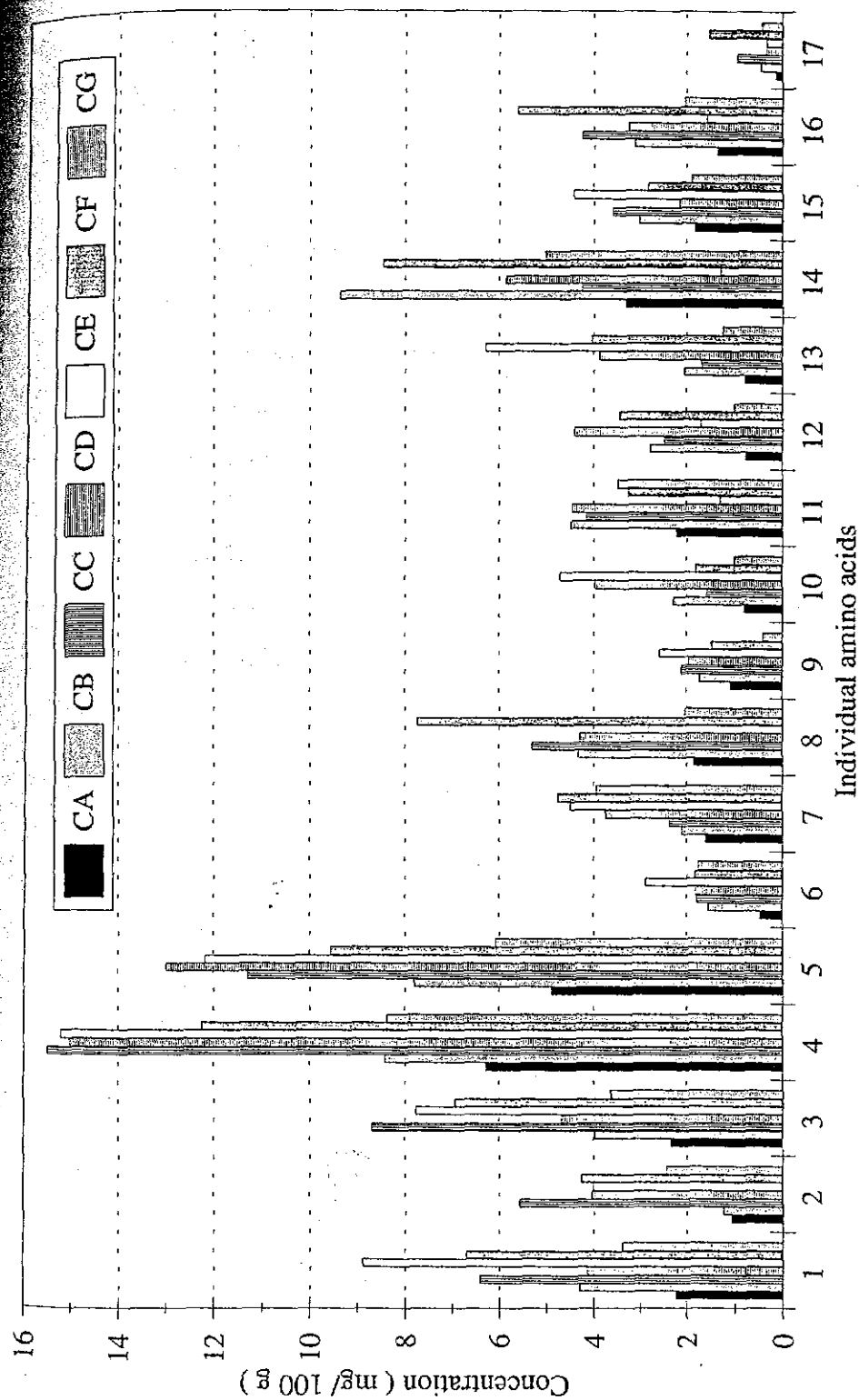


Fig (4. 10) Free amino acid content of set and stirred style thick yogurt produced from cow's milk using different production method. (1-Aspar. acid, 2-Threonine, 3-Serine, 4-Glut. acid, 5-Proline, 6-Glycine, 7-Alanine, 8-Valine, 9-Methionine, 10-Isoleucine, 11-Leucine, 12-Tyrosine, 13-Phenylalanine, 14-Lysine, 15-Histidine, 16-Arginine, 17-Tryptophan)

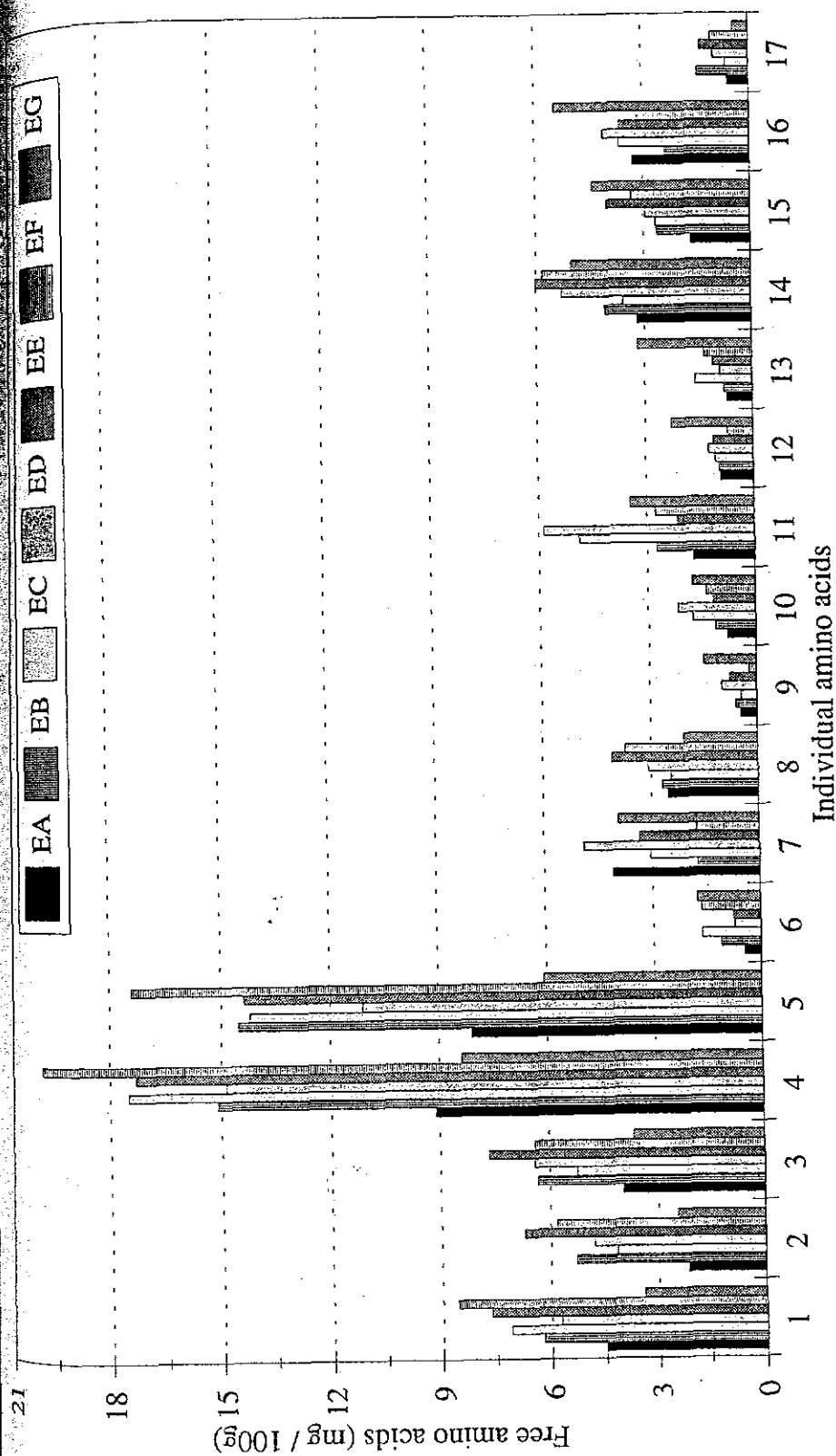


Fig (4.11) Free amino acids content of set and stirred style thick yogurt produced from ewe's milk using different production methods. (1-Aspar. acid, 2-Threonine, 3-Serine, 4-Glut. acid, 5- Proline, 6- Glycine, 7- Alanine, 8- Valine, 9- Methionine, 10- Isoleucine, 11- Leucine, 12- Tryosine, 13- Phenylalanine, 14- Lysine, 15- Hysyidine, 6- Arginine, 17- Tryptophon)

Whereas most other amino acids are consumed to some extent during growth of the bacteria in yogurt production, glutamic acid and proline are not utilized by the yogurt bacteria and hence accumulated in the yogurt. The final content of the other amino acids reflects the balance between proteolysis and consumption.

4.3.8 Microbiological properties of thick yogurt

4.3.8.1 Some microbiological changes in set and stirred style thick yogurt

As previously indicated yogurt samples were stored in a refrigerator at 4 °C, after incubation period, for 24 hours. The enumeration and viability of the yogurt starter cultures in set and stirred style thick and natural yogurts were counted by using LS-differential agar (see section 3.2.5.2.13) as recommended by Eloy and Lacrosse (1976). The enumeration of yogurt starter bacteria on solid media is based on the morphology of the colony and /or selectivity of the morphology of the medium.

The mean values of the *S. thermophilus* and *L. bulgaricus* for CA-CG and EA-EG coded samples are shown in Appendix (VII-1) and (VII-2). The enumeration of the starter bacteria in fresh yogurt samples (CA-CG and EA-EG coded) were between 5.5×10^8 - 2.2×10^9 cfu / g for *S. thermophilus* and 1.9×10^8 - 1.2×10^9 for *L. bulgaricus* for cow's milk yogurt and 5.6×10^8 - 2.3×10^9 cfu / g for *S. thermophilus* and 3.3×10^8 - 1.5×10^9 cfu / g for *L. bulgaricus* for ewe's milk yogurt respectively. The enumeration of starter culture in yogurt prior the production of thick yogurt was 2.3 - 1.4×10^8 cfu / g for *L. bulgaricus* and 4.9 - 6.1×10^8 cfu / g for *S. thermophilus*. The results were slightly lower than the data published by Tamime et al (1991 c). The viable counts of the starter bacteria in stirred style thick yogurt had increased to between 9 - 11×10^8 cfu / g for *L. bulgaricus* and 1.8×10^8 cfu / g for *S. thermophilus*.

These results are in general accordance with earlier findings (Hamann and Marth, 1984). It has been proposed by Hamann and Marth (1984) that the yogurt should contain at least 10^6 cfu/ml viable microorganisms at the time of sale. This value could be accepted as a possible lower limit. The level of total solids in the yogurt milk can influence the amount of starter organisms. Sellar and Babel (1978) noted variations in culture activity observed in different milk used for propagation of starter. Their result showed that with an increasing total solids the viable bacteria count increased. The results reported by Nakazawa et al (1991) support the result reported by Sellar and Babel (1978). However, Tramer (1973) noted that yogurt starter organisms, particularly *L. bulgaricus*, were inhibited above 21 % total solids.

4.8.1.2 Non-lactic acid, coliform, yeast and moulds in yogurt

In the present study, the coliform count of all the yogurt samples tested were absent. These results reflect the hygienic standards and sanitary conditions during the production of yogurt. According to Davis (1975) coliform bacteria is never found in a good yogurt after the first day. A growth of non-lactic acid bacteria, yeast and moulds was observed in stirred style thick yogurt (CG and EG coded sample) which was produced by using modified traditional method. A growth of non-lactic acid bacteria was observed in the majority of stirred style thick yogurt samples (CG and EG coded) but it was not observed for set style thick yogurt. The mean values ranged between 15×10^2 - 25×10^3 cfu/g for total count of non-lactic acid bacteria in CA-CF and EA-EF coded samples. The means were less than 10 cfu/g for yeast and moulds. Limited data are available for microbiological quality of thick yogurt. According to Tamime et al (1987) the limited standard level of total count of non-lactic acid bacteria for yogurt in Switzerland ranged between $25 - 50 \times 10^3$ cfu/g. Barnes et al (1979) have proposed the following standards for yeast and moulds in yogurt:- target <10, acceptable 10 - 50, acceptable with immediate action to improve 50 - 100 and unacceptable > 100 cfu/g respectively. However, if the above two standards are also applied to thick yogurt, the results should be acceptable for the

count of the non-lactic acid bacteria, yeast and moulds. The possible reasons for the presence of non-lactic acid bacteria, yeast and moulds could be attributed to poor hygiene and cross contamination from hand container and process equipments.

4.4 Some chemical, physical and microbiological changes in thick yogurt produced from cow's and ewe's milk during storage period

Shelf life expectations from commercial yogurt vary but generally are, approximately, four to six weeks from the date of production if cold storage is provided during distribution and retail marketing. Under these circumstances yogurt can be stored without a significant loss of quality. Nutritional quality is reasonably preserved at 4-6 °C storage. At the present time, an important factor is the open dating of perishable yogurt is to indicate when the products were packed or when they should be removed from market. Therefore many dairy components need to be rapidly measured to determine the shelf-life of perishable yogurt, *i.e.* the period of time for which a product will remain acceptable in quality to the consumer.

The shelf-life can be expressed by the number of days required to attain a certain bacteria count, unsatisfactory flavour, under specific storage conditions. The growth rate of bacteria during storage at low temperature gives a better index of the shelf-life. The optimum temperature for lipase and protease activity is often lower than the optimum for bacteria growth, and hence, off-flavours may develop even though bacterial counts are low. The flavour rating after different storage period at 7°C is, although subjective, presently the most useful shelf-life test. A slight increase in acidity is noticeable during storage period. Viability of the yogurt culture is also slightly reduced. These changes are relatively minor compared to the changes observed during fermentation.

4.4.1 Changes of titratable acidity and pH of set and stirred style thick yogurt during storage period

Limited information is available on the growth of yogurt starter cultures in UF whole milk retentate. The fermentation of whole milk concentrated to different total solids levels was carried out using yogurt starter cultures. After incubation period all samples were stored in refrigerator at 4°C. The total titratable acidity and pH was measured at 7 day intervals for 42 days. The mean value of total titratable acidity and pH are plotted as storage time versus pH or titratable acidity. They are presented in Fig (4.12) and (4.13) for titratable acidity of cow's milk and ewe's milk yogurt and Fig (4.14) and (4.15) for pH of cow's and ewe's milk yogurts respectively. The raw data are presented in Appendix (III-5) and (III-6) for pH, in Appendix (III-7) and (III-8) for titratable acidity. It can be seen from the graphs that the increases of the total titratable acidity were between 0.37 - 0.26 unit lactic acid % for CA - CF coded samples and 0.37 - 0.49 unit lactic acids % for EA - EF coded samples respectively. Similarly between 0.25 - 0.57 and 0.33 - 0.49 pH unit for cow's and ewe's milk yogurt samples (CA - CG and EA - EG coded samples). This small variation is considered insignificant and probably arises from inherent variation in starter culture activity. During 42 days storage period the maximum total titratable acidity was 2.09 (EF) and 2.05 (CF). These values are still within the range of Turkish Food Act which has a maximum of 2.5 % lactic acid. Considering the whole storage period, relative increases in pH were about 7 % for thick yogurts (CB - CD and EB - ED coded sample), 5-6 % for (CE) and (CF) coded samples and 13 % for control (CA) and 11 % (EA) coded samples. The increases in total titratable acidity were 17 % for CB-CF and 25 % for EB-EF coded samples respectively. The increase of total titratable acidity and pH was 23-25 % of initial level for CA-CF and EA-EF coded samples and 5 % of initial titratable acidity for CG and EG coded samples respectively. In spite of low lactose content in the UF retentate, the concentration of lactic acid was somewhat higher in the yogurt produced from UF retentate than control.

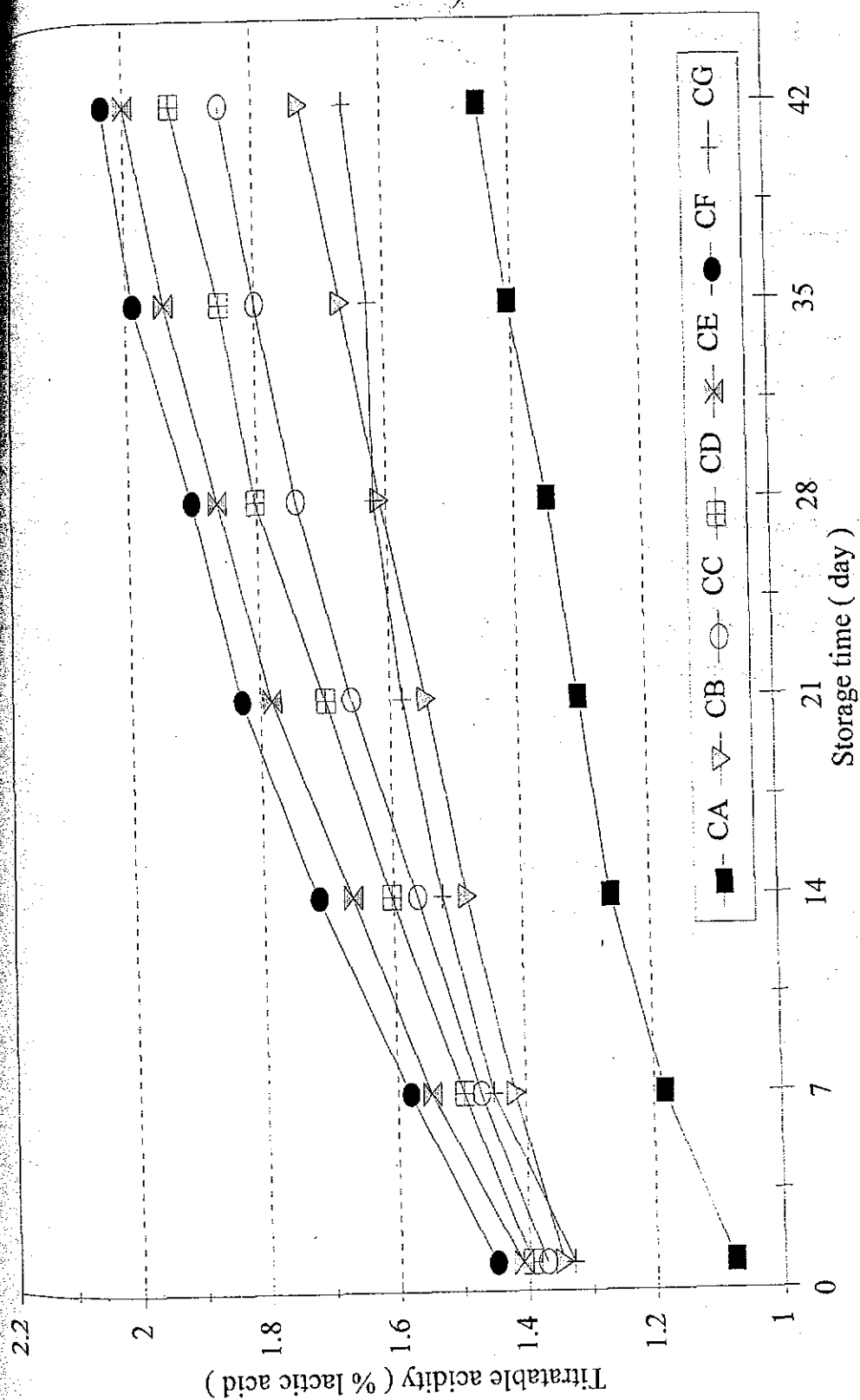


Fig (4.12) Changes of titratable acidity in yogurt produced from cow's milk during storage period

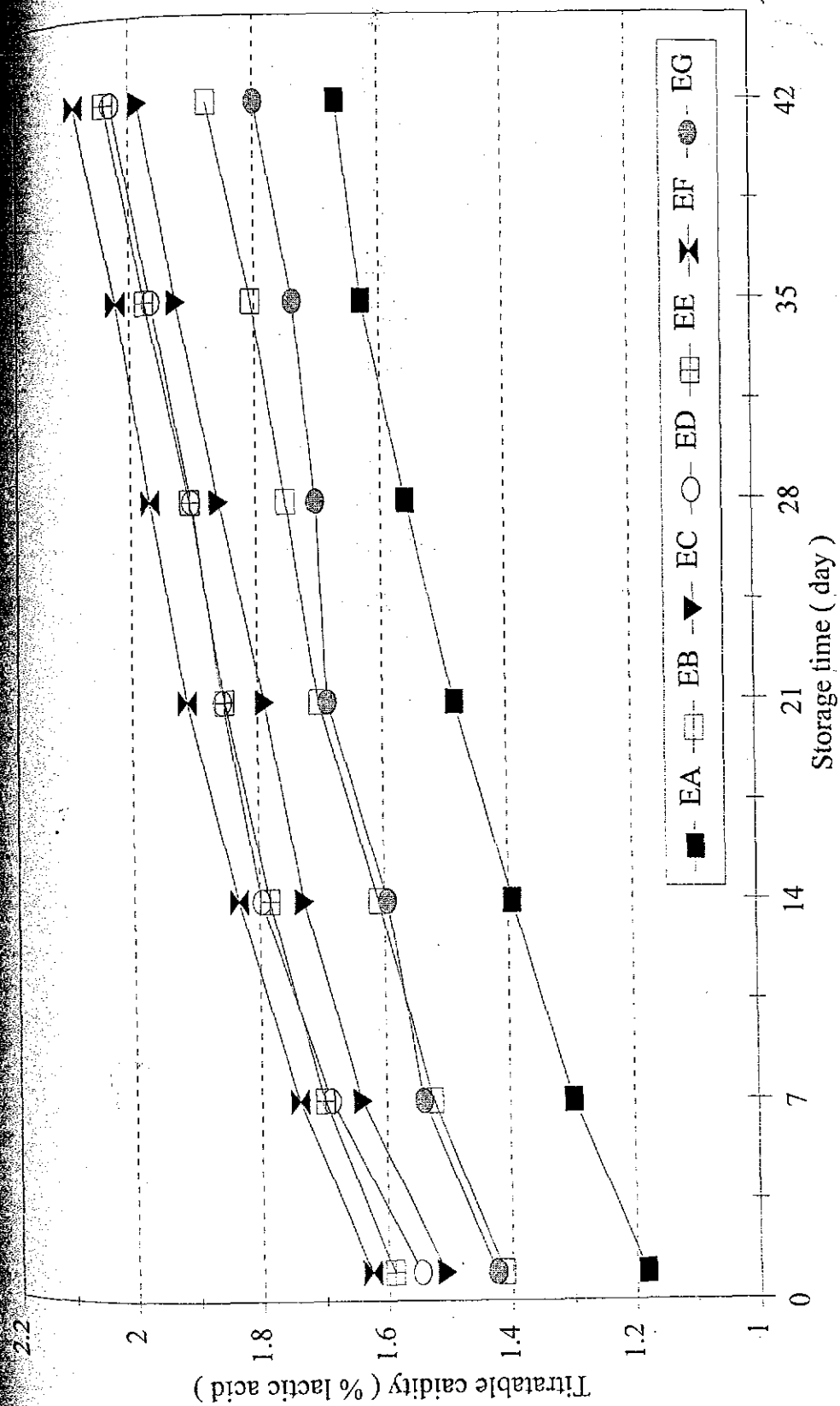


Fig (4.13) Changes of titratable acidity in yogurt produced from ewe's milk during storage period

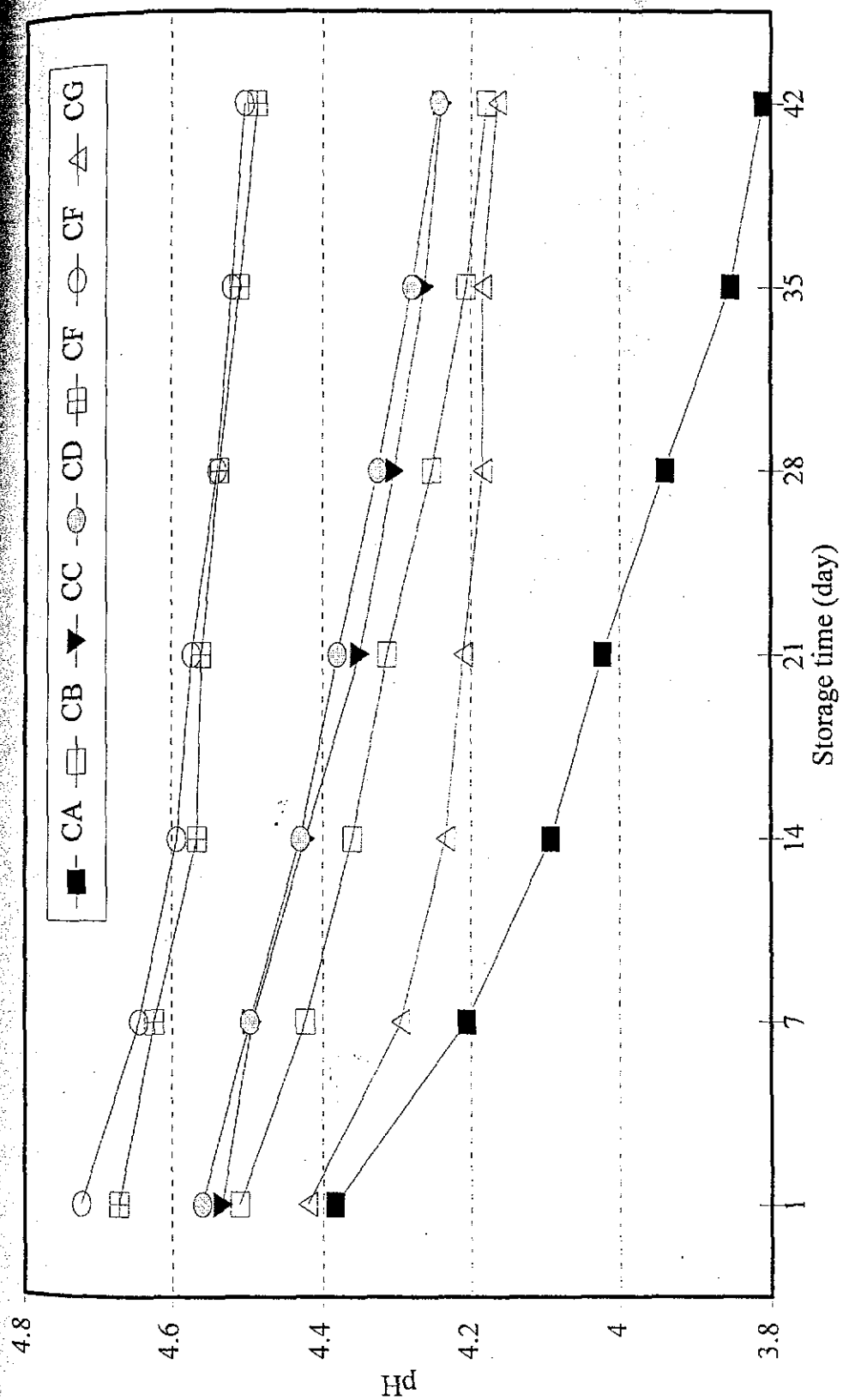


Fig (4. 14) Changes of pH in yogurt produced from cow's milk during the storage period

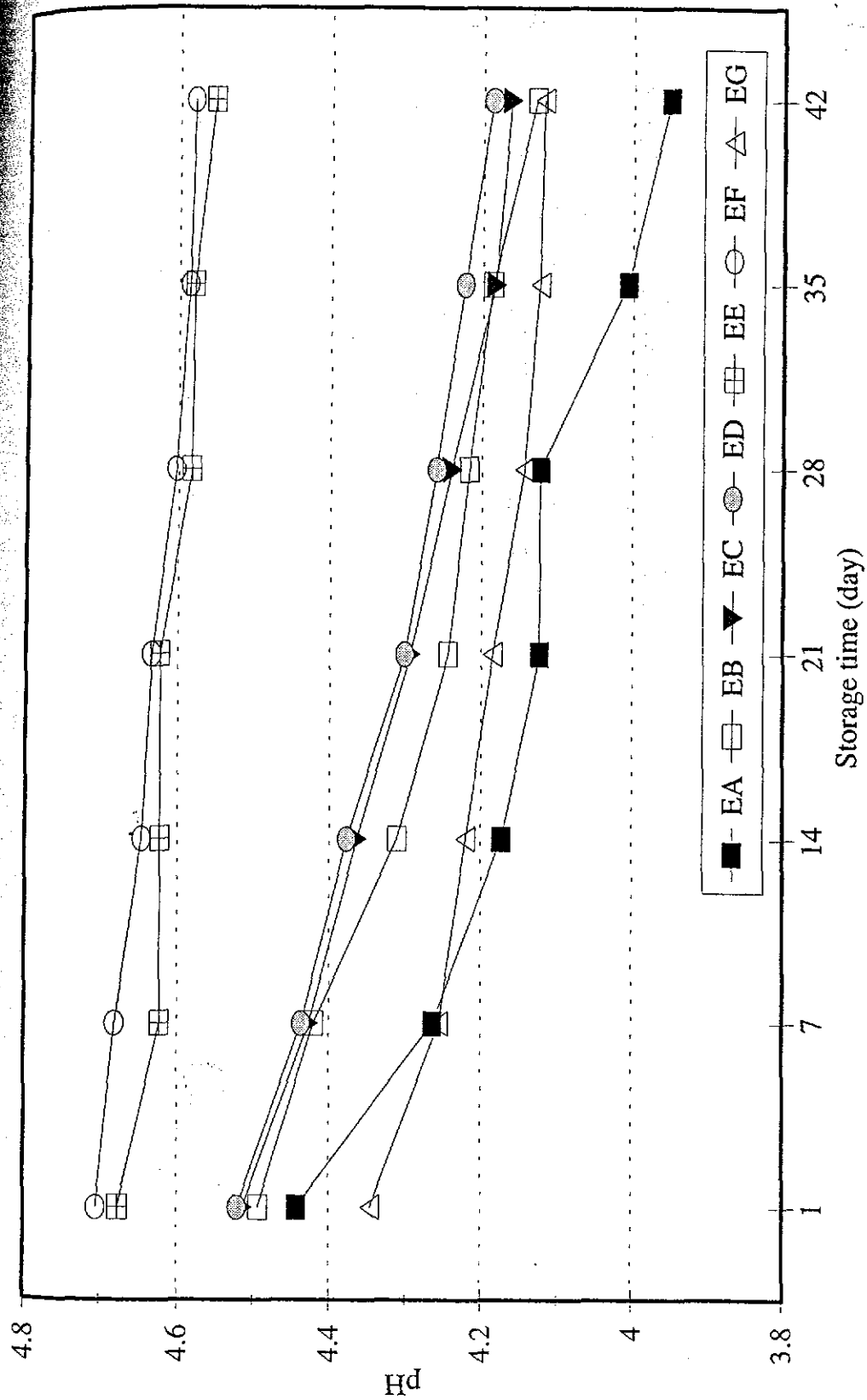


Fig (4. 15) Changes of pH in yogurt production from ewe's milk during storage period

The pH decrease and total titratable acidity increased during storage period of yogurt sample at 4 °C. The pH decrease during storage period was more in the control samples (CA, EA) than in the thick yogurt samples (CB - CF and EB - EF). The tendency of lower post-fermentation acidity in thick yogurt is obvious, this being due to the fact that the higher protein and mineral content of thick yogurt provides a better buffering in the samples. Becker and Puhon (1989) observed lower post-fermentation acidity in yogurt, produced from UF retentate, during 14 days storage at 4°C as compared to yogurt made from evaporated and reconstituted milk. Lactase activity of ewe's milk yogurt is slightly higher than that of cow's milk yogurt. These differences may originate from the lactose content of ewe's milk which is higher than cow's milk.

Kurmann (1968 b) and Fluckinger and Walser (1973) reported and it was quoted by Rasic and Kurmann (1978) that the yogurt samples with high pH at the beginning of storage showed stronger post-acidification than the controls with a lower pH. However, after longer interval of storage, pH values of both yogurt were equalized. The acidification rate of yogurt bacteria for post-fermentation depends upon the ability of starter bacteria, the rate of fermentation in bulk or in retail containers, cooling, storage temperature and the initial pH value. In 42 days storage period acidity level of samples were not equalized in this study. The acidity level was still low. The acid production of the streptococci stops at a pH of 3.9 - 4.3 and of the lactobacilli at a pH of 3.5 - 3.8. Therefore, the lactobacilli are the main contributor to acidification at pH value below 4.0 but at a pH above 4.0 both lactobacilli and streptococci contribute to post-acidification of yogurt (Rasic and Kurmann 1978). *S. thermophilus* tolerates up to 0.8 % lactic acid, whereas *L. bulgaricus* tolerate up to 1.7 % lactic acids. Beyond these levels, lactic acid inhibits the growth of bacteria. In addition to this, cooling of yogurt is an efficient method of stopping the growth of starter bacteria (Bottazzi 1983).

Parnell-Clunies et al (1986) produced yogurt using different heat treatment and stored for 42 days. The result was that pH was reduced after 42 days storage from 4.16 to 3.89 - 4.12. It was reported that the two means are not significantly different

99 % confidence level. Abrahamsen and Holmen (1980) compared quality of yogurt produced from processed milk with UF retentate at 14 % total solid. The results reported were that during storage period the pH decreased from 4.06 to 4.16 for UF retentate yogurt and 4.05 to 3.94 for control. Lactic acid % was 1.37 to 1.53 for UF retentate yogurt and 1.01 to 1.16 % lactic acid for control.

Poppino et al (1978) reported that yogurt made from 2.5:1 concentrated UF retentate and diluted milk showed no increase in acidity above 1.1 % lactic acid after seven days or longer storage. The acidity and pH changes in labneh produced from fortified retentate was reported by Hofi (1990) in 20 days storage period at 4 °C. The results ranged from 1.92 to 2.23 % total titratable acidity and pH ranged from 4.4 to 3.9 at the end of storage period respectively. These results do not agree with the results obtained in this study.

It can be concluded that the titratable acidity increased during storage period and pH decreased for all samples. However, these results showed that above 28 % total solids level increases of titratable acidity is lower than for other total solids levels. The titratable acidity in high total solids set style thick yogurt generally has slow post acidification because of buffer capacity of yogurt. As a result of this, set style thick yogurt, produced from fortified UF retentate can be stored for a longer time than natural yogurt if it is produced under hygienic conditions.

4.4.2 Changes of water holding capacity of set and stirred style thick yogurt during storage period

The water holding capacity of different yogurt during 42 days storage period are presented in Fig (4.16) for cow's milk yogurts and Fig (4.17) for ewe's milk yogurts. The raw data are given in Appendix (IV-1) and (IV-2) for cow's and ewe's milk yogurt samples. The water holding capacity was plotted versus storage time (days). These show a slight decrease for all samples. The two mean values are not

significantly different at the 95 % confidence level. It may originate from total titratable acidity increases during the storage period in samples. The WHC decreased 12 % to 22.2 % for CB - CG coded samples and 10.4 % for CA coded sample. The results for EB - EG coded samples were 17.5 % to 28.5 % . This can be attributed to starter activity in different types of milk. The results were slightly different between cow's and ewe's milk yogurt samples.

Parnel-Clunies et al (1986) reported that in the storage period, the water holding capacity of set-style natural yogurt was not significantly affected. In this study, the results confirm the results reported by them. It has been shown in other studies that yogurts prepared with casein-supplemented milk had low whey separation because of water binding by casein (Modler and Kaláb 1983; Modler et al 1983). Kehagias et al (1979) have found that the whey separation was reduced during cold storage. Kessler (1981) reported that the properties of yogurt change during storage period. It was found that the coagulum showed the greatest tendency to syneresis after about 8 days storage. When the product is stored for a long periods of time, particularly at higher temperatures and when the initial pH was low, the product becomes too acidic. It has been stated that extensive proteolysis can cause changes in the yogurt microstructure leading to syneresis. This is accompanied by the appearance of a slightly higher whey separation from sample. Becker and Puhan (1989) reported that during storage period water holding capacity slightly increased. The level is depend on solid non-fat contents.

It can be concluded that the water holding capacity slightly decreased during six week storage time, but the differences are not significant. The results confirm to the results reported by Parnel-Clunies et al (1986). Water holding capacity also depends on some other criteria *i.e.* protein level, storage temperature, proteolytic activity of starter culture.

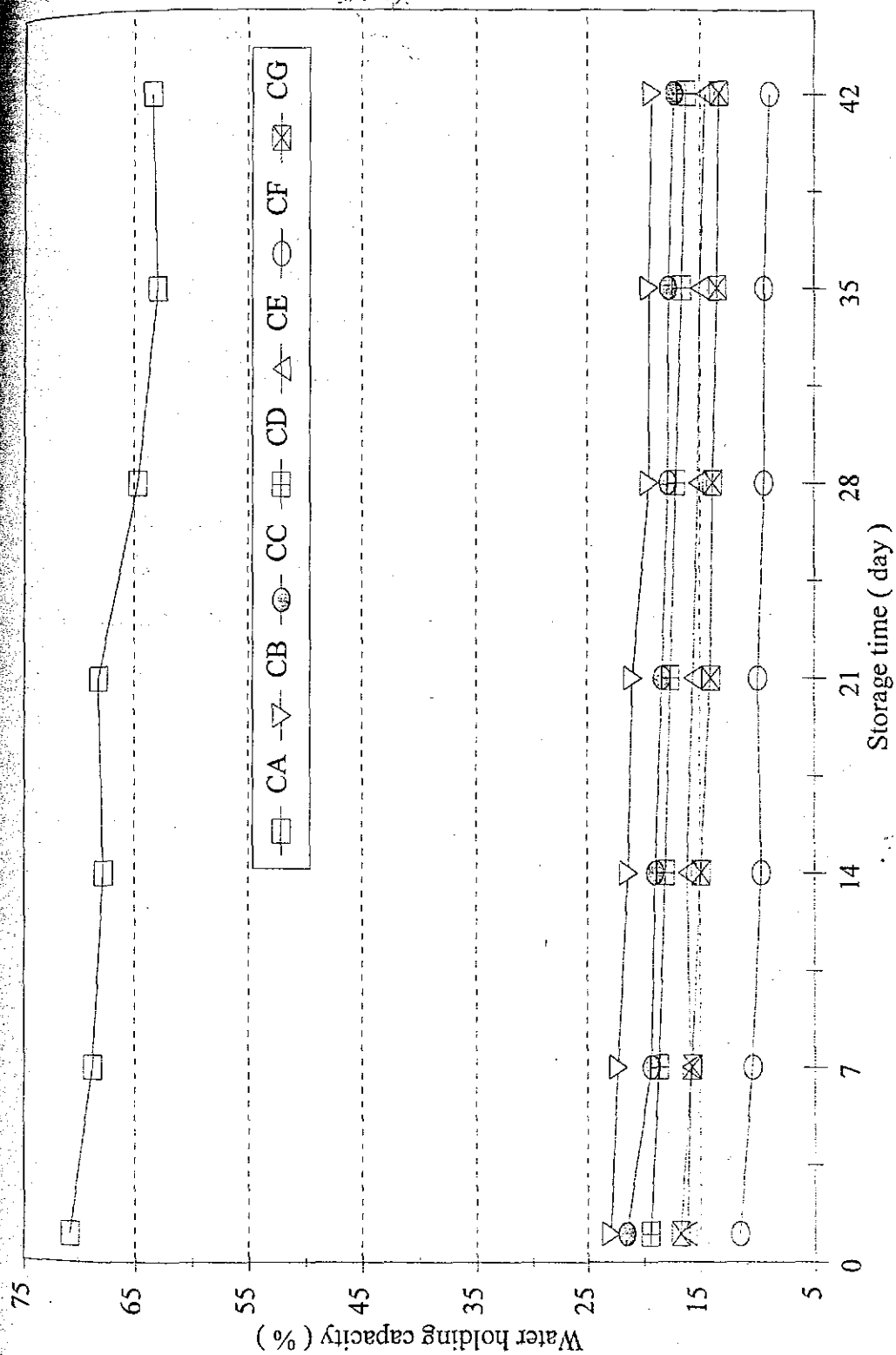


Fig (4.16) The changes of water holding capacity of set and stirred style thick yogurt produced from cow's milk during storage period

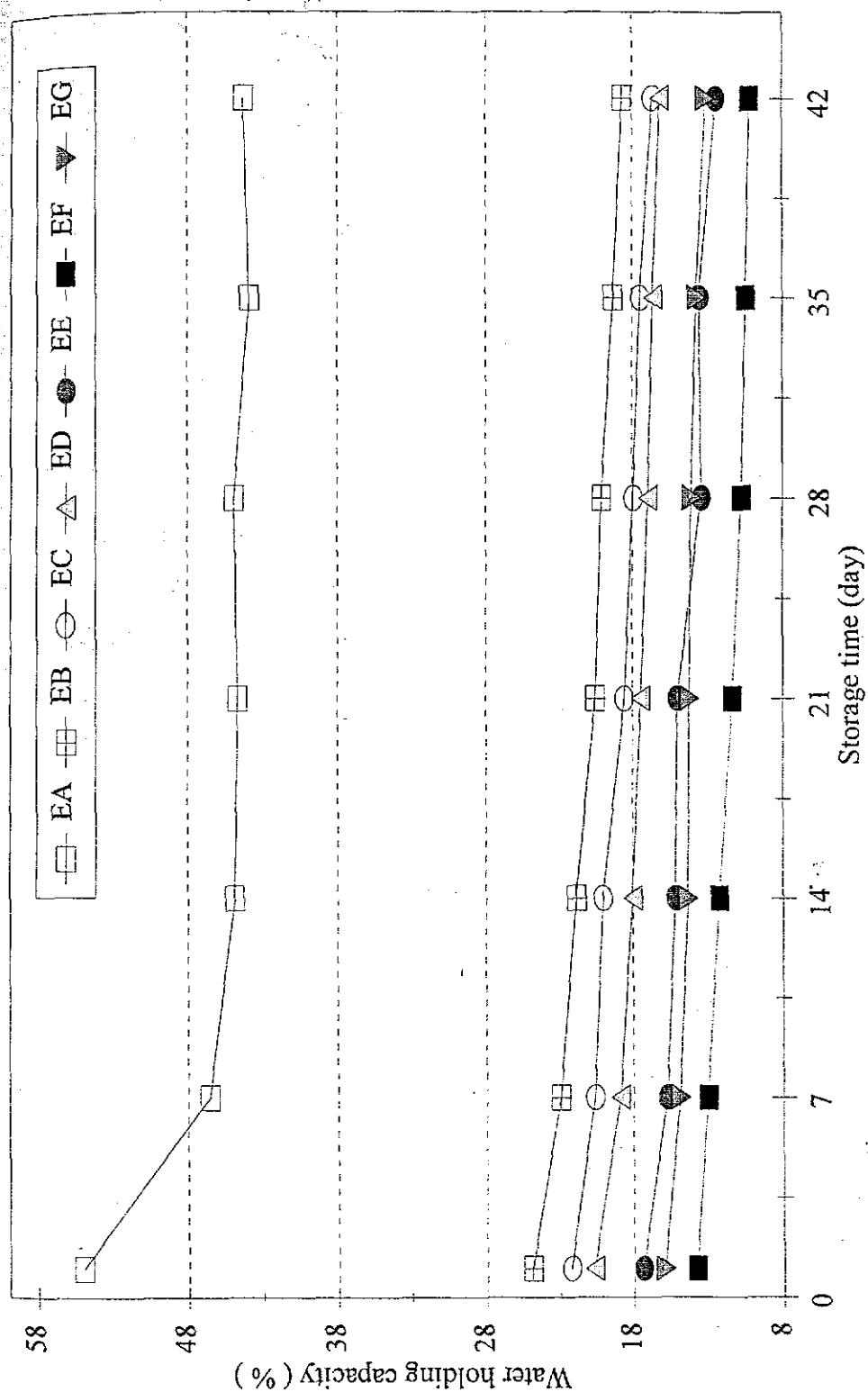


Fig (4.17) The changes of water holding capacity of set and stirred style thick yogurt produced from ewe's milk during storage period

4.4.3 Changes of firmness of set and stirred style thick yogurt during storage period

The firmness of various set and stirred style thick yogurt was measured by using the universal penetrometer over 42 days storage at seven days intervals at 4°C. The results are presented in Fig (4.18) and (4.19) for cow's and ewe's milk yogurt samples respectively and raw data are presented in Appendix (IV-3) and (IV-4). During the storage period, the firmness increased slightly for all yogurt samples.

As it can be seen from the results that between the beginning of storage and the end of storage, the firmness was slightly increased. However, the two means are not significantly different at the 95 % confidence level calculated with *paired-t* test. Similar statistical results were found for ewe's milk yogurt samples. The mean of control samples (CA and EA) are significantly different. But between set style thick yogurt samples, the source of milk (cow's or ewe's milk) does not effect the firmness significantly at similar total solid level.

The physical characteristics as a whole reach their optimum in 24-40 hours after the end of the lactic acid fermentation. As time passes the characteristics of the product tend to deteriorate owing to the action of enzymes coming from the lactic acid bacteria in yogurt. The firmness of coagulum depends on the end pH and starter culture selection (Loones, 1989). The decrease in pH affects the state of suspension of calcium. Below a pH of 5.0 ionic calcium is released from colloidal calcium phosphate to give a soluble form (Rajput et al 1983). The depth of penetration of the probe can be used as a significant parameter (Becker and Puhān 1989). A large variety of probes, penetration rates, penetration depths and temperatures have been used (Tamime 1977; Kessler 1981; Becker and Puhān 1989). Consequently, it is hardly possible to compare results. Parnell-Clunes (1986) reported that although there were differences between firmness at day 1 and 42, this difference was not significant for any samples, prepared using different treatments.

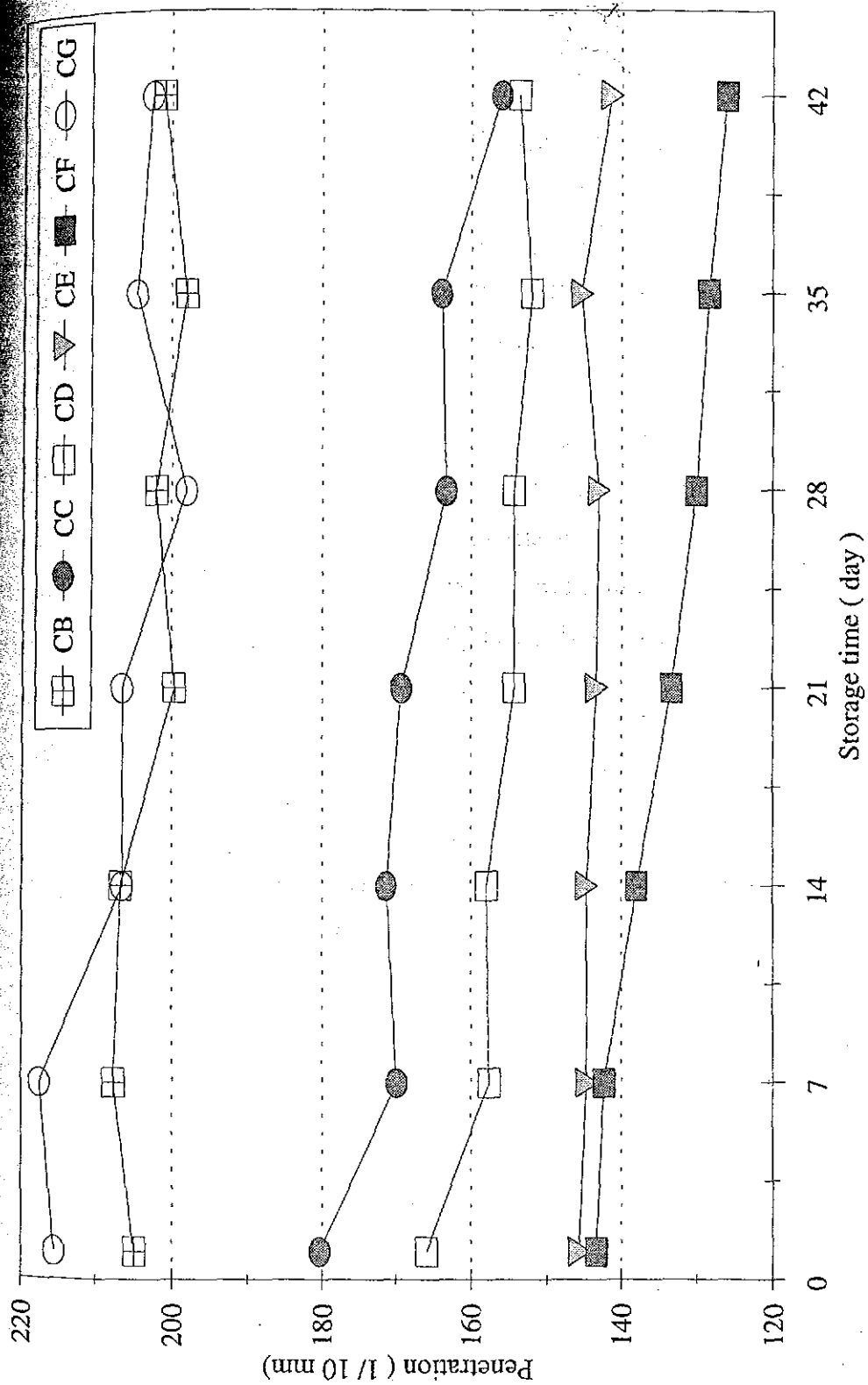


Fig (4.18) The changes of firmness in set and stirred style yogurt made from cow's milk by using different production method during storage period

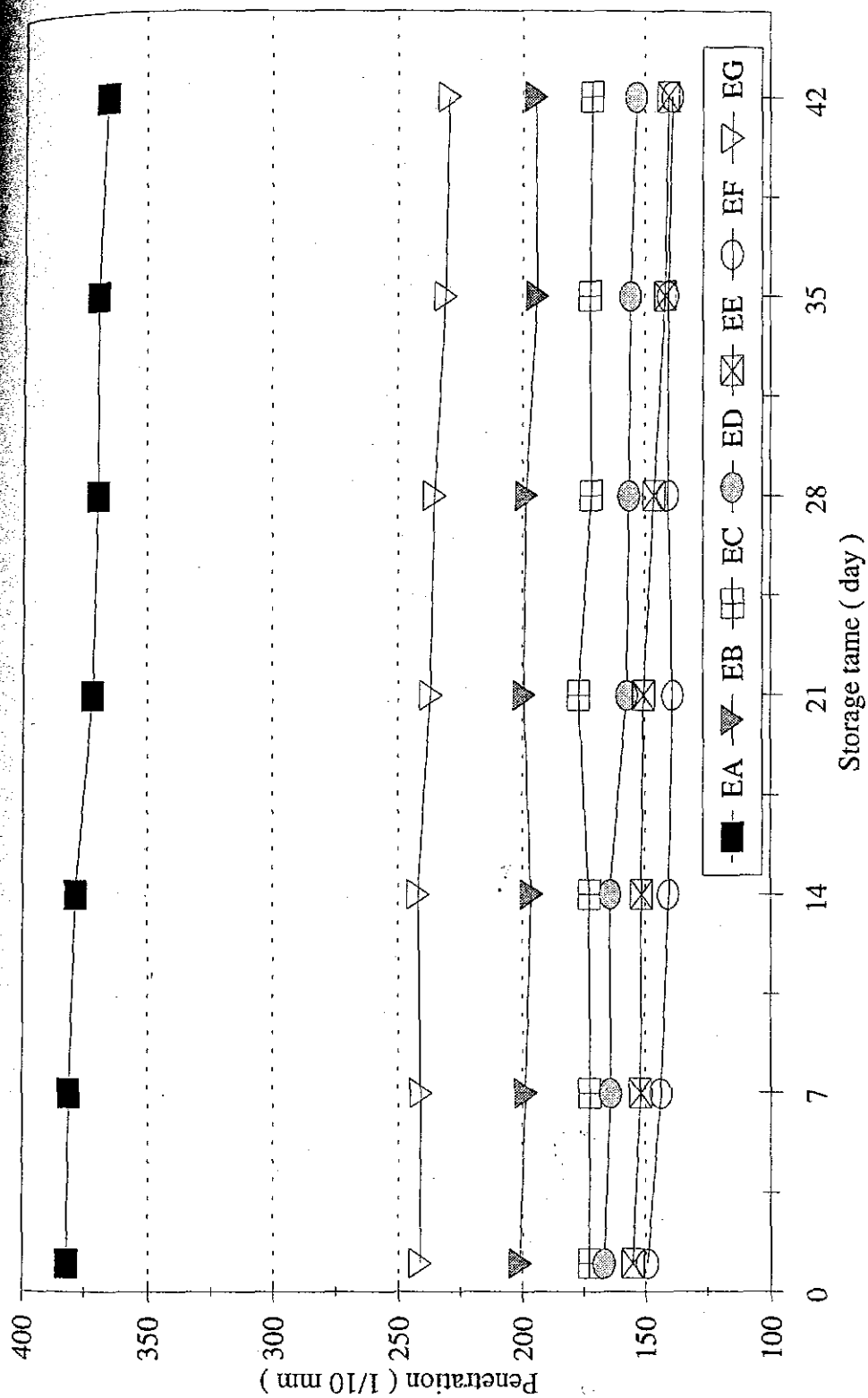


Fig (4.19) The changes of firmness in set and stirred style thick yogurt made from ewe's milk by using different production procedure during storage period

Mistry and Hassan (1992) reported that the control yogurt was softest and firmness of yogurt increased as protein content increased. Becker and Puhon (1989) noted that the effect of fat level on firmness in yogurt is very important. Kehagian et al (1984) have found that the penetration force remained practically constant during 10 days storage period.

It can be concluded that the firmness increased when total solids levels increased. During six weeks storage period firmness are slightly increased in all samples but the differences were not significant.

4.4.4 Changes of some volatile flavour compounds of set and stirred style thick yogurt during storage period

Acetaldehyde is produced during the production of yogurt in more significant amounts than any other volatile compounds. The rate of acetaldehyde production is very dependent on acidity level which means that at the titratable acidity of 0.90 % lactic acid obtained during the manufacture of yogurt a sufficient amount of aroma substance may be produced to contribute to the typical flavour of yogurt. Strains active in acetaldehyde production but capable to reduce it later, particularly during cold storage of yogurt, are considered to be of a lesser value in preparing the culture. Fig (4.20) for cow's milk and Fig (4.21) for ewe's milk yogurts show changes of acetaldehyde during storage period. The raw data are presented in Appendix (V-1) and (V-2) for cow's and ewe's milk yogurt samples. Acetaldehyde decreased between 17-30 % and 23 - 32 % of initial amount for cow's and ewe's milk thick yogurt at the end of 42 days storage period respectively. According to Goerner et al. (1972) a decreases of acetaldehyde content occurs during storage of yogurt. The acetaldehyde content also decreases at acidity level above 1.14 % lactic acid. In contrast to this, Vanderporten and Waes (1972) did not find any significant changes in the content of acetaldehyde during storage period of yogurt for 5 days.

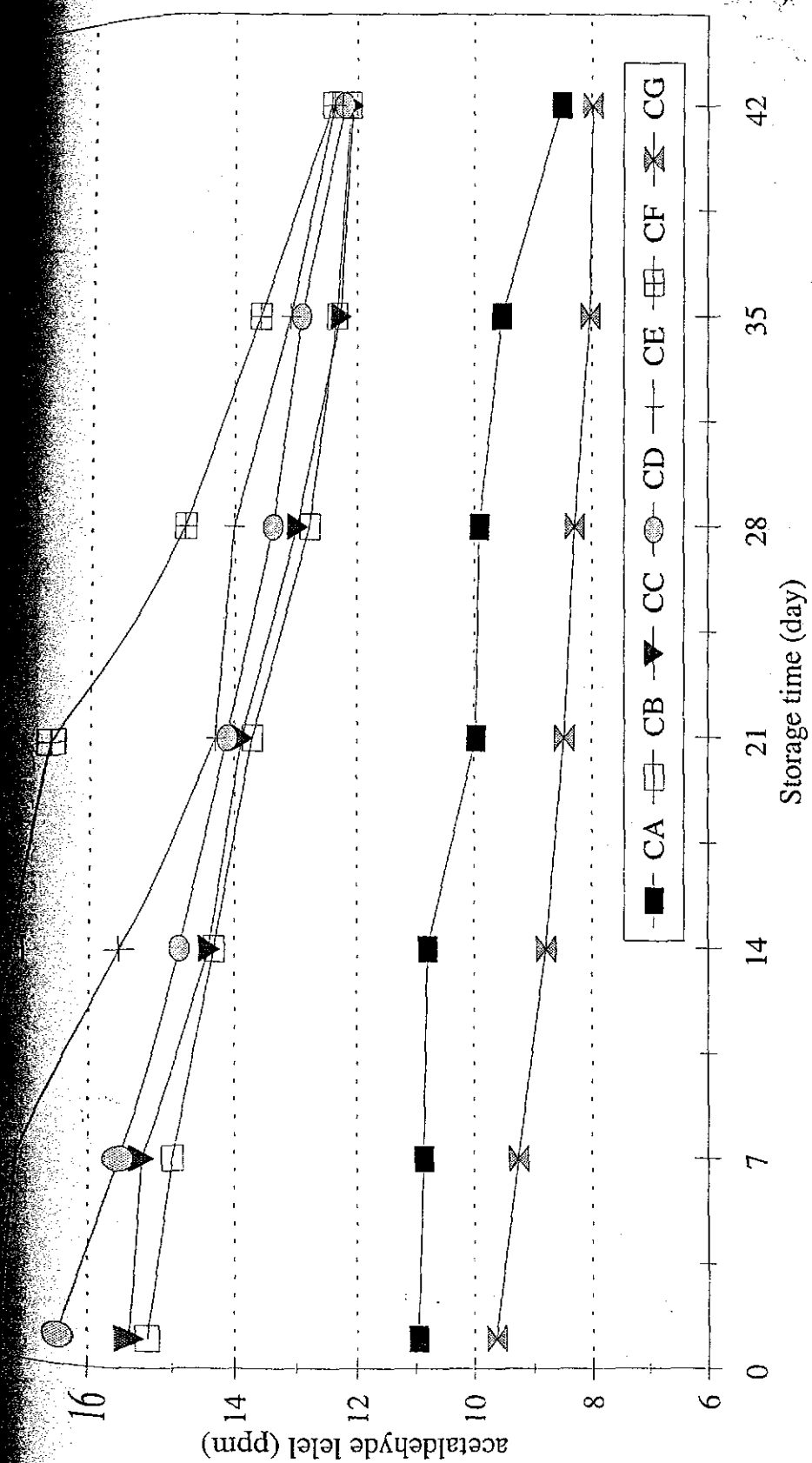


Fig (4.20) The changes of acetaldehyde level in yogurt produced from cow's milk during storage

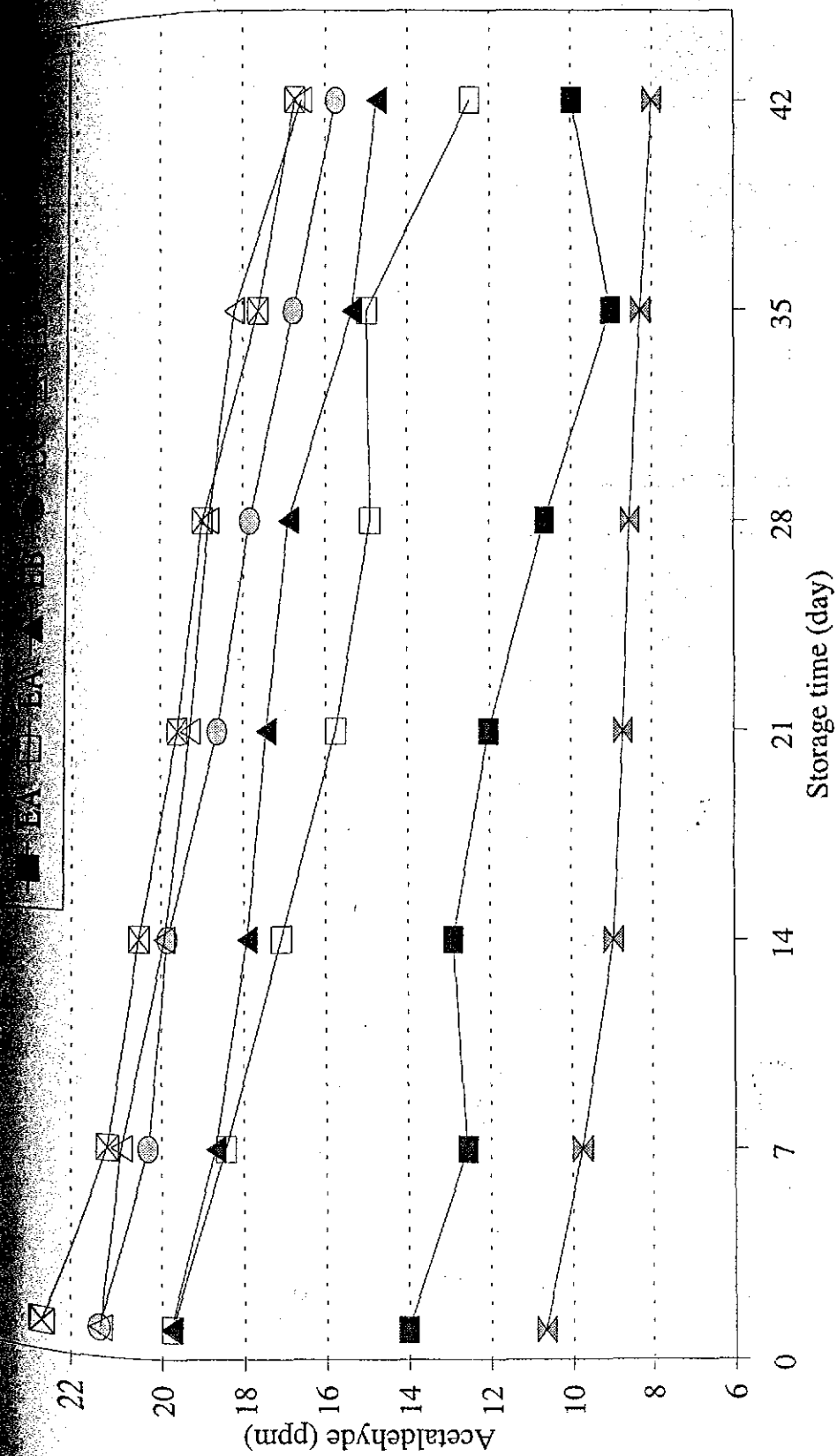


Fig (4.21) The changes of acetaldehyde level in yogurt produced from ewe's milk during storage period

Nevertheless, Hamdan et al. (1971) established that of three yogurts made with different starter cultures, two showed a decrease of acetaldehyde content during storage for two weeks while one retain constant values.

Fig (4.22) and Fig (4.23) for cow, ewe's milk yogurts show changes of acetone level during storage period. The raw data are presented in Appendix (V-3) and (V-4) for cow's and ewe's milk yogurt samples. Acetone level decreased 16 - 20 % for cow's milk yogurts (CA - CG coded samples) and 8 - 12 % for set style thick yogurt made from ewe's milk (EB-EF coded samples) respectively. It was 8 - 13 % for set style thick yogurt 14 % for control and 17 % for stirred style thick yogurt respectively. Diacetyl level of different yogurt are presented in Fig (4.24) and (4.25). The raw data are presented in Appendix (V-5) and (V-6) for cow's and ewe's milk yogurt samples. Diacetyl level decreased between 17.5 - 20 % and 17 - 23 % for cow's and ewe's milk control and thick yogurt (CA-CG and EA - EG coded sample) respectively. The decreases are slightly different between cow's and ewe's milk yogurts.

According to Gaffar (1992) acetone disappeared in natural commercial yogurt within first week of storage, whereas acetaldehyde and diacetyl declined steadily but were still present after two weeks storage. Kang et al (1988) studied flavour changes during ten days storage. It was reported that acetaldehyde increased after 10 days storage and acetone remained at almost the same level. Diacetyl increased first then decreased slightly at the end of the storage period. Mistry and Hassan (1992) reported that the high protein content of yogurts might be responsible for the high acetaldehyde content of the yogurts which range from approximately 33 to 57 ppm.

It can be concluded that all volatile aroma compounds analyzed decreased during storage period. Similar results were reported in literature.

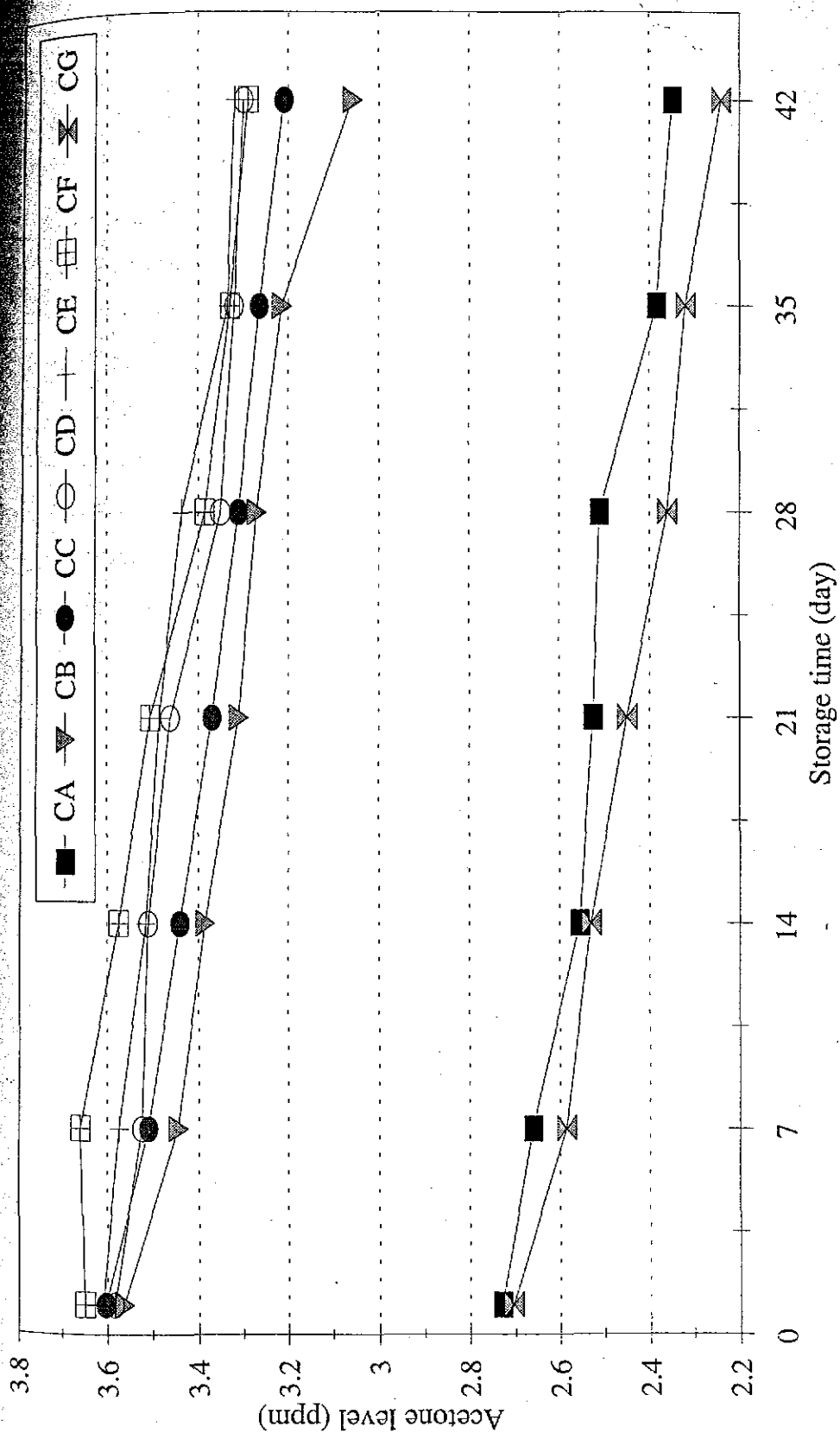


Fig (4.22) The changes of acetone level in yogurt produced from cow's milk during storage period

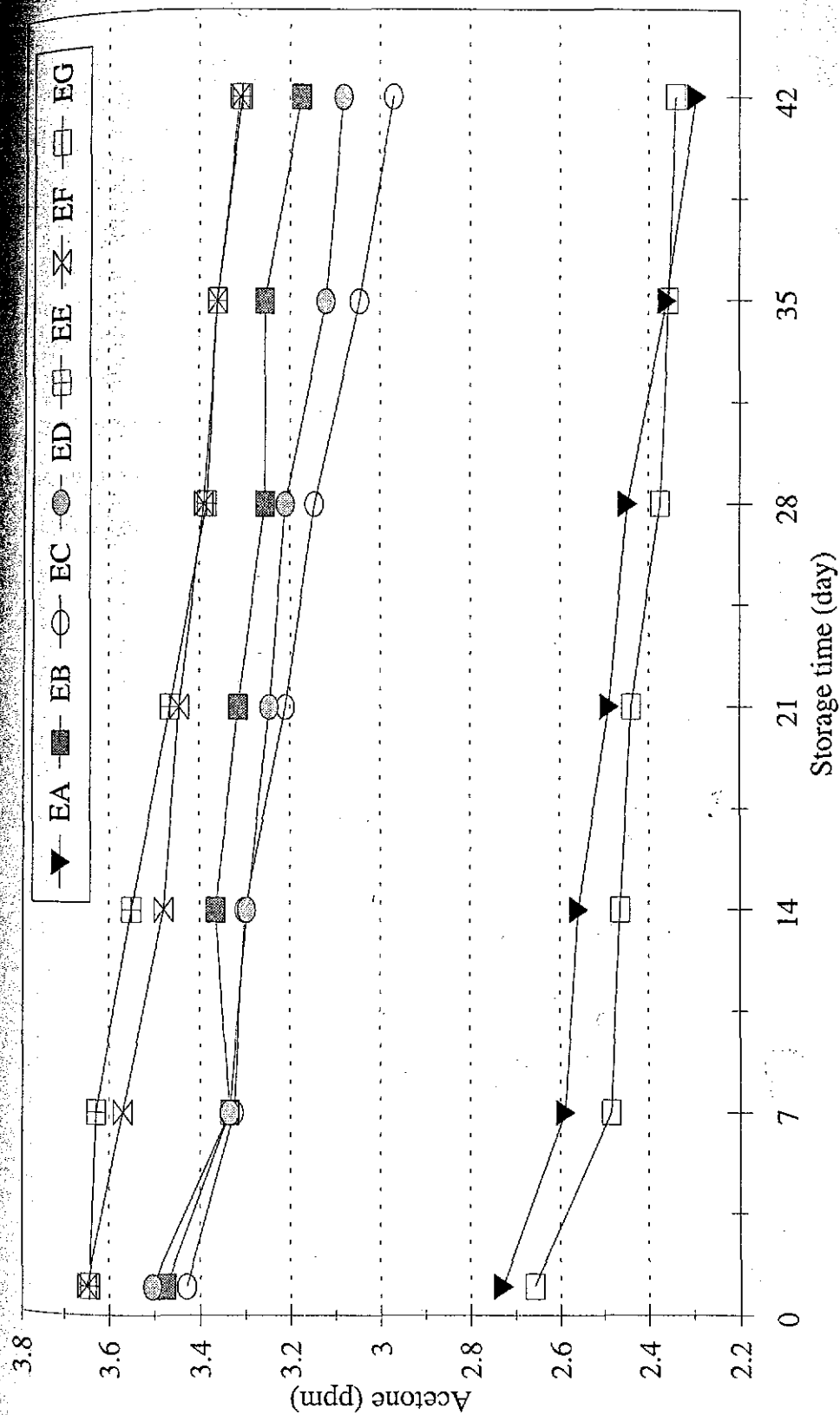


Fig (4. 23)The changes of acetone level in yogurt produced from ewe's milk during storage period

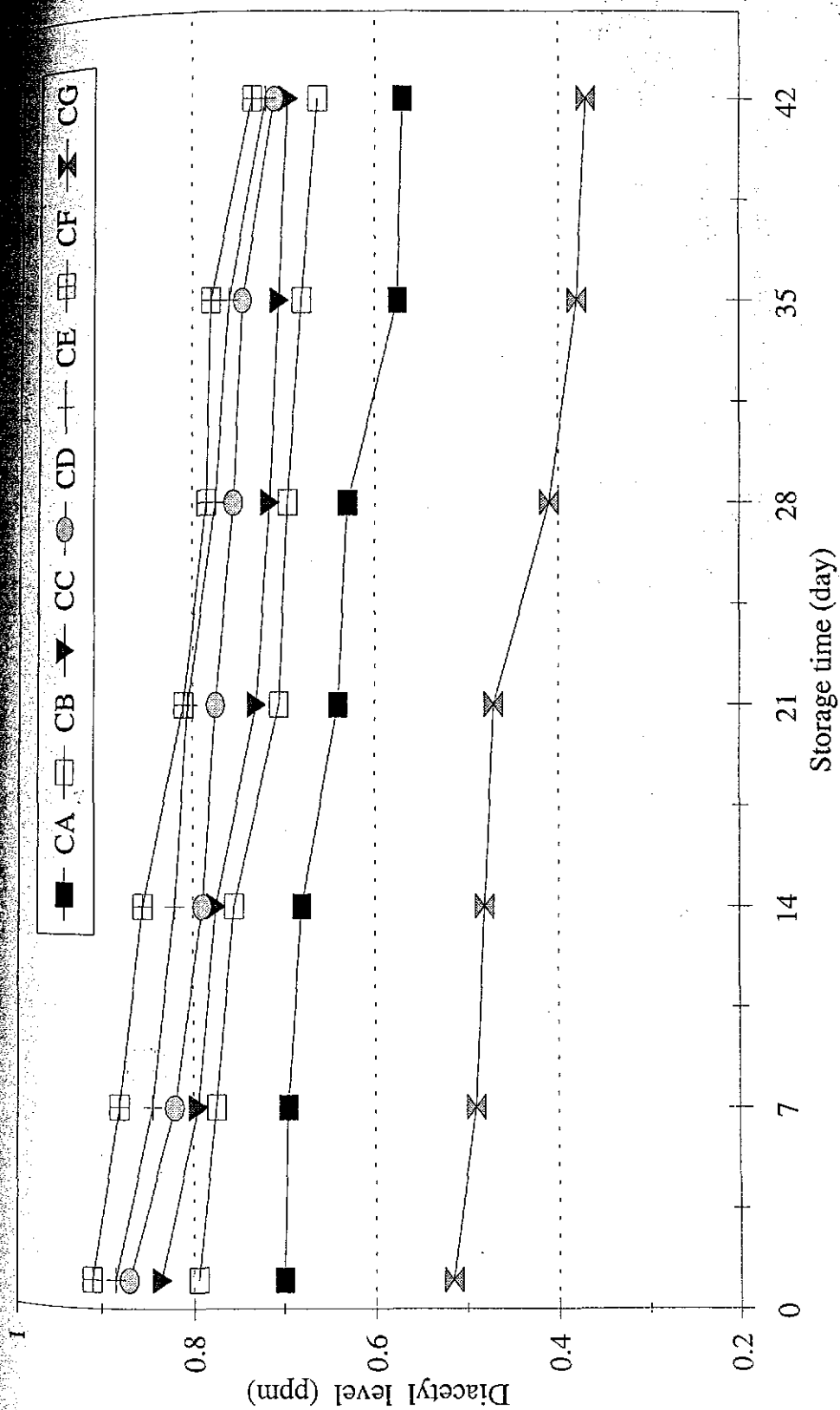


Fig (4. 24) The changes of diacetyl level in yogurt produced from cow's milk during storage period

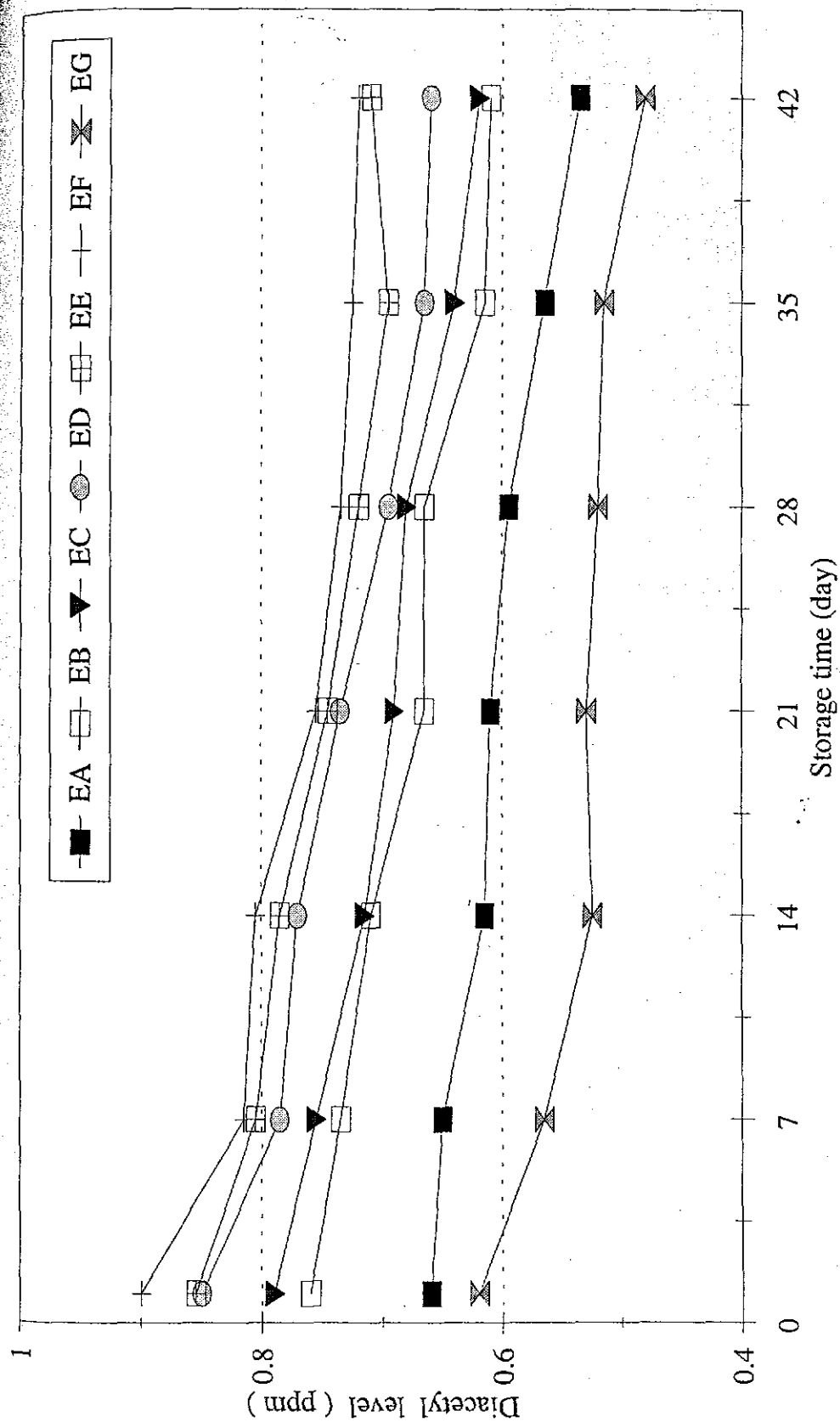


Fig (4. 25) The changes of diacetyl level in yogurt produced from ewe's milk during storage period

4.4.5 Changes of the free amino acids content of set and stirred style thick yogurt during storage period

There is limited data available on the free amino acids content of set and stirred style thick yogurts during storage time. The changes of total free amino acids content of yogurt samples during storage period are presented in Table (4.10) and individual free amino acids changes are presented in appendix (VI - 1- 14) for all type of yogurt samples made from cow's and ewe's milk. The free amino acids content of all yogurt samples increased during six weeks storage period at 4 °C. The changes were higher in set style thick yogurts (CB - CF and EB - EF coded samples) than in the controls (CA and EA coded samples) and stirred style yogurts (CG and EG coded samples). The increases were slightly higher in ewe's milk yogurt samples than in the cow's milk yogurt samples.

The smallest increases were observed in stirred style thick yogurts (CG and EG). The increase is about 30 % of the free amino acids of the first day level for control samples, 40 % for set style thick yogurt samples and 5-11 % for stirred style thick yogurt samples.

The individual free amino acids, proline and glutamic acid, were increased by considerable amounts during storage period because they were not used by the starter bacteria. All other individual free amino acids increased by small amounts. The changes originate from bacterial or enzymatic activity. The increases were not constant during storage period that is why it is difficult to make definite statement to explain these changes. The proteolysis is continuous during yogurt storage at 7°C. A considerable amount of peptidase appear in the yogurts after 24 hours and they continue to grow in the yogurts for 21 days.

Table (4.10) The changes of total free amino acids amounts during storage period (mg/100 ml)

Product Cods	Storage time (day)						
	1	7	14	21	28	35	42
CA	33.080	36.230	45.640	45.720	46.410	45.640	46.940
CB	62.990	77.790	85.070	86.620	89.110	90.660	92.130
CC	81.970	104.620	110.170	113.720	114.080	115.040	113.130
CD	80.950	103.210	111.340	114.510	116.660	117.660	118.730
CE	82.070	97.430	105.630	107.960	109.570	108.400	108.210
CF	86.530	103.710	108.560	108.010	110.590	110.940	115.660
CG	48.200	50.110	52.430	57.560	60.780	57.670	58.610
EA	48.260	70.560	71.840	70.210	70.510	66.370	60.980
EB	69.540	82.410	77.450	82.110	77.520	79.800	85.390
EC	68.420	79.300	83.120	87.890	83.440	93.340	92.490
ED	78.030	88.140	97.170	102.810	103.990	107.780	114.580
EE	83.090	100.600	105.240	110.930	115.300	118.360	118.490
EF	84.840	87.750	100.800	109.350	114.050	114.750	119.830
EG	59.214	64.295	65.491	73.979	74.029	62.714	62.459

The enzymes remain active during storage. The data compiled by Tamime and Deeth (1980) indicate that the profile of amino acids in milk and yogurt reported in the literature show many differences which are ascribed to origin of the milk, variability of the strains, rate of inoculation and starter bacteria and condition of storage. A great degree of cooling, to between 0 and 4 °C, slows down the changes in the substrate even more, though it does not stop them completely. Under these condition proteolysis is also slowed down considerably.

The "ready for use" yogurt has an increased amount of all free amino acids as compared with milk, particularly proline. The content of free amino acids increases with extended storage of yogurt. The high proteolytic action of *L. bulgaricus* gives rise to considerable increase of all amino acids, whereas *S. thermophilus* particularly during the first phases of growth, consumes free amino acids and only yields an increase of proline and alanine.

The temperature of storage of yogurt can affect the level of free amino acids in the product, i.e. the higher the storage temperature, the greater the increase in the free amino acids. Ottogalli et al. (1974) stored full and low fat natural yogurt at 4 and 20 °C for 60 days. The increases in the level of amino acids in these yogurt were at 4°C) 2.36 and 1.0, and (at 20°C) 7.57 and 14.65 mg/ 100 ml respectively. The amino acids content is also dependent on the titratable acidity of the product and method of production (Tamime and Robinson 1985).

It can be concluded that the total and individual free amino acids increased slightly in all yogurt samples because bacterial and enzymatic activity carried on slowly during storage. The free amino acids in set style thick yogurts were slightly higher than control and stirred style thick yogurts. This can be attributed to available protein in yogurt samples.

4.4.6 Changes of microbiological properties of set and stirred style thick yogurt during storage period

Yogurt is special in that it contain, up to the moment of consumption, large number of a specific, living yogurt bacteria. Microbiological analysis are expected to determine the content of specific fermentation agents and contaminant microorganisms. The means of the *S. thermophilus* and *L. bulgaricus* for yogurt samples are presented in Table (4.11) for cow's milk and in Table (4.12) for ewe's milk yogurts. Raw data are also presented in Appendix (VII-1) and (VII-2) for

cow's and ewe's milk yogurt samples. The analysis was carried out over 42 days at 7 days intervals. The yogurt starter bacteria counts in all yogurt samples decreased to 60-90 % of the initial count. This amount is still in the minimum requirement at the time of sale.

According to Hamann and Marth (1987) yogurt should contain at least 10^6 cfu/g or ml at the time of sale. Starter culture activity is affected by the rate of cooling after incubation, level of acidity at the end of the incubation period and the temperature and duration of storage. Cooling is important to control the metabolic activity of the starters, in particular; however, a warm starter (freshly incubated and uncooked) is sometimes used in cheese factories and, to some extent, in the yogurt industry.

Yogurt starter bacteria, *S. thermophilus* and *L. bulgaricus*, are thermophilic homofermentative bacteria whose characteristics were summarized previously. The ratio of *S. thermophilus* to *L. bulgaricus* determines flavour and body characteristics of the ripened yogurt. In yogurt manufacture a ratio of 1:1 to 1:3 have been used. In this study, the ratio between *S. thermophilus* and *L. bulgaricus* is variable, but generally it is about 1.5:1 through the storage period.

Abrahamsen and Holmen (1980) reported on a 12-14 % of total solid yogurt, made from fortified UF retentate, stored for two weeks at 4-5 °C. The number of bacteria decreased during two weeks storage period from 79×10^7 to 27×10^7 cfu/ml (34%) for 14 % total solids UF retentate yogurt and 49×10^7 to 34×10^7 cfu/ml (69%) for the control yogurt.

Mistry and Hassan (1992) reported that the bacterial count of fresh and two weeks old yogurts, produced under different production conditions, did not differ with process conditions. The sum of rods and cocci was approximately 1.2×10^9 cfu/ml. After two weeks storage there was a slight decrease in counts; the sum of rods and cocci was approximately 1.1×10^9 cfu/ml.

Table (4.11) Changes of *L.bulgaricus* in set and stirred style thick yogurt produced from cow's and ewe's milk using different production methods during storage period (cfu/g)

Product codes	Storage period (Day)						
	1	7	14	21	28	35	42
CA	3.6E+08	3.0E+08	2.3E+08	9.0E+07	7.0E+07	4.3E+07	3.6E+07
CB	3.9E+08	3.0E+08	2.4E+08	1.7E+08	1.0E+08	9.0E+07	5.9E+07
CC	2.6E+08	2.2E+08	1.7E+08	1.4E+08	1.1E+08	8.8E+07	7.0E+07
CD	7.6E+08	6.0E+08	2.8E+08	2.3E+08	1.6E+08	1.2E+08	7.6E+07
CE	7.5E+08	5.0E+08	4.7E+08	3.5E+08	2.3E+08	1.6E+08	1.2E+08
CF	8.8E+08	7.3E+08	5.2E+08	3.5E+08	2.0E+08	1.9E+08	1.0E+08
CG	3.1E+08	2.2E+08	2.1E+08	1.8E+08	1.5E+08	1.1E+08	8.0E+07
EA	3.3E+08	2.4E+08	2.0E+08	1.4E+08	9.9E+07	8.4E+07	7.1E+07
EB	2.8E+08	2.4E+08	1.7E+08	1.6E+08	1.4E+08	1.1E+08	8.0E+07
EC	2.9E+08	2.9E+08	2.3E+08	2.2E+08	1.9E+08	1.7E+08	9.8E+07
ED	4.0E+08	3.4E+08	2.6E+08	2.0E+08	1.8E+08	1.5E+08	1.2E+08
EE	4.2E+08	3.0E+08	2.9E+08	2.4E+08	2.2E+08	1.8E+08	1.5E+08
EF	8.0E+08	4.7E+08	3.6E+08	2.9E+08	2.6E+08	2.5E+08	2.3E+08
EG	3.8E+08	2.3E+08	2.2E+08	2.1E+08	1.8E+08	1.4E+08	1.1E+08

Kneifel et al (1993) reported that relative changes of $\pm 50\%$ of the original viable count were used as a measure for a significant change in bacterial populations during storage. In about 45 % of the yogurts the lactobacilli counts increased during two weeks. On the other hand, a relative increase of the streptococci was observed only with 25 % of the yogurts, whereas 30 % were constant in their bacterial population. In two weeks storage period the total count of *L. bulgaricus* was between 5.5×10^7 - 6.5×10^8 and 3.4×10^7 - 5.3×10^8 cfu/ ml while for *S.thermophilus* it was 3.5×10^7 - 1.2×10^9 and 1.0×10^6 - 1×10^9 cfu/ml. Rao et al. (1987) investigated microbiological quality of labneh during six months storage period. The end of storage period lactic acid bacteria decreased. However lactic acid bacteria count was stable for the first four month of storage.

Table (4.12) Changes of *S.thermophilus* in set and stirred style thick yogurt produced from cow's and ewe's milk using different production methods during storage period (cfu/g)

Product codes	Storage period (Day)						
	1	7	14	21	28	35	42
CA	5.5E+08	5.4E+08	4.1E+08	1.9E+08	9.5E+07	7.0E+07	5.5E+07
CB	5.4E+08	4.0E+08	3.5E+08	2.1E+08	1.3E+08	9.7E+07	6.5E+07
CC	5.6E+08	4.7E+08	3.6E+08	3.4E+08	2.0E+08	1.4E+08	8.8E+07
CD	1.0E+09	7.1E+08	4.7E+08	2.7E+08	1.6E+08	1.0E+08	9.0E+07
CE	1.6E+09	1.2E+09	7.3E+08	5.1E+08	3.3E+08	2.0E+08	1.4E+08
CF	1.2E+09	7.8E+08	7.0E+08	3.4E+08	3.0E+08	2.7E+08	1.4E+08
CG	4.3E+08	3.3E+08	3.0E+08	2.5E+08	1.9E+08	1.4E+08	1.1E+08
EA	6.5E+08	6.0E+08	5.9E+08	4.1E+08	2.6E+08	8.1E+07	6.6E+07
EB	7.1E+08	6.6E+08	4.9E+08	4.1E+08	3.4E+08	1.4E+08	1.1E+08
EC	7.1E+08	6.8E+08	5.9E+08	5.4E+08	4.6E+08	1.2E+08	9.3E+07
ED	7.0E+08	6.6E+08	6.1E+08	4.9E+08	3.9E+08	3.7E+08	2.5E+08
EE	7.2E+08	7.2E+08	6.2E+08	4.6E+08	4.2E+08	2.9E+08	2.3E+08
EF	2.3E+09	2.0E+09	1.3E+09	1.0E+09	7.9E+08	6.8E+08	3.5E+08
EG	5.6E+08	5.0E+08	4.4E+08	3.7E+08	2.9E+08	1.9E+08	1.7E+08

It can be concluded that during storage period *S. thermophilus* and *L. bulgaricus* count decreased and the ratio of the starter bacteria is not in constant balance. Coliform bacteria count was negative. The results are similar to the references.

4.5 Thick yogurt production by using modified traditional filtration method

4.5.1 Theory of batch cake filtration method

The analysis of cake filtration is well documented by Ward (1987). If the filter cloth resistance can be neglected implying that all the resistance to the flow of filtrate is across the cake, as it can when the cake forms and thickens, the resulting differential equation is

$$\frac{dV}{d\Theta} = - \left[\frac{A^2}{2C_v V} \right] \Delta p$$

where Δp is the pressure drop across the filter cake.

V is the volume of filtrate collected.

A is the filter cloth area.

Θ is the time of filtration

C_v is assumed constant (it depends on the filter cake permeability which is assumed constant since the solids forming the cake are assumed rigid and uniformly deposited and on the solids concentration which is constant for a particular experiment).

This gives in integrated form: -

$$V^2 = \left[\frac{A^2 \Delta p}{2C_v} \right] \Theta$$

A plot of V versus Θ on logarithmic co-ordinates should give lines of slope 0.5 indicating V^2 proportional to Θ .

4.5.2 Whey drainage results of batch filtration

These represent the average of 5 measurements made on the 5 x 30 litres of each type of milk bought for the experiments. The aim was to double the solids content of the yogurt to that value associated with "Greek" type yogurt in the UK (20-24% total solid) or süzme/ torba yogurt in Turkey. It can be seen that this was accomplished when a vacuum was applied but it was not possible for the same drainage time when the yogurt was allowed to drain naturally.

The filtration results, for the two kinds of yogurts, are presented in Fig (4.26) for the synthetic filter medium and in Fig (4.27) for cotton filter medium plotted the way suggested in theory. The raw data are given in Appendix (VIII-1), (VIII-2), (VIII-3) and (VIII-4). These values are the result of triplicate measurements. Straight lines are obtained with some slight deviation at long times, however, the slope is not 0.5 and alters slightly with pressure drop. A cross plot at constant time indicated a linear relation with pressure. The data was correlated, for each yogurt- filter cloth combination, with an equation of the form:

$$V = (A + B\Delta p) (\Theta^C)$$

where Δp is the pressure (in.Hg) and A, B and C are constants given in Table (4.13) together with the Pearson product moment correlation coefficient, r, for each data set.

Table (4.13) Constants from fit of data to $[V = (A + B\Delta p)(\Theta^C)]$.

Yogurt / Filter media combinations	A	B	C	r
Ewe's milk / Cotton	17.81	4.258	0.347	0.99
Cow's milk / Cotton	32.18	5.977	0.395	0.94
Ewe's milk / Synthetic fibre	7.85	7.256	0.332	0.99
Cow's milk / Synthetic fibre	14.05	6.622	0.507	0.99

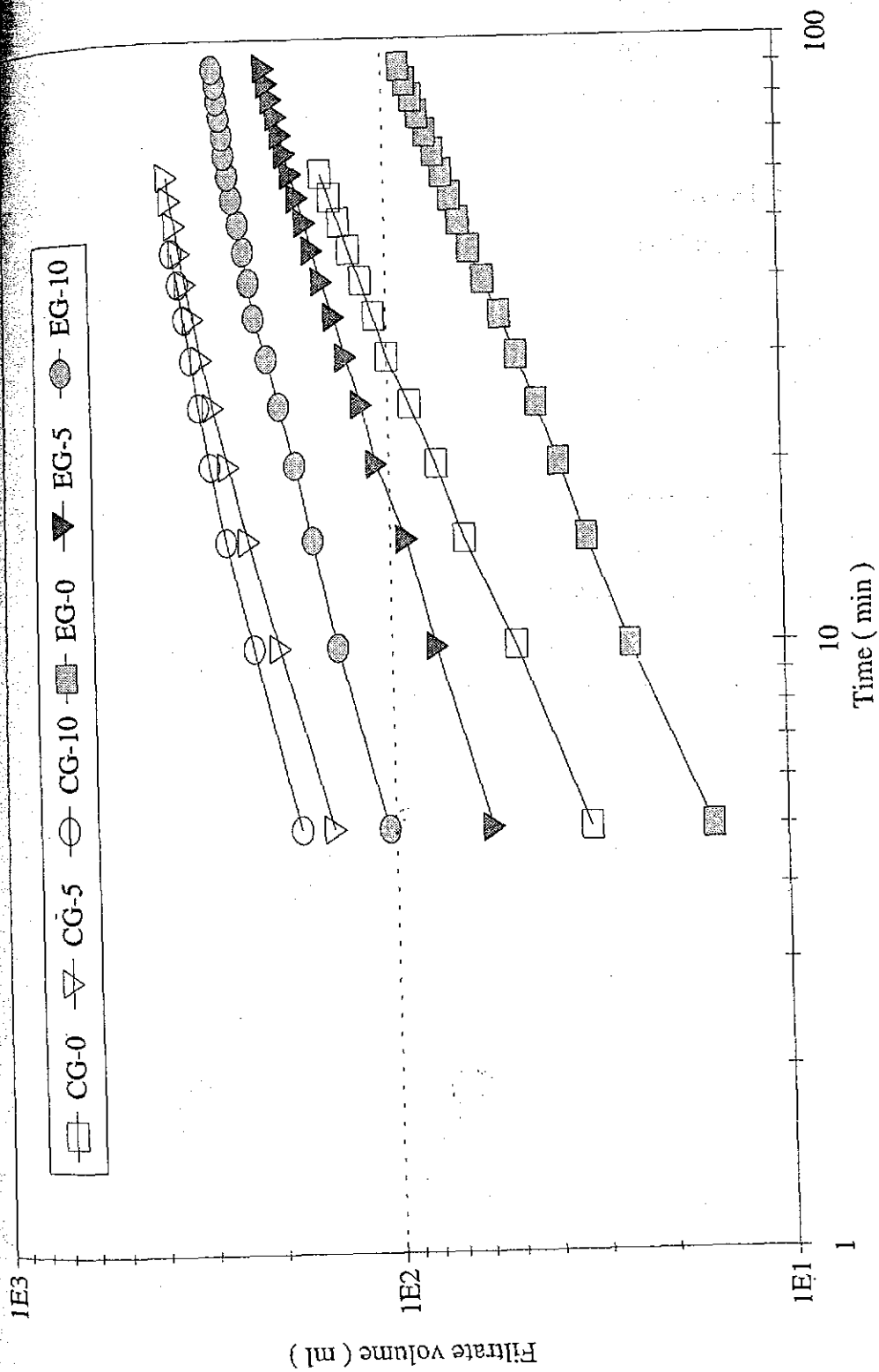


Fig (4.26) Plots of filtrate volume versus time for cow's and ewe's milk yogurt using the cotton fiber filter cloth

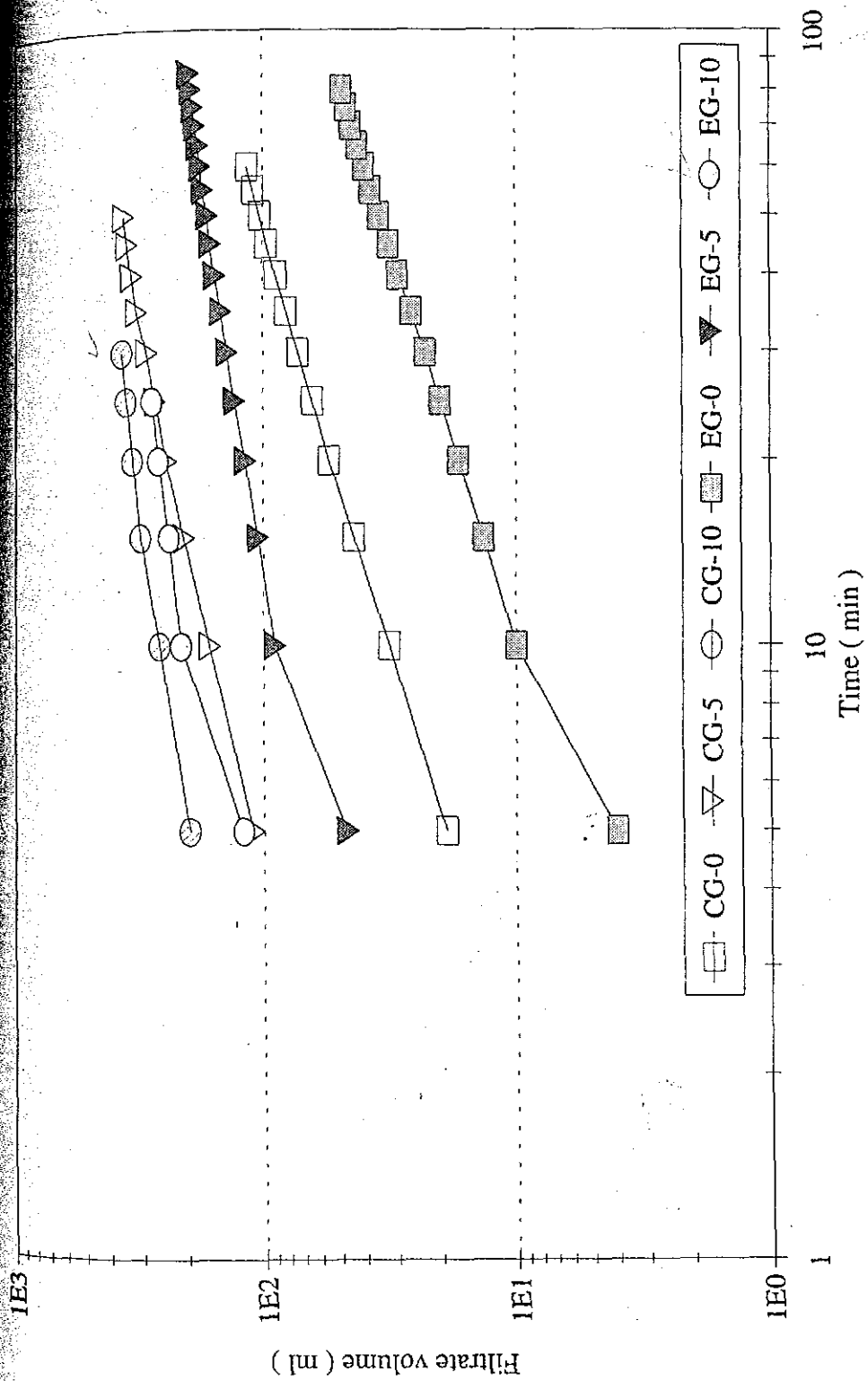


Fig (4.27) Plots of filtrate volume versus time for cow's and ewe's milk yogurt using synthetic filter cloth

The results for zero pressure drop correspond to natural drainage. The pressure drop is negligibly small compared to the 5 in.Hg and 10 in.Hg pressure drop used for the experiments but enough to cause natural drainage. Discrepancy is greatest for the highest pressure drop used in these experiments. An explanation for this is that the coagulum of yogurt is not strong unlike that in mineral filtration, so that under the load on the cake caused by the pressure drop the cake permeability changes with time as the cake is compressed and consolidated (compressible filter cake), c.f. the constant permeability used in the theory section.

When the cake was removed from the funnel, sliced across a diameter with a scalpel and the sections moved apart, a visual inspection of the cut showed the coagulum partially collapsed or crushed with it appearing driest adjacent to the filter cloth interface.

Tamime (1977) filtered yogurt of different solids content (from 12 to 20 % total solids) produced from reconstituted whole milk made with full fat spray dried milk powder using as the filter cloth a double layer of cheese cloth. The results indicated that the cake is quickly established to give straight lines as the analysis suggests but the slope is not 0.5 indicating a similar changes in permeability with time. Also, after some time the slope changes and this is thought to be due to a sudden change in cake structure caused by the weight of the cake and the pressure of the remaining yogurt since the yogurt is not rigid. A cross plot of the data indicated a linear relation with concentration so that for these data the correlation obtained was:-

$$V = (1408.9 - 54.14 * C) \Theta^{0.37}$$

Harwalkar and Kalab (1983) have presented similar results to the above for the influence of solids concentration on drainage rates. Yogurt was made from reconstituted milk, using non-fat dry milk powder. The total solids content was 10, 12.5 and 15 %.

the yogurt was allowed to drain naturally through a 120-mesh stainless steel screen into a measuring cylinder. Filtrate volume was expressed as a percentage of the original filter volume. Their data was plotted as suggested in theory. Again the data gives parallel straight lines after the first 10 minutes, once the filter cake is established. So that the equation of the form:-

$$V = (12.33 - 0.645 * C) \Theta^{0.415}$$

with the slope of 0.415 which is similar to that of 0.37 for the results reported by Tamime (1977). Both these values lie within the values given in Table (4.13). The exponent for Θ should be 0.5, however as noted previously, a possible explanation for the difference is that permeability is not constant i.e. C_v is not constant as the analysis requirements but is varying with time.

It can be concluded that the results indicate that a filter cake is formed during filtration. Due to the force, caused by the pressure difference applied or the weight of the cake as its thickness increases, the cake is partially collapsing so that the permeability changes with time (compressible filter cake). Also the initial solid content of the yogurt effects when this happens, since a thicker cake will be formed from an initially higher solids content yogurt for a given volume of filtrate collected, as the results show. Data taken from two sources found in the literature on the filtering of yogurt showed similar behaviour.

5.0 CONCLUSIONS

The fundamental difference in the manufacturing technique between modified traditional filtration and ultrafiltration is that the whey is drawn off after fermentation in the traditional process whereas the whey (permeate) is removed before fermentation in UF process. This difference leads to substantial changes in the composition of the yogurt. When yogurt is produced from UF milk retentate almost all the proteins are retained in the product, whereas some of the proteins are lost in the whey from traditional filtration process. The development of ultrafiltration technique into an industrial scale opens new possibilities as to rationalize and automatize the yogurt production and at the same time increase product yield due to a greater withhold of whey proteins in the product and a reduced loss of total protein as fines.

Obviously consumer reaction will need to be considered but the way is clearly open for some fundamental changes in the production of concentrated yogurt. The production of stirred style thick yogurt requires considerable labour and a controlled temperature and hygienic environment for drainage. The degree of drainage and the acidity of the curd are basic factors which determine the quality of the product but control of this factors are not always possible under the production conditions in industry.

It is evident from the results that modified traditional method can be used for the production of stirred style thick yogurt. The applied vacuum level for the modified traditional method can be recommended as 5 in.Hg and using cotton filter media. The advantages of these methods are short production time, low production cost and improved quality. The processing time of yogurt produced from fortified UF milk retentate is shorter as compared with the modified traditional method. These novel technologies can be recommended for the industrial production of set style thick Turkish yogurt. Thick yogurt produced UF milk retentate has one main disadvantage. which is high buffer capacity for high total solids milk.

Buffer capacity of high total solid UF milk retentate was observed to be high. Fermentation of fortified UF retentate can pose problems in achieving the desired pH for production of high total solids set style thick yogurt.

The average total solid content of thick yogurt produced by using modified traditional methods was 25 - 27 % containing 10 % protein and 10 % fat, 4.0 - 4.5 % lactose and 0.73 % ash. The protein and fat losses with UF permeate during the concentration period was not observed. However some protein and fat losses occurred in whey obtained from modified traditional method. Mineral contents of thick yogurt produced from fortified UF milk retentate are generally greater compared to control milk. The mineral content of yogurt produced by using modified traditional method is relatively lower than initial yogurt.

Traditional thick yogurt, produced commercially in Turkey, was analyzed. Its composition differs considerably from firm to firm.

The titratable acidity was higher in high total solids yogurt and during the storage period the titratable acidity increased but it was still in the range of Turkish Food Act. The pH was lower in high total solids yogurt produced from fortified UF retentate but it is higher in thick yogurt produced using modified traditional method. The pH decreased during storage period. The decreases were very slow for 30 and 32 % total solid levels.

Water holding capacity decreased due to total solids level and was not affected significantly by the storage period. Different production methods and milk used did not affect the water holding capacity significantly at the same total solid levels.

Set style thick yogurt, produced from fortified UF milk retentate, had a firmer texture than stirred style thick yogurt, produced by using modified traditional method. There were not marked differences in the firmness of thick yogurt produced from cow's and ewe's milk at the same total solid level. Firmness increased significantly when total solid level increased.

6.0 APPENDICES.

Appendix I:

- 1 Chemical composition of set style thick yogurt produced from fortified ultrafiltered cow's milk retentate.
- 2 Chemical composition of set style thick yogurt produced from fortified ultrafiltered ewe's milk retentate.
- 3 Chemical composition of stirred style thick yogurt and yogurt whey produced by using modified traditional batch cake filtration method for cow's milk.
- 4 Chemical composition of stirred style thick yogurt and yogurt whey produced by using modified traditional batch cake filtration method for ewe's milk.
- 5 Mineral content of thick yogurt produced from cow's milk.
- 6 Mineral content of thick yogurt produced from ewe's milk.

Appendix II:

- 1 Buffer capacity of fortified ultrafiltered cow's milk retentate.
- 2 Buffer capacity of fortified ultrafiltered ewe's milk retentate.

Appendix III:

- 1 Titratable acidity changes of fortified ultrafiltered cow's milk retentate during incubation period.
- 2 Titratable acidity changes of fortified ultrafiltered ewe's milk retentate during incubation period.
- 3 pH changes of fortified ultrafiltered cow's milk retentate during incubation period.
- 4 pH changes of fortified ultrafiltered ewe's milk retentate during incubation period.

- 5 Changes of pH in thick yogurt produced from cow's milk during storage period.
- 6 Changes of pH in thick yogurt produced from ewe's milk during storage period.
- 7 Changes of titratable acidity in thick yogurt produced from cow's milk during storage period.
- 8 Changes of titratable acidity in thick yogurt produced from ewe's milk during storage period.

Appendix IV:

- 1 Water holding capacity of thick yogurt produced from cow's milk by using different production process during storage period (%).
- 2 Water holding capacity of thick yogurt produced from ewe's milk by using different production process during storage period (%).
- 3 Penetration results of thick yogurt produced from different total solids cow's milk (result from three different areas).
- 4 Penetration results of thick yogurt produced from different total solids ewe's milk (result from three different areas).

Appendix V:

- 1 Changes of acetaldehyde for cow's milk yogurt during storage period (ppm).
- 2 Changes of acetaldehyde for ewe's milk yogurt during storage period (ppm).
- 3 Changes of acetone for cow's milk yogurt during storage period (ppm).
- 4 Changes of acetone for ewe's milk yogurt during storage period (ppm).
- 5 Changes of diacetyl for cow's milk yogurt during storage period (ppm).
- 6 Changes of diacetyl for ewe's milk yogurt during storage period (ppm).

Appendix VI:

- 1 Free amino acid changes during storage period from whole cow's milk yogurt (mg/100 g).
- 2 Free amino acid changes during storage period from about 24 % total

- solids thick yogurt produced from fortified ultrafiltered cow's milk retentate (mg/100 g).
- 3 Free amino acid changes during storage period from about 26 % total solids thick yogurt produced from fortified ultrafiltered cow's milk retentate (mg/100 g).
 - 4 Free amino acid changes during storage period from about 28 % total solids thick yogurt produced from fortified ultrafiltered cow's milk retentate (mg/100 g).
 - 5 Free amino acid changes during storage period from about 30 % total solids thick yogurt produced from fortified ultrafiltered cow's milk retentate (mg/100 g).
 - 6 Free amino acid changes during storage period from about 32 % total solids thick yogurt produced from fortified ultrafiltered cow's milk retentate (mg/100 g).
 - 7 Free amino acid changes during storage period from stirred style thick yogurt (CG coded) produced from cow's milk (mg / 100 g).
 - 8 Free amino acid changes during storage period from whole ewe's milk yogurt (mg/100 g).
 - 9 Free amino acid changes during storage period from about 24 % total solids thick yogurt produced from fortified ultrafiltered ewe's milk retentate (mg/100 g).
 - 10 Free amino acid changes during storage period from about 26 % total solids thick yogurt produced from fortified ultrafiltered ewe's milk retentate (mg/100 g).
 - 11 Free amino acid changes during storage period from about 28 % total solids thick yogurt produced from fortified ultrafiltered ewe's milk retentate (mg/100 g).
 - 12 Free amino acid changes during storage period from about 30 % total solids thick yogurt produced from fortified ultrafiltered ewe's milk retentate (mg/100 g).
 - 13 Free amino acid changes during storage period from about 32 % total

solids thick yogurt produced from fortified ultrafiltered ewe's milk retentate (mg/100 g).

14 Free amino acid changes during storage period from stirred style thick yogurt (EG coded) produced from ewe's milk (mg / 100 g).

Appendix VII:

- 1 Some microbiological changes in set and stirred style thick yogurts made from cow's milk using different production process during storage period at 5°C.
- 2 Some microbiological changes in set and stirred style thick yogurts made from ewe's milk using different production process during storage period at 5°C.

Appendix VIII:

- 1 Whey drainage from cow's milk yogurt by using cotton filter medium.
- 2 Whey drainage from cow's milk yogurt by using synthetic fiba filter medium.
- 3 Whey drainage from ewe's milk yogurt by using cotton filter medium.
- 4 Whey drainage from ewe's milk yogurt by using synthetic fiba filter medium.

Appendix IX:

- 1 Published paper.

Appendix I -1 Chemical composition of set style thick yogurt produced from fortified ultrafiltered cow's milk retentate.

Product codes	Trial	Total solids % (w/w)	Protein % (w/w)	Fat % (w/w)	Lactose % (w/w)	Ash % (w/w)	pH	Lactic acid %
CA	1	12.108	3.43	4.10	4.64	0.70	6.68	0.18
	2	12.122	3.48	4.00	4.75	0.73	6.70	0.18
	3	12.314	3.35	3.95	4.50	0.70	6.72	0.20
	Mean	12.181	3.42	4.02	4.63	0.71	6.70	0.19
	Std.er	0.0939	0.05	0.06	0.10	0.01	0.02	0.01
CB	1	23.743	8.00	9.90	4.60	1.18	6.52	0.21
	2	23.944	8.27	10.00	4.65	1.12	6.56	0.20
	3	24.397	8.30	10.40	4.72	0.98	6.46	0.25
	Mean	24.028	8.19	10.10	4.66	1.09	6.51	0.22
	Std.er	0.2734	0.13	0.22	0.05	0.08	0.04	0.02
CC	1	25.718	8.50	11.40	4.63	1.19	6.50	0.28
	2	26.162	8.62	11.70	4.69	1.15	6.52	0.24
	3	26.217	8.80	12.00	4.28	1.14	6.55	0.25
	Mean	26.033	8.64	11.70	4.53	1.16	6.52	0.26
	Std.er	0.2236	0.12	0.24	0.18	0.02	0.02	0.02
CD	1	27.816	9.50	12.60	4.47	1.25	6.50	0.26
	2	28.248	9.60	13.00	4.49	1.16	6.48	0.28
	3	28.197	9.90	12.20	4.30	1.21	6.53	0.20
	Mean	28.087	9.67	12.60	4.42	1.21	6.50	0.25
	Std.er	0.1926	0.17	0.33	0.08	0.04	0.02	0.03
CE	1	29.597	10.80	13.60	4.20	1.39	6.56	0.26
	2	29.930	11.00	13.40	4.35	1.18	6.54	0.20
	3	30.252	10.40	14.00	4.15	1.27	6.50	0.30
	Mean	29.926	10.73	13.67	4.23	1.28	6.53	0.25
	Std.er	0.2673	0.25	0.25	0.08	0.09	0.02	0.04
CF	1	31.696	11.70	14.70	4.10	1.44	6.50	0.30
	2	31.778	12.00	14.40	3.90	1.48	6.48	0.26
	3	32.488	11.90	15.00	4.03	1.31	6.53	0.28
	Mean	31.987	11.87	14.70	4.01	1.41	6.50	0.28
	Std.er	0.3555	0.12	0.24	0.08	0.07	0.02	0.02

Appendix I -2 Chemical composition of set style thick yogurt produced from fortified ultrafiltered ewe's milk retentate.

Product codes	Trial	Total solids % (w/w)	Protein % (w/w)	Fat % (w/w)	Lactose % (w/w)	Ash % (w/w)	pH	Lactic acid %
EA	1	17.306	5.40	5.90	5.07	0.94	6.67	0.22
	2	17.140	5.33	5.80	5.15	0.86	6.65	0.19
	3	16.920	5.53	6.60	4.89	0.92	6.70	0.20
	Mean	17.122	5.42	6.10	5.04	0.91	6.67	0.20
	Std.er	0.158	0.08	0.36	0.11	0.03	0.02	0.01
EB	1	23.805	8.35	8.50	5.00	1.19	6.58	0.25
	2	24.174	8.48	8.80	5.10	1.19	6.62	0.28
	3	24.530	8.59	9.00	4.95	1.30	6.65	0.30
	Mean	24.170	8.47	8.77	5.02	1.23	6.62	0.28
	Std.er	0.296	0.10	0.21	0.06	0.05	0.03	0.02
EC	1	25.796	8.89	10.90	4.76	1.25	6.63	0.28
	2	26.397	8.94	10.50	5.00	1.34	6.60	0.25
	3	26.572	9.36	10.00	5.10	1.41	6.56	0.24
	Mean	26.255	9.06	10.47	4.95	1.33	6.60	0.26
	Std.er	0.332	0.21	0.37	0.14	0.07	0.03	0.02
ED	1	27.832	10.10	11.40	4.99	1.34	6.62	0.26
	2	27.715	10.23	11.00	4.50	1.46	6.60	0.28
	3	28.315	10.50	11.30	4.80	1.39	6.55	0.30
	Mean	27.954	10.28	11.23	4.76	1.40	6.59	0.28
	Std.er	0.260	0.17	0.17	0.20	0.05	0.03	0.02
EE	1	29.860	11.75	12.10	4.56	1.45	6.62	0.24
	2	30.386	11.86	12.90	4.07	1.56	6.52	0.26
	3	29.995	11.50	12.50	4.95	1.49	6.60	0.30
	Mean	30.080	11.70	12.50	4.53	1.50	6.58	0.27
	Std.er	0.223	0.15	0.33	0.36	0.05	0.04	0.02
EF	1	31.799	12.75	13.70	3.71	1.64	5.50	0.30
	2	31.995	12.60	14.00	3.92	1.47	6.58	0.32
	3	32.145	12.96	13.50	4.31	1.38	6.62	0.30
	Mean	31.979	12.77	13.73	3.98	1.50	6.23	0.31
	Std.er	0.142	0.15	0.21	0.25	0.11	0.52	0.01

Products	Applied vacuum for filtration (ins.Hg)	Trial	Total Solids		Protein		Cotton		Slynt.		Cotton		Slynt.		Cotton		Slynt.		Cotton		Slynt.	
			Cotton	Slynt.	Cotton	Slynt.	Cotton	Slynt.	Cotton	Slynt.	Cotton	Slynt.	Cotton	Slynt.	Cotton	Slynt.	Cotton	Slynt.	Cotton	Slynt.	Cotton	Slynt.
Whole milk yogurt		1	12.33		3.31		3.90		4.59		0.71				4.30				0.93			
		2	12.11		3.40		4.10		4.75		0.72				4.22				1.04			
		3	12.27		3.37		3.90		4.66		0.71				4.37				1.09			
		Mean	12.24		3.36		3.97		4.67		0.71				4.30				1.02			
		Std.er	0.09		0.04		0.09		0.07		0.00				0.06				0.07			
Modified Traditional Batch Yogurt	0	1	15.1300	15.1290	4.65	4.25	5.30	5.00	4.46	4.51	0.73	0.74	4.35	4.33	4.35	4.33	1.05		1.05		1.00	
		1	26.300	24.593	10.50	9.65	10.20	10.10	4.61	4.30	0.72	0.74	4.25	4.33	4.25	4.33	1.10		1.10		1.15	
		2	26.830	25.643	10.42	9.92	10.50	10.30	4.35	4.50	0.70	0.71	4.35	4.28	4.35	4.28	1.24		1.24		1.30	
		3	26.580	23.648	10.16	9.71	10.70	9.80	4.70	4.18	0.73	0.71	4.20	4.40	4.20	4.40	1.20		1.20		1.00	
		Mean	26.570	24.628	10.36	9.76	10.47	10.07	4.55	4.33	0.72	0.72	4.27	4.34	4.27	4.34	1.18		1.18		1.15	
		Std.er	0.216	0.815	0.145	0.116	0.205	0.205	0.148	0.132	0.012	0.014	0.062	0.049	0.062	0.049	0.059		0.059		0.122	
	10	1	26.070	22.519	10.43	8.35	10.30	11.30	4.10	4.20	0.72	0.71	4.20	4.22	4.20	4.22	1.20		1.20		1.25	
		2	26.187	23.496	10.10	9.00	10.70	11.70	4.50	4.13	0.70	0.70	4.15	4.30	4.15	4.30	1.35		1.35		1.18	
		Mean	26.129	23.007	10.27	8.68	10.50	11.50	4.30	4.17	0.71	0.71	4.18	4.26	4.18	4.26	1.28		1.28		1.22	
		Std.er	0.06	0.49	0.17	0.33	0.20	0.20	0.20	0.04	0.01	0.01	0.02	0.04	0.02	0.04	0.08		0.08		0.04	
		1	5.283	5.540	0.21	0.45	0.10	0.20	4.47	4.30	0.67	0.70	4.35	4.36	4.35	4.36	1.05		1.05		0.98	
		1	5.870	5.880	0.84	0.88	0.20	0.30	4.25	4.30	0.72	0.74	4.35	4.30	4.35	4.30	1.08		1.08		1.30	
Modified Traditional Yogurt Whey	5	2	5.950	6.096	0.86	0.95	0.10	0.20	4.17	4.42	0.71	0.72	4.20	4.33	4.20	4.33	1.28		1.28		1.20	
		Mean	5.910	5.988	0.85	0.92	0.15	0.25	4.21	4.36	0.72	0.73	4.28	4.32	4.28	4.32	1.18		1.18		1.25	
		Std.er	0.04	0.11	0.01	0.03	0.05	0.05	0.04	0.06	0.01	0.01	0.07	0.02	0.07	0.02	0.10		0.10		0.05	
		1	6.250	7.830	0.90	1.09	0.80	1.00	4.19	4.47	0.72	0.73	4.20	4.28	4.20	4.28	1.32		1.32		1.20	
		2	6.400	8.200	1.30	1.22	1.00	1.20	4.26	4.13	0.70	0.72	4.15	4.24	4.15	4.24	1.37		1.37		1.27	
		Mean	6.325	8.015	1.10	1.16	0.90	1.10	4.23	4.30	0.71	0.73	4.18	4.26	4.18	4.26	1.35		1.35		1.24	
		Std.er	0.08	0.18	0.20	0.06	0.10	0.10	0.03	0.17	0.01	0.01	0.02	0.02	0.02	0.02	0.03		0.03		0.04	
	10	1	6.250	7.830	0.90	1.09	0.80	1.00	4.19	4.47	0.72	0.73	4.20	4.28	4.20	4.28	1.32		1.32		1.20	
		2	6.400	8.200	1.30	1.22	1.00	1.20	4.26	4.13	0.70	0.72	4.15	4.24	4.15	4.24	1.37		1.37		1.27	
		Mean	6.325	8.015	1.10	1.16	0.90	1.10	4.23	4.30	0.71	0.73	4.18	4.26	4.18	4.26	1.35		1.35		1.24	
		Std.er	0.08	0.18	0.20	0.06	0.10	0.10	0.03	0.17	0.01	0.01	0.02	0.02	0.02	0.02	0.03		0.03		0.04	
		1	6.250	7.830	0.90	1.09	0.80	1.00	4.19	4.47	0.72	0.73	4.20	4.28	4.20	4.28	1.32		1.32		1.20	
		2	6.400	8.200	1.30	1.22	1.00	1.20	4.26	4.13	0.70	0.72	4.15	4.24	4.15	4.24	1.37		1.37		1.27	

Appendix I - 3 Chemical composition of stirred style thick yogurt and yogurt whey produced by using modified traditional batch cake filtration method for cow's milk. [% (w/w)]

Products	Applied vacuum for filtration (ins.Hg)	Trial	Total Solids		Protein		Fat		Lactose		Cotton		Shynt.		Cotton		Shynt.		Cotton		Shynt.	
			Cotton	Shynt.	Cotton	Shynt.	Cotton	Shynt.	Cotton	Shynt.	Cotton	Shynt.	Cotton	Shynt.	Cotton	Shynt.	Cotton	Shynt.	Cotton	Shynt.	Cotton	Shynt.
Whole milk yogurt		1	16.950		5.61		5.60		4.84		0.91				4.50				1.20			
		2	17.140		5.50		5.60		4.80		0.89				4.40				1.33			
		Mean	17.045		5.56		5.60		4.82		0.90				4.45				1.27			
		Std.er	0.095		0.06		0.00		0.02		0.01				0.05				0.07			
Modified Traditional Batch Yogurt	0	1	19.200	18.860	6.85	6.20	6.75	6.15	4.70	4.60	0.90	0.91			4.40	4.35			1.48	1.32		
		1	25.373	23.540	9.67	9.10	11.50	9.80	4.15	4.23	0.92	0.90			4.19	4.30			1.35	1.26		
	5	2	27.643	24.180	10.16	9.50	12.10	10.20	4.30	4.20	0.83	0.92			4.28	4.15			1.26	1.32		
		Mean	26.508	23.860	9.92	9.30	11.80	10.00	4.23	4.22	0.88	0.91			4.24	4.23			1.31	1.29		
		Std.er	1.135	0.320	0.25	0.20	0.30	0.20	0.07	0.02	0.05	0.01			0.04	0.07			0.05	0.03		
	10	1	25.513	23.614	9.30	8.50	10.70	10.60	4.60	4.20	0.84	0.89			4.19	4.25			1.30	1.34		
		2	26.489	22.914	10.10	8.70	11.00	9.80	4.28	4.00	0.90	0.90			4.20	4.30			1.40	1.28		
		Mean	26.001	23.264	9.70	8.60	10.85	10.20	4.44	4.10	0.87	0.90			4.20	4.28			1.35	1.31		
		Std.er	0.488	0.350	0.40	0.10	0.15	0.40	0.16	0.10	0.03	0.01			0.00	0.02			0.05	0.03		
Modified Traditional Yogurt Whey	0	1	5.758	5.863	0.22	0.30	0.20	0.20	4.60	4.71	0.81	0.85			4.34	4.35			1.00	1.20		
		1	6.033	7.313	0.62	1.31	0.10	0.20	4.40	4.60	0.84	0.90			4.28	4.30			0.90	0.96		
	5	2	6.039	7.327	0.67	1.14	0.10	0.20	4.30	4.75	0.84	0.90			4.19	4.18			0.98	1.02		
		Mean	6.036	7.320	0.64	1.22	0.10	0.20	4.35	4.68	0.84	0.90			4.24	4.24			0.94	0.99		
		Std.er	0.003	0.007	0.03	0.09	0.00	0.00	0.05	0.08	0.00	0.00			0.04	0.06			0.04	0.03		
	10	1	7.199	10.096	0.99	2.99	0.30	1.00	4.60	4.70	0.88	0.86			4.30	4.27			0.99	0.96		
		2	7.229	10.118	0.81	2.71	0.30	1.20	4.77	4.65	0.90	0.89			4.32	4.29			1.05	0.95		
		Mean	7.214	10.107	0.90	2.85	0.30	1.10	4.69	4.68	0.89	0.88			4.31	4.28			1.02	0.96		
		Std.er	0.015	0.011	0.09	0.14	0.00	0.10	0.08	0.02	0.01	0.02			0.01	0.01			0.03	0.01		

Appendix 1 - 4 Chemical composition of stirred style thick yogurt and yogurt whey produced by using modified traditional batch cake filtration method for ewe's milk. [% (w/w)]

Appendix I - 5 Mineral content of yogurt produced from cow's milk.

Sample Codes	Tiral	Concentrations (mg/L)					
		Na	K	Mg	Ca	P	S
CA	1	432.96	1362.86	116.62	1044.52	1619.22	277.19
	2	490.93	1330.90	98.35	961.00	2569.53	367.37
	3	573.05	1335.18	104.64	989.52	2217.96	320.51
	Mean	498.98	1342.98	106.54	998.35	2135.57	321.69
	Std.er	57.47	14.17	7.58	34.67	392.31	36.82
CB	1	519.09	1498.89	169.98	2431.18	3152.13	679.05
	2	470.13	1513.91	167.56	2969.66	2109.77	637.42
	3	371.70	1147.00	138.43	2272.20	1592.35	474.82
	Mean	453.64	1386.60	158.66	2557.68	2284.75	597.10
	Std.er	61.29	169.54	14.34	298.46	648.69	88.12
CC	1	539.28	1524.52	180.55	2493.01	3790.71	494.00
	2	484.43	1398.11	128.24	2296.00	2716.26	481.39
	3	428.96	1440.85	151.06	2321.00	1979.69	444.81
	Mean	484.22	1454.49	153.29	2370.01	2828.89	473.40
	Std.er	45.04	52.50	21.41	87.58	743.62	20.86
CD	1	517.04	1499.85	197.02	2932.41	3239.82	480.80
	2	338.12	1269.13	168.70	2640.59	3058.27	450.30
	3	455.43	1493.59	176.86	3927.95	2488.92	549.00
	Mean	436.86	1420.86	180.86	3166.99	2929.00	493.36
	Std.er	74.21	107.32	11.90	551.12	319.89	41.26
CE	1	609.55	1666.98	201.15	3064.44	3658.22	533.92
	2	446.75	1263.05	159.09	2384.68	2891.93	455.92
	3	397.25	1366.88	185.07	3045.60	2000.05	644.01
	Mean	484.52	1432.30	181.77	2831.58	2850.07	544.62
	Std.er	90.69	171.27	17.33	316.10	677.59	77.16
CF	1	519.48	1536.35	215.77	3399.28	3267.75	483.32
	2	475.46	1591.48	179.58	3412.16	3640.11	499.96
	3	451.78	1541.93	187.93	3260.46	2383.72	457.25
	Mean	482.24	1556.59	194.42	3357.30	3097.19	480.18
	Std.er	28.05	24.78	15.47	68.68	526.90	17.58
CG	1	773.85	2249.08	138.15	490.77	1640.11	480.46
	2	703.85	2060.98	124.95	454.52	1336.20	447.93
	Mean	738.85	2155.03	131.55	472.65	1488.16	464.20
	Std.er	28.57	76.79	5.39	14.80	124.07	13.28

Appendix I - 6 Mineral content of thick yogurt produced from ewe's milk.

Sample Codes	Tiral	Concentrations (mg/L)					
		Na	K	Mg	Ca	P	S
EA	1	489.97	1145.66	179.92	1710.86	2141.55	750.56
	2	496.81	1173.37	176.59	1618.40	2567.34	688.44
	3	526.78	940.06	145.21	1486.61	1947.70	564.02
	Mean	504.52	1086.36	167.24	1605.29	2218.86	667.67
	Std.er	15.99	104.07	15.64	92.02	258.81	77.56
EB	1	430.69	1105.05	230.14	2793.86	3504.69	785.85
	2	418.05	1098.74	229.78	2670.58	3323.47	735.74
	3	482.96	1240.56	230.37	2801.85	3826.67	840.37
	Mean	443.90	1148.12	230.10	2755.43	3551.61	787.32
	Std.er	28.10	65.42	0.24	60.09	208.09	42.73
EC	1	436.42	1118.78	242.85	2876.23	3805.56	1103.96
	2	449.82	1155.48	282.74	2944.46	2082.54	1020.04
	3	423.24	1252.42	207.89	2609.70	3317.90	917.22
	Mean	436.50	1175.56	244.49	2810.13	3068.67	1013.74
	Std.er	10.85	56.37	30.58	144.44	725.16	76.37
ED	1	462.33	1144.57	245.89	3208.46	2809.82	726.74
	2	475.81	1211.04	272.16	3267.17	3264.77	827.84
	3	444.04	1337.86	287.64	3595.64	3205.63	549.85
	Mean	460.73	1231.16	268.56	3357.09	3093.41	701.48
	Std.er	13.02	80.18	17.23	170.38	201.97	114.89
EE	1	443.41	1232.57	267.67	3316.62	3448.42	1033.91
	2	451.77	1287.97	274.59	3323.07	2569.72	1039.64
	3	402.72	1103.18	234.69	3192.50	3077.15	579.07
	Mean	432.63	1207.91	258.99	3277.40	3031.76	884.21
	Std.er	21.42	77.43	17.41	60.09	360.16	215.77
EF	1	375.05	1147.22	231.03	3070.95	1950.62	543.40
	2	399.04	1167.23	243.76	3304.57	2023.13	545.22
	3	415.62	1108.49	207.66	2884.81	2875.84	813.88
	Mean	396.57	1140.98	227.48	3086.78	2283.20	634.16
	Std.er	16.65	24.38	14.95	171.73	420.10	127.08
EG	1	645.00	1975.00	145.00	745.00	1875.00	845.00
	2	796.00	2316.00	196.00	618.00	2151.00	915.00
	Mean	720.50	2145.50	170.50	681.50	2013.00	880.00
	Std.er	75.50	170.50	25.50	63.50	138.00	35.00

Appendix II - 1 Buffer capacity of fortified ultrafiltered cow's milk retentate

Sample codes	Trial	Lactic acid (μ moles/g media)								
		0.0	16.0	32.0	48.0	64.0	80.0	96.0	112.0	128.0
CA	1	6.54	6.00	5.58	5.20	4.93	4.58	4.36	4.20	4.02
	2	6.42	5.87	5.54	5.28	5.14	4.84	4.54	4.42	4.13
	3	6.52	6.10	5.68	5.20	4.93	4.58	4.36	4.20	4.00
	Mean	6.49	5.99	5.60	5.23	5.00	4.67	4.42	4.27	4.05
	Std.Er.	0.05	0.09	0.06	0.04	0.10	0.12	0.08	0.10	0.06
CB	1	6.20	6.11	5.90	5.58	5.28	5.06	4.96	4.82	4.70
	2	6.31	6.20	5.88	5.55	5.36	5.16	5.00	4.86	4.69
	3	6.28	6.15	5.82	5.60	5.43	5.25	5.10	4.95	4.78
	Mean	6.26	6.15	5.87	5.58	5.36	5.16	5.02	4.88	4.72
	Std.Er.	0.05	0.04	0.03	0.02	0.06	0.08	0.06	0.05	0.04
CC	1	6.24	6.08	5.88	5.68	5.49	5.32	5.21	5.10	4.96
	2	6.30	6.10	5.86	5.70	5.55	5.35	5.19	5.05	4.93
	3	6.26	6.04	5.84	5.69	5.53	5.35	5.21	5.04	4.88
	Mean	6.27	6.07	5.86	5.69	5.52	5.34	5.20	5.06	4.92
	Std.Er.	0.02	0.02	0.02	0.01	0.02	0.01	0.01	0.03	0.03
CD	1	6.26	6.12	5.97	5.79	5.60	5.45	5.31	5.18	5.07
	2	6.22	6.05	5.90	5.70	5.50	5.37	5.18	5.03	4.97
	3	6.15	6.00	5.84	5.62	5.46	5.22	5.10	5.00	4.92
	Mean	6.21	6.06	5.90	5.70	5.52	5.35	5.20	5.07	4.99
	Std.Er.	0.05	0.05	0.05	0.07	0.06	0.10	0.09	0.08	0.06
CE	1	6.16	6.00	5.82	5.61	5.45	5.29	5.18	5.05	4.98
	2	6.23	6.10	5.92	5.75	5.60	5.46	5.33	5.21	5.07
	3	6.30	6.15	5.96	5.71	5.57	5.40	5.24	5.14	5.07
	Mean	6.23	6.08	5.90	5.69	5.54	5.38	5.25	5.13	5.04
	Std.Er.	0.04	0.05	0.05	0.07	0.07	0.08	0.08	0.08	0.04
CF	1	6.27	6.13	5.99	5.78	5.63	5.46	5.34	5.21	5.16
	2	6.22	6.10	5.95	5.71	5.55	5.39	5.26	5.13	5.04
	3	6.17	6.03	5.86	5.68	5.53	5.36	5.25	5.16	5.10
	Mean	6.22	6.09	5.93	5.72	5.57	5.40	5.28	5.17	5.10
	Std.Er.	0.02	0.01	0.02	0.04	0.04	0.04	0.04	0.04	0.06

Appendix II - 2 Buffer capacity of fortified ultrafiltered ewe's milk retentate

Sample codes	Trial	Lactic acid ($\mu\text{mmol} / \text{g media}$)								
		0.0	16.0	32.0	48.0	64.0	80.0	96.0	112.0	128.0
EA	1	6.32	5.98	5.63	5.32	5.16	4.86	4.64	4.40	4.18
	2	6.42	6.15	5.75	5.51	5.26	5.00	4.80	4.40	4.20
	3	6.56	6.10	5.80	5.47	5.25	5.05	4.75	4.58	4.35
	Mean	6.43	6.08	5.73	5.43	5.22	4.97	4.73	4.46	4.24
	Std.Er.	0.10	0.07	0.07	0.08	0.04	0.08	0.07	0.08	0.08
EB	1	6.50	6.15	5.85	5.65	5.40	5.20	5.02	4.98	4.86
	2	6.42	6.10	5.77	5.72	5.40	5.26	5.16	4.94	4.80
	3	6.36	6.02	5.80	5.56	5.40	5.24	5.16	5.02	4.88
	Mean	6.43	6.09	5.81	5.64	5.40	5.23	5.11	4.98	4.85
	Std.Er.	0.06	0.05	0.03	0.07	0.00	0.02	0.07	0.03	0.03
EC	1	6.54	6.12	5.89	5.65	5.50	5.34	5.20	5.04	4.90
	2	6.38	6.06	5.83	5.58	5.43	5.27	5.13	5.02	4.89
	3	6.39	6.10	5.80	5.62	5.37	5.25	5.15	4.96	4.81
	Mean	6.44	6.09	5.84	5.62	5.43	5.29	5.16	5.01	4.87
	Std.Er.	0.07	0.02	0.04	0.03	0.05	0.04	0.03	0.03	0.04
ED	1	6.40	6.08	5.90	5.65	5.44	5.30	5.18	4.96	4.88
	2	6.38	6.06	5.85	5.63	5.46	5.34	5.22	5.12	5.00
	3	6.35	5.96	5.78	5.60	5.41	5.30	5.17	5.08	4.98
	Mean	6.38	6.03	5.84	5.63	5.44	5.31	5.19	5.05	4.95
	Std.Er.	0.02	0.05	0.05	0.02	0.02	0.02	0.02	0.07	0.05
EE	1	6.26	5.96	5.72	5.60	5.48	5.38	5.17	5.00	4.91
	2	6.27	5.97	5.75	5.62	5.51	5.32	5.20	5.12	5.02
	3	6.23	5.88	5.74	5.59	5.40	5.30	5.20	5.14	5.00
	Mean	6.25	5.94	5.74	5.60	5.46	5.33	5.19	5.09	4.98
	Std.Er.	0.02	0.04	0.01	0.01	0.05	0.03	0.01	0.06	0.05
EF	1	6.20	5.96	5.75	5.62	5.50	5.37	5.28	5.19	5.04
	2	6.24	5.81	5.65	5.53	5.41	5.30	5.25	5.18	5.08
	3	6.19	5.86	5.77	5.58	5.42	5.34	5.27	5.14	5.00
	Mean	6.21	5.88	5.72	5.58	5.44	5.34	5.27	5.17	5.04
	Std.Er.	0.02	0.06	0.05	0.04	0.04	0.03	0.01	0.02	0.03

Appendix III- 1 Titratable acidity changes of fortified ultrafiltered cow's milk retentate during incubation period

Product codes	Trial	Incubation time (min)									
		0	30	60	90	120	150	180	210	240	270
CA	1	0.18	0.24	0.36	0.48	0.64	0.80	0.95			
	2	0.20	0.24	0.34	0.42	0.58	0.76	0.90			
	3	0.24	0.30	0.38	0.48	0.60	0.84	0.89			
	Mean	0.21	0.26	0.36	0.46	0.61	0.80	0.91			
	Std. er.	0.02	0.03	0.02	0.03	0.02	0.03	0.03			
CB	1	0.28	0.34	0.42	0.54	0.66	0.96	1.10	1.26	1.37	
	2	0.24	0.30	0.38	0.52	0.85	1.04	1.24	1.34	1.41	
	3	0.22	0.28	0.36	0.50	0.74	0.95	1.18	1.30	1.39	
	Mean	0.25	0.31	0.39	0.52	0.75	0.98	1.17	1.30	1.39	
	Std. er.	0.02	0.02	0.02	0.02	0.08	0.04	0.06	0.03	0.02	
CC	1	0.26	0.32	0.44	0.62	0.86	1.08	1.22	1.38	1.49	1.57
	2	0.22	0.30	0.42	0.58	0.80	1.00	1.28	1.40	1.52	1.63
	3	0.20	0.26	0.38	0.55	0.78	1.03	1.20	1.35	1.48	1.59
	Mean	0.23	0.29	0.41	0.58	0.81	1.04	1.23	1.38	1.50	1.60
	Std. er.	0.02	0.02	0.02	0.03	0.03	0.03	0.03	0.02	0.02	0.02
CD	1	0.24	0.30	0.38	0.58	0.86	1.14	1.30	1.47	1.55	1.65
	2	0.30	0.36	0.44	0.66	0.94	1.08	1.24	1.40	1.52	1.63
	3	0.22	0.28	0.39	0.54	0.75	1.00	1.20	1.39	1.48	1.6
	Mean	0.25	0.31	0.40	0.59	0.85	1.07	1.25	1.42	1.52	1.63
	Std. er.	0.03	0.03	0.03	0.05	0.08	0.06	0.04	0.04	0.03	0.02
CE	1	0.28	0.36	0.42	0.60	0.76	1.06	1.25	1.40	1.53	1.62
	2	0.30	0.40	0.50	0.64	0.94	1.22	1.40	1.52	1.61	1.7
	3	0.24	0.30	0.42	0.58	0.88	1.04	1.26	1.42	1.55	1.63
	Mean	0.27	0.35	0.45	0.61	0.86	1.11	1.30	1.45	1.56	1.65
	Std. er.	0.02	0.04	0.04	0.02	0.07	0.08	0.07	0.05	0.03	0.04
CF	1	0.25	0.29	0.40	0.64	0.84	1.18	1.42	1.56	1.65	1.72
	2	0.24	0.29	0.38	0.58	0.93	1.16	1.34	1.50	1.60	1.69
	3	0.22	0.28	0.39	0.62	0.88	1.08	1.26	1.43	1.55	1.64
	Mean	0.24	0.29	0.39	0.61	0.88	1.14	1.34	1.50	1.60	1.68
	Std. er.	0.01	0.00	0.01	0.02	0.04	0.04	0.07	0.05	0.04	0.03

Appendix III- 2 Titratable acidity changes of fortified ultrafiltered ewe's milk retentate during incubation period

Product codes	Trial	Incubation time (min)									
		0	30	60	90	120	150	180	210	240	270
EA	1	0.18	0.22	0.29	0.33	0.47	0.68	0.88			
	2	0.22	0.25	0.31	0.36	0.48	0.70	0.92			
	3	0.20	0.27	0.33	0.42	0.62	0.75	0.95			
	Mean	0.20	0.25	0.31	0.37	0.52	0.71	0.92			
	Std.er	0.02	0.02	0.02	0.04	0.07	0.03	0.03			
EB	1	0.22	0.25	0.31	0.40	0.61	0.81	0.98	1.12	1.25	1.36
	2	0.18	0.26	0.34	0.44	0.68	0.85	1.00	1.15	1.27	1.39
	3	0.24	0.29	0.36	0.45	0.70	0.90	1.04	1.22	1.34	1.42
	Mean	0.21	0.27	0.34	0.43	0.66	0.85	1.01	1.16	1.29	1.39
	Std.er	0.02	0.02	0.02	0.02	0.04	0.04	0.02	0.04	0.04	0.02
EC	1	0.21	0.26	0.36	0.48	0.68	0.87	1.08	1.28	1.39	1.47
	2	0.20	0.30	0.38	0.50	0.74	0.95	1.13	1.34	1.47	1.55
	3	0.25	0.29	0.36	0.55	0.75	0.98	1.18	1.38	1.51	1.58
	Mean	0.22	0.28	0.37	0.51	0.72	0.93	1.13	1.33	1.46	1.53
	Std.er	0.02	0.02	0.01	0.03	0.03	0.05	0.04	0.04	0.05	0.05
ED	1	0.19	0.29	0.40	0.52	0.67	0.88	1.13	1.24	1.40	1.51
	2	0.21	0.27	0.33	0.45	0.75	0.90	1.24	1.45	1.54	1.60
	3	0.25	0.30	0.40	0.60	0.80	1.00	1.19	1.39	1.47	1.56
	Mean	0.22	0.29	0.38	0.52	0.74	0.93	1.19	1.36	1.47	1.56
	Std.er	0.02	0.01	0.03	0.06	0.05	0.05	0.04	0.09	0.06	0.04
EE	1	0.20	0.27	0.36	0.58	0.78	1.08	1.26	1.45	1.56	1.63
	2	0.22	0.25	0.38	0.55	0.82	1.03	1.22	1.52	1.60	1.68
	3	0.22	0.27	0.36	0.52	0.72	0.99	1.18	1.34	1.46	1.58
	Mean	0.21	0.26	0.37	0.55	0.77	1.03	1.22	1.44	1.54	1.63
	Std.er	0.01	0.01	0.01	0.02	0.04	0.04	0.03	0.07	0.06	0.04
EF	1	0.22	0.26	0.39	0.55	0.80	0.98	1.24	1.48	1.57	1.65
	2	0.24	0.30	0.42	0.62	0.90	1.10	1.30	1.53	1.60	1.69
	3	0.25	0.30	0.40	0.54	0.85	1.00	1.22	1.42	1.51	1.62
	Mean	0.24	0.29	0.40	0.57	0.85	1.03	1.25	1.48	1.56	1.65
	Std.er	0.01	0.02	0.01	0.04	0.04	0.05	0.03	0.04	0.04	0.03

Appendix III - 3 pH changes of fortified ultrafiltered cow's milk retentate during incubation period

Product Codes	Trial	Incubation time (min)									
		0	30	60	90	120	150	180	210	240	270
CA	1	6.34	6.18	6.09	5.82	5.25	4.89	4.60			
	2	6.25	6.15	6.00	5.78	5.20	4.75	4.55			
	3	6.40	6.26	6.05	5.80	5.30	4.90	4.60			
	Mean	6.33	6.20	6.05	5.80	5.25	4.85	4.58			
	Std.er	0.06	0.05	0.04	0.02	0.04	0.07	0.02			
CB	1	6.00	5.95	5.90	5.80	5.55	5.10	4.90	4.70		
	2	6.19	6.15	6.04	5.82	5.45	5.05	4.85	4.64		
	3	6.23	6.18	6.00	5.90	5.50	5.20	4.95	4.72		
	Mean	6.14	6.09	5.98	5.84	5.50	5.12	4.90	4.69		
	Std.er	0.10	0.10	0.06	0.04	0.04	0.06	0.04	0.03		
CC	1	6.20	6.15	6.04	5.86	5.57	5.35	5.17	5.02	4.90	4.76
	2	6.15	6.10	6.00	5.80	5.53	5.29	5.13	4.98	4.83	4.71
	3	6.19	6.10	5.95	5.70	5.42	5.22	5.05	4.96	4.83	4.7
	Mean	6.18	6.12	6.00	5.79	5.51	5.29	5.12	4.99	4.85	4.72
	Std.er	0.02	0.02	0.04	0.07	0.06	0.05	0.05	0.02	0.03	0.03
CD	1	6.10	6.03	5.88	5.72	5.60	5.44	5.25	5.10	4.97	4.79
	2	6.18	6.10	6.00	5.83	5.53	5.29	5.15	5.02	4.90	4.73
	3	6.22	6.15	6.03	5.95	5.57	5.25	5.10	4.95	4.82	4.7
	Mean	6.17	6.09	5.97	5.83	5.57	5.33	5.17	5.02	4.90	4.74
	Std.er	0.05	0.05	0.06	0.09	0.03	0.08	0.06	0.06	0.06	0.04
CE	1	6.15	6.07	5.88	5.73	5.50	5.30	5.13	5.00	4.88	4.75
	2	6.16	6.10	6.02	5.84	5.52	5.28	5.14	5.02	4.92	4.78
	3	6.20	6.12	6.00	5.87	5.59	5.32	5.02	4.90	4.80	4.7
	Mean	6.17	6.10	5.97	5.81	5.54	5.30	5.10	4.97	4.87	4.74
	Std.er	0.02	0.02	0.06	0.06	0.04	0.02	0.05	0.05	0.05	0.03
CF	1	6.11	6.04	5.82	5.76	5.57	5.35	5.17	5.03	4.91	4.74
	2	6.14	6.09	5.98	5.86	5.60	5.40	5.22	5.06	4.95	4.78
	3	6.21	6.17	6.10	5.96	5.64	5.30	5.14	4.99	4.86	4.72
	Mean	6.15	6.10	5.97	5.86	5.60	5.35	5.18	5.03	4.91	4.75
	Std.er	0.04	0.05	0.11	0.08	0.03	0.04	0.03	0.03	0.04	0.02

Appendix III - 4 pH changes of fortified ultrafiltered ewe's milk retentate during incubation period

Product Codes	Trial	Incubation time (min)									
		0	30	60	90	120	150	180	210	240	270
EA	1	6.26	6.02	5.79	5.27	4.90	4.70	4.56			
	2	6.13	6.04	5.83	5.46	5.00	4.80	4.63			
	3	6.20	6.16	5.99	5.78	5.28	4.92	4.60			
	Mean	6.20	6.07	5.87	5.50	5.06	4.81	4.60			
	Std.er	0.05	0.06	0.09	0.21	0.16	0.09	0.03			
EB	1	6.28	6.22	5.98	5.79	5.40	5.15	4.90	4.76	4.64	
	2	6.22	6.18	6.09	5.90	5.60	5.35	5.10	4.90	4.71	
	3	6.12	6.12	6.05	5.84	5.50	5.20	5.03	4.80	4.66	
	Mean	6.21	6.17	6.04	5.84	5.50	5.23	5.01	4.82	4.67	
	Std.er	0.07	0.04	0.05	0.04	0.08	0.08	0.08	0.06	0.03	
EC	1	6.24	6.19	6.03	5.85	5.55	5.32	5.15	4.98	4.83	4.7
	2	6.18	6.17	6.08	5.90	5.60	5.43	5.25	5.13	4.97	4.75
	3	6.19	6.24	6.16	6.04	5.85	5.48	5.22	5.04	4.90	4.72
	Mean	6.20	6.20	6.09	5.93	5.67	5.41	5.21	5.05	4.90	4.72
	Std.er	0.03	0.03	0.05	0.08	0.13	0.07	0.04	0.06	0.06	0.02
ED	1	6.20	6.14	6.05	5.90	5.66	5.42	5.20	5.06	4.92	4.76
	2	6.18	6.11	5.96	5.75	5.50	5.33	5.15	5.00	4.87	4.73
	3	6.17	6.13	6.02	5.88	5.57	5.32	5.14	5.00	4.87	4.7
	Mean	6.18	6.13	6.01	5.84	5.58	5.36	5.16	5.02	4.89	4.73
	Std.er	0.01	0.01	0.04	0.07	0.07	0.04	0.03	0.03	0.02	0.02
EE	1	6.16	6.10	5.95	5.67	5.46	5.25	5.10	4.97	4.84	4.73
	2	6.16	6.10	6.00	5.80	5.52	5.30	5.16	5.02	4.86	4.7
	3	6.29	6.20	6.06	5.90	5.60	5.40	5.24	5.05	4.90	4.75
	Mean	6.20	6.13	6.00	5.79	5.53	5.32	5.17	5.01	4.87	4.73
	Std.er	0.06	0.05	0.04	0.09	0.06	0.06	0.06	0.03	0.02	0.02
EF	1	6.18	6.12	6.00	5.91	5.64	5.45	5.17	5.05	4.93	4.77
	2	6.28	6.20	6.10	5.96	5.70	5.40	5.15	5.02	4.90	4.76
	3	6.14	6.09	6.02	5.87	5.60	5.44	5.21	5.07	4.95	4.8
	Mean	6.20	6.14	6.04	5.91	5.65	5.43	5.18	5.05	4.93	4.78
	Std.er	0.06	0.05	0.04	0.04	0.04	0.02	0.02	0.02	0.02	0.02

Appendix III - 5 Changes of pH in thick yogurt produced from cow's milk during storage period

Product Codes	Trial	pH during storage period (day)						
		1	7	14	21	28	35	42
CA	1	4.45	4.25	4.13	4.02	3.94	3.86	3.80
	2	4.40	4.22	4.10	4.05	3.96	3.90	3.88
	3	4.30	4.15	4.05	4.00	3.92	3.80	3.75
	Mean	4.38	4.21	4.09	4.02	3.94	3.85	3.81
	Std.er	0.06	0.04	0.03	0.02	0.02	0.04	0.05
CB	1	4.55	4.46	4.40	4.37	4.28	4.23	4.20
	2	4.50	4.41	4.35	4.30	4.23	4.20	4.18
	3	4.48	4.40	4.33	4.27	4.25	4.19	4.16
	Mean	4.51	4.42	4.36	4.31	4.25	4.21	4.18
	Std.er	0.03	0.03	0.03	0.04	0.02	0.02	0.02
CC	1	4.52	4.55	4.48	4.40	4.36	4.32	4.26
	2	4.58	4.50	4.42	4.35	4.30	4.26	4.22
	3	4.50	4.43	4.37	4.30	4.25	4.21	4.24
	Mean	4.53	4.49	4.42	4.35	4.30	4.26	4.24
	Std.er	0.03	0.05	0.04	0.04	0.04	0.04	0.02
CD	1	4.54	4.50	4.45	4.40	4.35	4.31	4.28
	2	4.56	4.49	4.42	4.39	4.34	4.30	4.25
	3	4.58	4.50	4.42	4.35	4.29	4.23	4.20
	Mean	4.56	4.50	4.43	4.38	4.33	4.28	4.24
	Std.er	0.02	0.00	0.01	0.02	0.03	0.04	0.03
CE	1	4.70	4.66	4.61	4.60	4.58	4.53	4.50
	2	4.66	4.62	4.54	4.52	4.50	4.50	4.48
	3	4.65	4.59	4.55	4.56	4.53	4.50	4.48
	Mean	4.67	4.62	4.57	4.56	4.54	4.51	4.49
	Std.er	0.02	0.03	0.03	0.03	0.03	0.01	0.01
CF	1	4.70	4.62	4.60	4.59	4.54	4.52	4.50
	2	4.72	4.66	4.58	4.55	4.55	4.51	4.51
	3	4.74	4.65	4.60	4.58	4.53	4.53	4.50
	Mean	4.72	4.64	4.59	4.57	4.54	4.52	4.50
	Std.er	0.02	0.02	0.01	0.02	0.01	0.01	0.00
CG	1	4.46	4.32	4.28	4.22	4.18	4.17	4.15
	2	4.38	4.27	4.19	4.20	4.19	4.20	4.18
	Mean	4.42	4.30	4.24	4.21	4.19	4.19	4.17
	Std.er	0.04	0.03	0.04	0.01	0.01	0.02	0.01

Appendix III - 6 Changes of pH in thick yogurt produced from ewe's milk during storage period

Product Codes	Trial	pH during storage period (day)						
		1	7	14	21	28	35	42
EA	1	4.37	4.19	4.17	4.12	4.05	3.95	3.90
	2	4.46	4.25	4.15	4.10	4.20	4.07	4.00
	3	4.50	4.35	4.20	4.15	4.12	4.00	3.95
	Mean	4.44	4.26	4.17	4.12	4.12	4.01	3.95
	Std.er	0.05	0.07	0.02	0.02	0.06	0.05	0.04
EB	1	4.45	4.37	4.30	4.22	4.19	4.17	4.13
	2	4.48	4.41	4.26	4.25	4.24	4.21	4.16
	3	4.55	4.48	4.37	4.26	4.22	4.18	4.10
	Mean	4.49	4.42	4.31	4.24	4.22	4.19	4.13
	Std.er	0.04	0.05	0.05	0.02	0.02	0.02	0.02
EC	1	4.48	4.39	4.33	4.30	4.25	4.21	4.18
	2	4.50	4.41	4.34	4.30	4.27	4.15	4.15
	3	4.56	4.48	4.42	4.28	4.20	4.19	4.16
	Mean	4.51	4.43	4.36	4.29	4.24	4.18	4.16
	Std.er	0.03	0.04	0.04	0.01	0.03	0.02	0.01
ED	1	4.54	4.45	4.39	4.32	4.22	4.22	4.18
	2	4.52	4.43	4.34	4.22	4.26	4.20	4.20
	3	4.50	4.43	4.40	4.36	4.30	4.25	4.18
	Mean	4.52	4.44	4.38	4.30	4.26	4.22	4.19
	Std.er	0.02	0.01	0.03	0.06	0.03	0.02	0.01
EE	1	4.70	4.62	4.64	4.64	4.60	4.60	4.58
	2	4.67	4.65	4.64	4.65	4.59	4.59	4.55
	3	4.66	4.60	4.59	4.58	4.56	4.55	4.53
	Mean	4.68	4.62	4.62	4.62	4.58	4.58	4.55
	Std.er	0.02	0.02	0.02	0.03	0.02	0.02	0.02
EF	1	4.74	4.70	4.66	4.63	4.59	4.56	4.57
	2	4.70	4.69	4.65	4.62	4.62	4.60	4.56
	3	4.67	4.65	4.63	4.65	4.60	4.60	4.61
	Mean	4.70	4.68	4.65	4.63	4.60	4.59	4.58
	Std.er	0.03	0.02	0.01	0.01	0.01	0.02	0.02
EG	1	4.38	4.21	4.20	4.15	4.15	4.13	4.10
	2	4.31	4.30	4.24	4.22	4.14	4.12	4.14
	Mean	4.35	4.26	4.22	4.19	4.15	4.13	4.12
	Std.er	0.04	0.04	0.02	0.03	0.01	0.00	0.02

Appendix III - 7 Changes of titratable acidity in thick yogurt from cow's milk during storage period

Product codes	Trial	Storage period (day)						
		1	7	14	21	28	35	42
CA	1	1.08	1.17	1.25	1.30	1.34	1.39	1.42
	2	1.15	1.25	1.34	1.35	1.38	1.43	1.49
	3	1.00	1.13	1.20	1.27	1.33	1.40	1.44
	Mean	1.08	1.18	1.26	1.31	1.35	1.41	1.45
	Std.er.	0.06	0.05	0.06	0.03	0.02	0.02	0.03
CB	1	1.33	1.39	1.45	1.52	1.59	1.64	1.68
	2	1.37	1.45	1.50	1.56	1.61	1.67	1.74
	3	1.34	1.41	1.51	1.55	1.63	1.69	1.76
	Mean	1.35	1.42	1.49	1.54	1.61	1.67	1.73
	Std.er.	0.02	0.02	0.03	0.02	0.02	0.02	0.03
CC	1	1.37	1.48	1.59	1.67	1.75	1.79	1.85
	2	1.35	1.43	1.51	1.63	1.70	1.77	1.82
	3	1.40	1.50	1.59	1.68	1.77	1.84	1.89
	Mean	1.37	1.47	1.56	1.66	1.74	1.80	1.85
	Std.er.	0.02	0.03	0.04	0.02	0.03	0.03	0.03
CD	1	1.44	1.51	1.62	1.71	1.80	1.87	1.95
	2	1.39	1.50	1.60	1.72	1.83	1.90	1.95
	3	1.36	1.48	1.59	1.67	1.78	1.80	1.89
	Mean	1.40	1.50	1.60	1.70	1.80	1.86	1.93
	Srd.er.	0.03	0.01	0.01	0.02	0.02	0.04	0.03
CE	1	1.45	1.60	1.71	1.85	1.88	1.97	2.00
	2	1.42	1.56	1.68	1.80	1.90	1.96	2.03
	3	1.36	1.48	1.60	1.70	1.81	1.90	1.97
	Mean	1.41	1.55	1.66	1.78	1.86	1.94	2.00
	Std.er.	0.04	0.05	0.05	0.06	0.04	0.03	0.02
CF	1	1.45	1.57	1.67	1.79	1.85	1.95	2.01
	2	1.42	1.55	1.70	1.80	1.89	1.97	2.00
	3	1.48	1.62	1.78	1.90	1.97	2.05	2.09
	Mean	1.45	1.58	1.72	1.83	1.90	1.99	2.03
	Std.er.	0.02	0.03	0.05	0.05	0.05	0.04	0.04
CG	1	1.30	1.40	1.51	1.56	1.58	1.65	1.65
	2	1.36	1.50	1.54	1.60	1.66	1.60	1.67
	Mean	1.33	1.45	1.53	1.58	1.62	1.63	1.66
	Std.er.	0.03	0.05	0.015	0.02	0.04	0.025	0.01

Appendix III - 8 Changes of titratable acidity in thick yogurt from ewe's milk during storage period

Product codes	Trial	Storage period (day)						
		1	7	14	21	28	35	42
EA	1	1.14	1.26	1.34	1.45	1.54	1.62	1.68
	2	1.23	1.34	1.44	1.52	1.60	1.65	1.65
	3	1.18	1.30	1.40	1.48	1.54	1.62	1.68
	Mean	1.18	1.30	1.39	1.48	1.56	1.63	1.67
	Std.er.	0.04	0.03	0.04	0.03	0.03	0.01	0.01
EB	1	1.34	1.46	1.54	1.66	1.72	1.78	1.85
	2	1.40	1.53	1.63	1.70	1.75	1.80	1.88
	3	1.50	1.58	1.66	1.75	1.79	1.84	1.90
	Mean	1.41	1.52	1.61	1.70	1.75	1.81	1.88
	Std.er.	0.07	0.05	0.05	0.04	0.03	0.02	0.02
EC	1	1.44	1.58	1.67	1.75	1.80	1.86	1.92
	2	1.52	1.66	1.76	1.82	1.91	1.98	2.04
	3	1.56	1.68	1.76	1.80	1.87	1.94	2.00
	Mean	1.51	1.64	1.73	1.79	1.86	1.93	1.99
	Std.er.	0.05	0.04	0.04	0.03	0.05	0.05	0.05
ED	1	1.50	1.63	1.75	1.80	1.86	1.93	2.01
	2	1.60	1.76	1.84	1.91	1.95	2.00	2.08
	3	1.54	1.68	1.80	1.86	1.90	1.97	2.00
	Mean	1.55	1.69	1.80	1.86	1.90	1.97	2.03
	Std.er.	0.04	0.05	0.04	0.04	0.04	0.03	0.04
EE	1	1.60	1.70	1.79	1.85	1.89	2.00	2.05
	2	1.65	1.76	1.80	1.86	1.90	1.96	2.01
	3	1.52	1.64	1.76	1.85	1.93	1.97	2.06
	Mean	1.59	1.70	1.78	1.85	1.91	1.98	2.04
	Std.er.	0.05	0.05	0.02	0.00	0.02	0.02	0.02
EF	1	1.60	1.72	1.80	1.88	1.94	2.00	2.08
	2	1.72	1.80	1.89	1.96	1.99	2.05	2.09
	3	1.56	1.70	1.81	1.90	1.98	2.02	2.10
	Mean	1.63	1.74	1.83	1.91	1.97	2.02	2.09
	Std.er.	0.07	0.04	0.04	0.03	0.02	0.02	0.01
EG	1	1.45	1.57	1.60	1.68	1.73	1.77	1.81
	2	1.40	1.51	1.59	1.70	1.68	1.71	1.79
	Mean	1.43	1.54	1.60	1.69	1.71	1.74	1.80
	Std.er.	0.025	0.03	0.005	0.01	0.025	0.03	0.01

Appendix IV - 1 Water holding capacity of thick yogurt produced from cow's milk by using different production process during storage period (%).

Product Codes	Trial	Storage period (day)						
		1	7	14	21	28	35	42
CA	1	70.50	67.80	68.59	67.83	63.58	63.74	64.11
	2	71.05	69.75	66.92	68.51	65.91	62.15	62.70
	Mean	70.78	68.78	67.76	68.17	64.75	62.95	63.41
CB	1	22.41	21.90	20.78	21.42	19.23	19.07	19.40
	2	23.60	22.70	21.90	20.50	19.80	19.90	19.10
	Mean	23.01	22.30	21.34	20.96	19.52	19.49	19.25
CC	1	22.49	19.60	19.25	18.72	17.81	18.02	17.67
	2	20.55	19.15	18.75	17.96	17.80	17.50	16.90
	Mean	21.52	19.38	19.00	18.34	17.81	17.76	17.29
CD	1	19.12	18.54	18.53	18.01	17.36	16.52	15.66
	2	19.75	18.80	17.63	17.15	16.85	16.70	16.80
	Mean	19.44	18.67	18.08	17.58	17.11	16.61	16.23
CE	1	16.75	16.45	16.89	16.37	15.50	15.63	14.23
	2	15.56	15.23	15.40	14.78	14.90	14.42	14.94
	Mean	16.16	15.84	16.15	15.58	15.20	15.03	14.59
CF	1	10.75	9.09	8.83	8.57	8.56	8.71	8.17
	2	12.45	11.89	10.67	11.50	10.42	10.20	9.87
	Mean	11.60	10.49	9.75	10.04	9.49	9.46	9.02
CG	1	16.25	16.12	15.45	13.89	13.25	13.27	13.32
	2	17.30	15.38	14.46	14.27	14.61	13.84	13.50
	Mean	16.78	15.75	14.96	14.08	13.93	13.56	13.41

Appendix IV - 2 Water holding capacity of thick yogurt produced from ewe's milk by using different production process during storage period (%).

Product Codes	Trial	Storage period (day)						
		1	7	14	21	28	35	42
EA	1	53.93	47.93	44.76	45.22	46.13	45.10	45.36
	2	56.25	45.27	45.10	44.18	43.80	42.78	43.27
	Mean	55.09	46.60	44.93	44.70	44.97	43.94	44.32
EB	1	25.68	23.62	22.83	21.60	20.55	19.85	19.98
	2	24.20	22.27	20.85	19.50	19.70	18.87	17.50
	Mean	24.94	22.95	21.84	20.55	20.13	19.36	18.74
EC	1	20.87	18.75	17.80	17.69	17.33	17.07	16.40
	2	23.70	22.50	22.30	19.50	18.60	17.96	17.00
	Mean	22.29	20.63	20.05	18.60	17.97	17.52	16.70
ED	1	19.89	18.65	17.90	17.23	16.79	16.95	16.34
	2	21.61	19.14	18.18	17.76	17.18	16.37	16.05
	Mean	20.75	18.90	18.04	17.50	16.99	16.66	16.20
EE	1	16.68	15.25	14.67	14.69	12.80	13.63	12.23
	2	18.16	16.24	15.73	15.40	14.05	13.50	12.67
	Mean	17.42	15.75	15.20	15.05	13.43	13.57	12.45
EF	1	13.46	12.83	11.89	11.29	9.69	10.70	9.50
	2	14.16	13.27	12.60	11.50	11.90	10.30	11.00
	Mean	13.81	13.05	12.25	11.40	10.80	10.50	10.25
EG	1	16.46	15.57	14.98	15.03	14.47	14.20	13.75
	2	15.39	14.24	13.75	13.45	13.74	13.32	12.52
	Mean	15.93	14.91	14.37	14.24	14.11	13.76	13.14

Appendix IV -3 Penetration results of thick yogurt produced from different total solids
cow's milk (resault from three different areas)

Product codes	Trial	Storage time (day)						
		1	7	14	21	28	35	42
CA		400 >	400 >	400 >	400 >	400 >	400 >	400 >
CB	1	200	206	198	195	200	192	195
	2	205	212	208	193	200	195	198
	3	210	205	214	211	206	207	210
	Mean	205	208	207	200	202	198	201
	Std.er	4	3	7	8	3	6	6
CC	1	175	170	172	170	159	159	159
	2	182	165	167	162	162	165	150
	3	184	175	175	176	169	168	159
	Mean	180	170	171	169	163	164	156
	Std.er	4	4	3	6	4	4	4
CD	1	168	155	164	150	150	150	147
	2	164	158	157	158	160	157	160
	3	166	160	153	155	153	149	154
	Mean	166	158	158	154	154	152	154
	Std.er	2	2	5	3	4	4	5
CE	1	147	146	141	140	146	149	140
	2	140	145	148	148	143	142	147
	3	150	143	145	142	140	145	137
	Mean	146	145	145	143	143	145	141
	Std.er	4	1	3	3	2	3	4
CF	1	140	143	139	136	130	129	127
	2	142	139	135	128	129	124	126
	3	148	145	140	136	131	132	125
	Mean	143	142	138	133	130	128	126
	Std.er	3	2	2	4	1	3	1
CG	1	206	203	215	218	207	210	200
	2	225	232	198	195	189	199	205
	Mean	216	218	207	207	198	205	203
	Std.er	10	15	9	12	9	6	3

Appendix IV - 4 Penetration results of thick yogurt produced from different total solids
ewe's milk (result from three different areas)

Product codes	Trial	Storage time (day)						
		1	7	14	21	28	35	42
EA	1	389	388	385	369	377	376	376
	2	382	383	387	366	369	368	365
	3	377	373	364	380	362	364	356
	Mean	382	381	378	372	369	369	365
	Std.er	5	6	10	6	6	5	8
EB	1	198	191	203	189	197	187	201
	2	210	210	187	197	189	192	195
	3	196	196	199	212	210	203	189
	Mean	201	199	196	199	199	194	195
	Std.er	6	8	7	10	9	7	5
EC	1	171	176	178	181	172	170	172
	2	170	160	169	180	177	180	175
	3	178	182	173	172	168	168	168
	Mean	173	173	173	177	172	173	172
	Std.er	4	9	4	4	4	5	3
ED	1	172	152	163	162	157	153	159
	2	164	177	159	155	149	162	152
	3	165	164	171	156	164	153	151
	Mean	167	164	164	157	157	156	154
	Std.er	4	10	5	3	6	4	4
EE	1	162	157	157	156	148	144	143
	2	158	151	145	147	147	142	140
	3	147	149	153	150	145	140	140
	Mean	155	152	152	151	147	142	141
	Std.er	6	4	5	4	1	2	1
EF	1	152	145	133	136	138	143	139
	2	153	145	142	144	142	140	140
	3	143	142	147	137	143	140	138
	Mean	149	144	141	139	141	141	139
	Std.er	4	2	6	4	2	1	1
EG	1	253	234	255	240	245	240	235
	2	230	248	229	234	226	222	224
	Mean	242	241	242	237	236	231	230
	Std.er	12	7	13	3	10	9	6

Appendix V - 1 Changes of acetaldehyde for cow's milk yogurt during storage period (ppm)

Product Codes	Trial	Storage time (day)						
		1	7	14	21	28	35	42
CA	1	10.78	11.03	10.57	9.5	9.55	9.42	8.41
	2	11.13	10.7	11.02	10.45	10.3	9.68	8.62
	Mean	10.96	10.87	10.80	9.98	9.93	9.55	8.52
CB	1	15.77	15.09	14.22	13.25	12.52	12.17	13.02
	2	14.96	14.89	14.45	14.18	13.05	12.5	11.19
	Mean	15.37	14.99	14.34	13.72	12.79	12.34	12.11
CC	1	15.35	15.3	14.75	14.17	12.69	12.1	11.54
	2	15.83	15.56	14.09	13.61	13.29	12.46	12.62
	Mean	15.59	15.43	14.42	13.89	12.99	12.28	12.08
CD	1	16.63	15.87	14.85	13.45	12.96	12.45	12.17
	2	15.98	15.54	14.92	14.79	13.8	13.39	12.25
	Mean	16.31	15.71	14.89	14.12	13.38	12.92	12.21
CE	1	17.76	16.69	15.83	14.64	13.9	12.49	11.9
	2	16.33	16.93	15.58	13.98	14.12	13.71	12.86
	Mean	17.05	16.81	15.71	14.31	14.01	13.10	12.38
CF	1	18.47	18.11	17.4	16.75	14.41	13.7	11.84
	2	17.26	16.89	16.45	16.14	15.18	13.48	12.99
	Mean	17.87	17.50	16.93	16.45	14.80	13.59	12.42
CG	1	10.05	8.69	8.42	8.25	7.64	7.41	7.55
	2	9.23	9.83	9.16	8.72	8.96	8.65	8.41
	Mean	9.64	9.26	8.79	8.49	8.30	8.03	7.98

Appendix V - 2 Changes of acetaldehyde for ewe's milk yogurt during storage period (ppm)

Product Codes	Trial	Storage time (day)						
		1	7	14	21	28	35	42
EA	1	13.94	12.96	13.51	11.96	11.22	9.50	9.80
	2	14.13	12.18	12.32	12.13	10.09	8.55	10.17
	Mean	14.03	12.57	12.92	12.05	10.66	9.03	9.99
EB	1	19.86	19.52	17.48	16.06	15.75	14.93	12.39
	2	19.71	17.40	16.69	15.46	14.07	15.05	12.58
	Mean	19.79	18.46	17.09	15.76	14.91	14.99	12.49
EC	1	19.27	18.21	17.27	16.71	16.08	15.48	14.91
	2	20.35	19.19	18.58	18.16	17.70	15.23	14.59
	Mean	19.81	18.70	17.93	17.44	16.89	15.36	14.75
ED	1	21.36	20.74	19.29	18.08	17.75	17.20	16.45
	2	21.49	19.89	20.41	19.17	17.93	16.33	15.05
	Mean	21.43	20.32	19.85	18.63	17.84	16.77	15.75
EE	1	21.65	21.00	20.38	18.73	18.56	18.47	16.39
	2	21.13	20.86	19.45	19.84	19.12	17.94	16.73
	Mean	21.39	20.93	19.92	19.29	18.84	18.21	16.56
EF	1	22.73	20.85	20.26	19.45	19.33	17.89	16.74
	2	22.41	21.63	20.77	19.75	18.64	17.37	16.71
	Mean	22.57	21.24	20.52	19.60	18.99	17.63	16.73
EG	1	10.57	9.92	8.91	8.59	8.60	8.30	8.02
	2	10.78	9.60	9.05	8.87	8.50	8.25	7.95
	Mean	10.68	9.76	8.98	8.73	8.55	8.28	7.99

Appendix V - 3 Changes of acetone for cow's milk yogurt during storage period (ppm)

Product Codes	Trial	Storage time (day)						
		1	7	14	21	28	35	42
CA	1	2.62	2.75	2.63	2.60	2.64	2.51	2.36
	2	2.84	2.57	2.48	2.45	2.38	2.26	2.34
	Mean	2.73	2.66	2.56	2.53	2.51	2.39	2.35
CB	1	3.56	3.46	3.41	3.21	3.29	3.19	2.98
	2	3.57	3.43	3.36	3.41	3.25	3.24	3.14
	Mean	3.57	3.45	3.39	3.31	3.27	3.22	3.06
CC	1	3.55	3.49	3.51	3.35	3.47	3.30	3.26
	2	3.66	3.53	3.37	3.39	3.15	3.23	3.16
	Mean	3.61	3.51	3.44	3.37	3.31	3.27	3.21
CD	1	3.59	3.45	3.57	3.54	3.39	3.28	3.21
	2	3.58	3.60	3.45	3.38	3.31	3.36	3.39
	Mean	3.59	3.53	3.51	3.46	3.35	3.32	3.30
CE	1	3.55	3.56	3.55	3.51	3.39	3.47	3.37
	2	3.67	3.59	3.48	3.46	3.48	3.20	3.27
	Mean	3.61	3.58	3.52	3.49	3.44	3.34	3.32
CF	1	3.72	3.65	3.57	3.54	3.46	3.43	3.42
	2	3.58	3.67	3.58	3.47	3.31	3.23	3.16
	Mean	3.65	3.66	3.58	3.51	3.39	3.33	3.29
CG	1	2.66	2.60	2.56	2.47	2.37	2.36	2.25
	2	2.75	2.57	2.50	2.43	2.35	2.28	2.24
	Mean	2.71	2.59	2.53	2.45	2.36	2.32	2.24

Appendix V - 4 Changes of acetone for ewe's milk yogurt during storage period (ppm)

Product Codes	Trial	Storage time (day)						
		1	7	14	21	28	35	42
EA	1	2.62	2.64	2.56	2.53	2.44	2.35	2.30
	2	2.84	2.54	2.56	2.45	2.46	2.37	2.29
	Mean	2.73	2.59	2.56	2.49	2.45	2.36	2.30
EB	1	3.58	3.32	3.38	3.33	3.25	3.26	3.20
	2	3.37	3.35	3.35	3.30	3.26	3.25	3.15
	Mean	3.48	3.34	3.37	3.32	3.26	3.26	3.18
EC	1	3.45	3.40	3.41	3.28	3.23	3.09	3.19
	2	3.41	3.25	3.19	3.14	3.06	3.00	2.75
	Mean	3.43	3.33	3.30	3.21	3.15	3.05	2.97
ED	1	3.66	3.19	3.34	3.34	3.27	3.15	3.09
	2	3.35	3.48	3.25	3.15	3.15	3.09	3.07
	Mean	3.51	3.34	3.30	3.25	3.21	3.12	3.08
EE	1	3.72	3.69	3.60	3.49	3.37	3.35	3.30
	2	3.57	3.57	3.50	3.44	3.40	3.37	3.31
	Mean	3.65	3.63	3.55	3.47	3.39	3.36	3.31
EF	1	3.72	3.59	3.45	3.41	3.33	3.35	3.29
	2	3.58	3.55	3.51	3.48	3.45	3.37	3.33
	Mean	3.65	3.57	3.48	3.45	3.39	3.36	3.31
EG	1	2.58	2.31	2.28	2.30	2.27	2.30	2.33
	2	2.74	2.66	2.65	2.58	2.48	2.41	2.35
	Mean	2.66	2.49	2.47	2.44	2.38	2.36	2.34

Appendix V - 5 Changes of diacetyl for cow's milk yogurt during storage period (ppm)

Product Codes	Trial	Storage time (day)						
		1	7	14	21	28	35	42
CA	1	0.74	0.67	0.71	0.65	0.66	0.58	0.59
	2	0.66	0.72	0.65	0.63	0.60	0.57	0.55
	Mean	0.70	0.70	0.68	0.64	0.63	0.58	0.57
CB	1	0.74	0.72	0.71	0.69	0.71	0.70	0.68
	2	0.85	0.83	0.80	0.72	0.68	0.66	0.65
	Mean	0.80	0.78	0.76	0.71	0.70	0.68	0.66
CC	1	0.83	0.79	0.78	0.75	0.70	0.70	0.71
	2	0.84	0.80	0.77	0.71	0.73	0.71	0.68
	Mean	0.84	0.80	0.78	0.73	0.72	0.71	0.70
CD	1	0.86	0.80	0.77	0.75	0.74	0.70	0.73
	2	0.88	0.84	0.81	0.80	0.77	0.79	0.69
	Mean	0.87	0.82	0.79	0.78	0.76	0.75	0.71
CE	1	0.91	0.79	0.79	0.75	0.80	0.77	0.73
	2	0.86	0.90	0.85	0.86	0.75	0.75	0.71
	Mean	0.89	0.85	0.82	0.81	0.78	0.76	0.72
CF	1	0.89	0.87	0.81	0.78	0.76	0.73	0.68
	2	0.93	0.89	0.90	0.84	0.81	0.83	0.79
	Mean	0.91	0.88	0.86	0.81	0.79	0.78	0.74
CG	1	0.55	0.52	0.53	0.48	0.45	0.35	0.36
	2	0.48	0.46	0.43	0.46	0.37	0.41	0.38
	Mean	0.52	0.49	0.48	0.47	0.41	0.38	0.37

Appendix V - 6 Changes of diacetyl for ewe's milk yogurt during storage period (ppm)

Product Codes	Trials	Storage time (day)						
		1	7	14	21	28	35	42
EA	1	0.62	0.68	0.59	0.57	0.59	0.55	0.52
	2	0.70	0.62	0.64	0.65	0.60	0.58	0.55
	Mean	0.66	0.65	0.62	0.61	0.60	0.57	0.54
EB	1	0.74	0.70	0.69	0.64	0.68	0.61	0.57
	2	0.78	0.77	0.73	0.69	0.65	0.62	0.65
	Mean	0.76	0.74	0.71	0.67	0.67	0.62	0.61
EC	1	0.77	0.75	0.73	0.70	0.69	0.65	0.63
	2	0.81	0.76	0.70	0.68	0.67	0.63	0.61
	Mean	0.79	0.76	0.72	0.69	0.68	0.64	0.62
ED	1	0.81	0.83	0.79	0.69	0.68	0.65	0.67
	2	0.89	0.74	0.75	0.78	0.71	0.68	0.65
	Mean	0.85	0.79	0.77	0.74	0.70	0.67	0.66
EE	1	0.84	0.83	0.76	0.72	0.71	0.74	0.69
	2	0.87	0.78	0.81	0.77	0.73	0.65	0.73
	Mean	0.86	0.81	0.79	0.75	0.72	0.70	0.71
EF	1	0.85	0.80	0.79	0.75	0.70	0.67	0.70
	2	0.95	0.83	0.82	0.76	0.77	0.78	0.74
	Mean	0.90	0.82	0.81	0.76	0.74	0.73	0.72
EG	1	0.60	0.55	0.54	0.49	0.55	0.55	0.47
	2	0.64	0.58	0.51	0.57	0.49	0.48	0.49
	Mean	0.62	0.57	0.53	0.53	0.52	0.52	0.48

Appendix VI - 1 Free amino acid changes during storage period from
whole cow's milk yogurt (mg / 100g)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	2.237	2.723	3.282	3.824	3.471	3.951	3.887
Threoinine	1.063	2.571	2.673	3.819	3.729	2.142	3.563
Serine	2.330	2.866	3.880	3.540	4.404	4.289	2.720
Glutam. acid	6.291	5.739	8.981	8.232	7.333	8.432	7.694
Proline	4.902	6.301	6.662	6.742	5.850	5.829	6.345
Glycine	0.478	0.519	1.654	1.431	1.352	1.038	1.613
Alanine	1.599	1.145	1.996	2.434	3.057	2.086	3.110
Valine	1.856	1.419	2.494	2.823	2.209	2.930	1.556
Methionine	1.080	0.486	0.481	0.742	0.444	0.582	0.419
Isoleusin	0.800	0.735	0.999	1.523	0.712	1.497	0.403
Leusine	2.231	2.515	2.793	2.531	3.378	2.944	2.425
Tryosine	0.786	1.429	1.190	0.866	1.784	1.212	1.421
Phenylalanine	0.797	0.645	1.079	1.262	1.171	1.655	1.534
Lysine	3.290	3.585	3.998	3.164	2.651	2.474	4.739
Hystidine	1.836	1.347	1.436	1.077	2.763	1.597	2.562
Arginine	1.357	1.263	1.665	1.508	1.576	2.279	2.605
Tryptophan	0.147	0.942	0.374	0.204	0.526	0.704	0.342
Total	33.08	36.23	45.64	45.72	46.41	45.64	46.94

Appendix VI - 2 Free amino acid changes during storage period from about 24 % total solids
thick yogurt produced from fortified ultrafiltered cow's milk retentate (mg / 100 g)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	4.303	7.104	6.889	6.382	6.656	6.770	7.204
Threoinine	1.236	1.747	1.903	2.073	2.016	2.581	2.091
Serine	3.971	6.642	6.815	6.525	6.469	5.940	7.593
Glutam. acid	8.422	13.702	13.908	14.321	14.045	14.270	13.357
Proline	7.789	8.130	9.540	8.601	8.245	8.340	7.287
Glycine	1.544	2.126	2.649	2.641	2.674	2.817	2.493
Alanine	2.104	2.973	3.454	3.113	3.564	3.570	3.360
Valine	4.314	4.814	5.100	5.577	5.180	5.720	6.790
Methionine	1.732	1.441	1.947	2.443	3.628	3.658	3.726
Isoleusin	2.294	1.096	1.478	2.410	2.460	2.433	1.714
Leusine	4.472	5.148	6.806	7.857	9.453	9.220	11.073
Tryosine	2.781	3.115	2.340	2.994	3.962	3.360	4.727
Phenylalanine	2.051	2.234	2.859	3.444	3.110	3.400	3.812
Lysine	9.368	9.460	9.690	8.370	8.420	8.507	8.405
Hystidine	3.027	3.796	3.504	3.653	3.413	3.879	2.541
Arginine	3.130	3.781	5.542	5.599	5.037	5.520	5.475
Tryptophan	0.450	0.479	0.650	0.618	0.780	0.675	0.486
Total	62.99	77.79	85.07	86.62	89.11	90.66	92.13

Appendix VI - 3 Free amino acid changes during storage period from about 26 % total solids, thick yogurt produced from fortified ultrafiltered cow's milk retentate (mg / 100 g)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	6.415	6.390	9.046	9.690	10.110	9.718	10.150
Threoinine	5.560	7.230	7.740	7.870	7.970	8.563	9.370
Serine	8.699	10.351	10.992	10.330	10.050	10.310	10.850
Glutam. acid	15.490	18.080	18.720	19.600	18.370	19.830	18.030
Proline	11.280	16.910	16.660	16.350	16.850	15.728	15.200
Glycine	1.788	3.255	2.768	3.350	3.129	2.434	2.820
Alanine	2.375	2.647	3.394	3.370	3.980	4.928	4.199
Valine	5.299	5.660	5.990	7.260	8.390	7.997	7.630
Methionine	2.117	3.552	3.372	2.930	3.350	3.961	3.344
Isoleusin	1.584	2.622	2.117	2.050	3.100	3.606	2.841
Leusine	4.147	5.330	5.710	5.683	4.470	4.580	6.060
Tryosine	2.482	4.007	4.754	5.480	5.560	5.054	4.890
Phenylalanine	1.692	4.096	4.054	3.950	4.570	4.410	4.455
Lysine	4.250	5.030	5.590	6.950	5.729	5.970	5.140
Hystidine	3.600	3.180	3.980	3.970	3.280	3.150	3.262
Arginine	4.232	5.530	4.640	4.150	4.741	4.350	4.330
Tryptophan	0.954	0.750	0.640	0.740	0.434	0.458	0.558
Total	81.97	104.62	110.17	113.72	114.08	115.04	113.13

Appendix VI - 4 Free amino acid changes during storage period from about 28 % total solids thick yogurt produced from fortified ultrafiltered cow's milk retentate (mg / 100 g)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	4.148	4.797	7.748	7.550	8.776	9.122	7.846
Threoinine	4.026	5.890	6.697	6.664	7.220	7.937	6.783
Serine	4.674	7.823	7.261	7.109	7.423	8.277	8.687
Glutam. acid	15.020	20.510	19.390	20.216	19.908	20.896	20.178
Proline	12.976	13.758	15.603	14.424	14.201	12.338	14.762
Glycine	1.808	1.740	3.355	3.496	3.491	3.855	4.249
Alanine	3.752	3.077	4.974	4.913	4.191	4.175	5.392
Valine	4.292	5.382	5.088	5.790	5.578	6.517	7.036
Methionine	1.956	2.388	4.492	3.234	3.587	3.083	3.853
Isoleusin	3.961	4.851	2.799	5.110	5.505	4.783	5.217
Leusine	4.454	5.982	7.520	7.321	7.763	7.244	7.714
Tryosine	4.415	6.899	4.440	5.827	5.890	5.608	5.181
Phenylalanine	3.871	3.799	4.696	4.345	4.493	3.861	3.247
Lysine	5.853	6.145	7.106	7.844	8.097	8.455	8.194
Hystidine	2.163	3.636	3.720	3.671	3.888	3.247	3.648
Arginine	3.233	6.012	5.779	6.242	5.668	7.308	6.115
Tryptophan	0.342	0.523	0.666	0.751	0.978	0.955	0.626
Total	80.95	103.21	111.34	114.51	116.66	117.66	118.73

Appendix VI - 5 Free amino acid changes during storage period from about 30 % total solids thick yogurt produced from fortified ultrafiltered cow's milk retentate (mg / 100 g)

Amino acid name	Storage period of yogurt (day)						
	1	7	14	21	28	35	42
Aspar.acid	8.884	7.052	9.265	10.842	10.021	9.064	10.694
Threonine	4.013	2.875	2.507	6.038	6.497	6.530	6.953
Serine	7.770	9.872	10.627	11.058	11.917	10.894	10.364
Glutam. acid	15.188	16.887	19.586	18.126	18.854	18.835	18.254
Proline	12.171	14.754	16.844	16.038	16.384	16.165	16.036
Glycine	2.890	3.229	3.860	3.375	3.660	3.558	3.136
Alanine	4.485	5.687	4.287	5.421	4.803	4.665	4.122
Valine	2.818	3.205	3.895	3.013	3.977	3.419	3.715
Methionine	2.579	2.267	2.255	2.960	2.820	2.296	2.744
Isoleusin	4.706	5.534	5.843	5.002	5.725	6.846	7.391
Leusine	1.286	2.436	2.903	2.403	2.158	2.597	2.577
Tryosine	1.715	3.845	3.319	2.152	2.931	3.074	3.402
Phenylalanine	6.279	6.264	7.824	7.979	8.015	8.826	8.760
Lysine	1.289	2.870	2.004	3.306	3.040	3.641	2.681
Hystidine	4.424	8.737	8.924	8.272	7.184	6.063	6.035
Arginine	1.570	1.920	1.689	1.972	1.588	1.927	1.343
Tryptophan	0.345	0.391	0.456	0.399	0.521	0.614	0.458
Total	82.07	97.43	105.63	107.96	109.57	108.40	108.21

Appendix VI - 6 Free amino acid changes during storage period from about 32 % total solids
thick yogurt produced from fortified ultrafiltered cow's milk retentate (mg / 100 g)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	6.705	8.247	9.187	9.214	8.943	9.049	11.216
Threoinine	4.269	7.370	7.925	7.336	6.985	6.652	9.375
Serine	6.947	8.157	9.483	9.105	7.528	7.181	7.663
Glutam. acid	12.248	15.198	14.517	14.975	15.063	14.294	15.639
Proline	9.559	11.459	10.609	12.076	12.648	12.745	12.554
Glycine	1.830	2.647	3.968	4.872	3.018	3.205	4.615
Alanine	4.751	2.493	3.087	3.153	4.790	4.705	3.298
Valine	7.736	8.842	8.841	7.961	7.585	6.996	7.097
Methionine	1.473	2.275	2.149	2.595	2.751	3.672	3.215
Isoleusin	1.816	2.489	3.168	1.511	2.987	3.639	2.841
Leusine	3.250	7.727	7.151	6.367	7.571	8.326	9.581
Tryosine	3.435	3.949	3.525	3.146	4.124	4.634	4.074
Phenylalanine	4.041	2.997	4.003	5.399	4.258	5.080	4.318
Lysine	8.479	9.538	8.766	8.143	8.658	8.036	7.399
Hystidine	2.833	2.570	2.630	2.410	2.799	3.150	3.502
Arginine	5.615	7.080	8.904	8.466	9.447	8.146	8.304
Tryptophan	1.542	0.675	0.651	1.281	1.435	1.434	0.967
Total	86.53	103.71	108.56	108.01	110.59	110.94	115.66

Appendix VI - 7 Free amino acid changes during storage period from stirred style thick yogurt (CG coded) produced from cow's milk (mg / 100 g)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	3.390	4.671	4.264	4.448	4.152	4.891	5.713
Threoinine	2.440	2.971	3.501	3.739	4.854	4.591	3.997
Serine	3.650	3.128	3.501	3.739	3.341	4.893	4.050
Glutam. acid	8.370	10.176	10.272	10.921	10.410	11.419	11.458
Proline	6.085	6.925	5.499	6.730	6.006	6.282	7.308
Glycine	1.740	0.871	1.297	1.792	1.346	1.813	1.240
Alanine	3.940	3.661	5.987	4.514	5.565	4.289	4.688
Valine	2.040	1.724	1.795	1.869	4.010	1.672	2.082
Methionine	0.410	0.428	0.266	0.448	0.628	0.351	0.347
Isoleusin	1.005	0.860	0.934	1.106	1.101	0.756	1.097
Leusine	3.488	2.640	2.637	3.495	3.155	2.783	3.090
Tryosine	1.005	1.756	0.869	1.379	1.273	1.395	1.716
Phenylalanine	1.250	1.177	1.138	1.904	1.721	1.858	1.341
Lysine	5.030	4.789	5.957	5.251	5.692	5.826	5.465
Hystidine	1.890	1.619	1.552	2.338	3.054	1.985	2.043
Arginine	2.039	2.166	2.422	3.244	3.897	2.422	2.552
Tryptophan	0.430	0.546	0.541	0.640	0.570	0.439	0.418
Total	48.20	50.11	52.43	57.56	60.78	57.67	58.61

Appendix VI - 8 Free amino acid changes during storage period from whole ewe's milk yogurt (mg / 100 g)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	4.498	5.734	6.818	7.495	6.833	6.398	5.040
Threoinine	2.149	4.435	4.742	4.514	4.320	4.510	4.171
Serine	3.976	4.513	4.733	4.582	4.941	4.556	3.717
Glutam. acid	9.108	14.961	14.968	15.652	15.941	14.753	12.875
Proline	8.092	13.541	12.328	12.612	12.773	12.319	11.670
Glycine	0.436	1.601	1.396	1.257	1.060	0.941	0.744
Alanine	4.103	5.221	5.346	4.053	4.742	4.479	4.475
Valine	2.532	2.804	2.568	2.512	2.095	1.706	0.259
Methionine	0.453	0.259	0.245	0.315	0.197	0.221	0.795
Isoleusin	0.766	1.173	1.199	1.569	0.760	0.746	0.807
Leusine	1.704	3.262	3.325	3.729	3.395	3.934	3.262
Tryosine	0.915	1.058	1.122	1.114	1.120	1.127	1.221
Phenylalanine	0.705	1.236	1.191	1.018	0.845	0.714	1.271
Lysine	3.236	3.902	4.767	3.608	4.243	3.692	4.175
Hystidine	1.671	2.484	2.710	2.154	2.133	1.955	2.046
Arginine	3.326	3.189	3.036	2.860	3.445	2.749	3.521
Tryptophan	0.590	1.144	1.347	1.169	1.669	1.566	0.925
Total	48.26	70.52	71.84	70.21	70.51	66.37	60.98

Appendix VI - 9 Free amino acid changes during storage period from about 24 % total solids thick yogurt produced from fortified ultrafiltered ewe's milk retentate (mg / 100 g)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	6.23	7.795	6.702	7.784	7.007	7.127	7.421
Threonine	5.29	5.327	5.779	5.688	5.265	6.160	5.257
Serine	6.32	5.260	6.825	7.084	5.646	6.088	6.251
Glutam. acid	15.09	14.431	17.219	18.217	16.658	17.848	14.656
Proline	14.50	13.274	13.587	12.516	13.541	14.029	11.322
Glycine	1.07	2.426	1.155	1.685	1.525	0.831	1.407
Alanine	1.71	1.965	3.199	3.072	3.414	4.393	4.902
Valine	2.70	3.263	2.445	2.987	2.190	2.946	3.668
Methionine	0.57	0.769	0.648	1.437	1.511	0.975	3.120
Isoleusin	1.11	1.925	1.898	2.257	1.809	1.132	2.033
Leusine	2.74	4.868	3.077	3.977	3.403	3.873	5.046
Tryosine	0.93	1.544	0.960	0.542	0.184	1.362	2.990
Phenylalanine	0.78	1.846	1.335	2.606	2.178	1.173	3.231
Lysine	4.11	5.349	5.015	4.573	4.609	4.688	4.492
Hystidine	2.63	5.644	2.909	2.482	2.639	2.550	3.835
Arginine	2.34	4.852	3.443	3.789	4.951	3.217	4.589
Tryptophan	1.42	1.871	1.250	1.414	0.992	1.410	1.171
Total	69.54	82.41	77.45	82.11	77.52	79.80	85.39

Appendix VI - 10 Free amino acid changes during storage period from about 26 % total solids thick yogurt produced from fortified ultrafiltered ewe's milk retentate (mg / 100 g.)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	7.111	9.510	8.650	8.170	7.525	7.320	7.720
Threoinine	4.155	5.544	6.481	5.343	7.042	6.967	6.189
Serine	5.250	6.136	7.168	6.747	6.121	8.935	8.282
Glutam. acid	17.520	19.372	19.130	17.759	15.740	18.730	17.952
Proline	14.177	17.587	15.322	11.846	13.786	13.013	11.289
Glycine	1.620	1.090	1.031	1.542	1.424	1.547	1.518
Alanine	3.060	3.814	4.397	8.808	4.668	5.399	5.443
Valine	2.440	2.310	3.851	3.701	3.440	4.259	4.623
Methionine	0.441	0.673	1.395	2.481	2.321	3.953	2.095
Isoleusin	1.750	0.619	1.686	1.933	1.920	2.635	2.930
Leusine	4.890	6.886	5.021	5.620	5.680	6.115	7.028
Tryosine	1.035	1.311	1.818	2.441	2.100	2.200	4.864
Phenylalanine	1.580	1.590	1.237	2.817	2.928	2.505	2.933
Lysine	3.585	5.069	5.801	7.540	6.940	6.458	5.860
Hystidine	2.646	3.357	3.528	3.360	4.070	4.360	5.590
Arginine	3.652	3.298	4.452	5.540	4.480	4.773	4.887
Tryptophan	0.615	0.644	0.800	0.410	0.780	1.495	1.005
Total	68.42	79.30	83.12	87.89	83.44	93.34	92.49

Appendix VI - 11 Free amino acid changes during storage period from about 28 % total solids thick yogurt produced from fortified ultrafiltered ewe's milk retentate (mg / 100 g)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	5.744	7.337	7.230	8.628	9.116	9.745	10.231
Threoinine	4.808	5.205	5.624	5.257	5.472	5.164	8.265
Serine	6.419	6.287	6.572	8.261	8.131	8.887	8.724
Glutam. acid	14.877	17.989	16.976	17.738	18.038	18.730	19.350
Proline	11.085	12.329	12.067	12.188	12.068	12.635	12.877
Glycine	0.716	1.261	1.391	2.079	1.203	1.652	1.330
Alanine	4.920	4.083	4.347	5.794	6.348	5.877	6.692
Valine	3.098	3.952	5.948	5.289	4.874	5.594	6.042
Methionine	0.943	1.332	2.418	2.174	2.797	2.701	3.845
Isoleusin	2.150	2.955	4.358	5.333	3.256	3.278	3.519
Leusine	5.864	4.734	6.728	7.093	7.620	8.519	8.588
Tryosine	1.218	1.477	1.880	2.252	2.317	2.519	3.872
Phenylalanine	0.875	1.216	1.634	2.157	2.060	2.860	3.179
Lysine	5.313	7.511	7.626	7.724	7.632	7.102	6.830
Hystidine	2.940	3.962	4.985	5.614	5.967	5.697	4.779
Arginine	4.106	5.556	6.944	5.232	5.929	5.664	5.221
Tryptophan	0.949	0.954	0.438	0.000	1.158	1.156	1.238
Total	76.03	88.14	97.17	102.81	103.99	107.78	114.58

Appendix VI - 12 Free amino acid changes during storage period from about 30 % total solids thick yogurt produced from fortified ultrafiltered ewe's milk retentate (mg / 100 g)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	7.676	10.508	9.624	10.919	10.961	10.138	10.731
Threoinine	6.717	8.054	8.090	8.692	8.528	8.490	9.074
Serine	7.688	9.166	9.108	9.718	11.265	11.526	10.605
Glutam. acid	17.320	19.054	18.925	18.552	19.684	19.942	20.567
Proline	14.339	14.905	14.093	14.129	13.280	13.998	14.846
Glycine	0.734	0.933	1.463	1.357	1.116	1.929	1.047
Alanine	3.345	4.554	5.991	6.376	7.102	6.487	6.336
Valine	4.089	5.295	5.273	5.847	6.628	6.190	7.724
Methionine	0.732	1.769	2.070	2.586	3.373	3.586	3.258
Isoleusin	1.138	1.508	1.804	2.581	2.086	2.028	2.503
Leusine	2.129	3.827	5.366	5.243	6.776	6.185	5.421
Tryosine	1.081	1.317	2.204	2.714	2.273	2.575	2.506
Phenylalanine	1.054	1.087	2.164	2.745	2.297	2.594	2.282
Lysine	6.026	7.180	7.503	7.641	7.118	7.069	7.407
Hystidine	4.032	4.287	4.213	3.590	4.214	7.622	5.610
Arginine	3.649	6.012	6.112	7.367	6.416	6.353	7.717
Tryptophan	1.344	1.141	1.237	0.878	2.177	1.647	0.855
Total	83.09	100.60	105.24	110.93	115.30	118.36	118.49

Appendix VI - 13 Free amino acid changes during storage period from about 32 % total solids thick yogurt produced from fortified ultrafiltered ewe's milk retentate (mg / 100 g.)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	8.566	8.423	10.460	11.351	10.551	10.772	10.220
Threonine	5.814	5.463	6.995	9.664	8.180	8.903	9.611
Serine	6.382	5.808	9.024	10.269	9.660	9.182	9.324
Glutam. acid	19.912	19.580	17.099	18.795	19.993	20.121	20.065
Proline	17.387	18.217	15.799	14.002	15.193	15.176	14.916
Glycine	1.626	0.816	1.308	1.258	2.097	1.558	2.344
Alanine	1.748	2.568	4.602	5.025	4.963	5.586	5.722
Valine	3.732	3.381	4.944	5.474	5.902	6.077	6.396
Methionine	0.191	0.717	0.997	1.945	2.016	2.785	3.368
Isoleusin	1.344	2.140	2.902	2.016	2.262	3.289	3.692
Leusine	2.769	3.655	4.254	5.616	7.093	6.862	6.363
Tyrosine	0.692	0.792	1.187	1.543	2.828	2.986	3.739
Phenylalanine	1.320	1.693	1.257	1.840	3.263	3.382	2.909
Lysine	5.832	5.473	8.378	8.523	6.689	5.721	7.221
Hystidine	3.336	5.026	5.453	5.439	6.418	6.123	6.785
Arginine	3.164	3.104	5.531	5.797	5.853	5.230	5.516
Tryptophan	1.026	0.894	0.605	0.788	1.085	1.002	1.637
Total	84.84	87.75	100.80	109.35	114.05	114.75	119.83

Appendix VI - 14 Free amino acid changes during storage period in stirred style thick yogurt (EG coded) produced from ewe's milk (mg / 100 g)

Amino acid name	Storage time (day)						
	1	7	14	21	28	35	42
Aspar.acid	3.390	4.671	4.264	4.448	4.152	4.891	4.713
Threoinine	2.440	2.971	3.501	3.739	4.854	4.591	3.997
Serine	3.650	3.128	3.501	3.739	3.341	4.893	4.050
Glutam. acid	8.370	8.176	8.272	8.921	8.410	7.419	8.458
Proline	6.085	6.925	5.499	6.730	6.006	5.282	5.308
Glycine	1.740	6.925	1.297	1.792	1.346	1.813	1.240
Alanine	3.940	3.661	5.987	4.514	5.565	4.289	4.688
Valine	2.040	2.436	3.684	4.419	4.839	3.517	3.741
Methionine	1.460	1.120	1.075	1.142	1.343	1.097	1.546
Isoleusin	1.754	2.191	2.207	2.798	3.084	1.178	1.355
Leusine	3.488	4.383	5.905	7.950	5.510	5.318	5.969
Tryosine	2.293	1.756	2.409	3.287	3.569	1.510	1.661
Phenylalanine	3.214	2.177	3.076	3.176	3.254	3.752	3.297
Lysine	5.030	6.362	5.957	6.251	5.692	5.826	5.465
Hystidine	4.422	3.414	3.637	4.061	5.712	2.675	2.463
Arginine	5.468	3.453	4.679	6.372	6.782	3.973	3.946
Tryptophan	0.430	0.546	0.541	0.640	0.570	0.690	0.562
Total	59.214	64.295	65.491	73.979	74.029	62.714	62.459

Appendix VII - 1 Some microbiological changes in set and stirred style thick yogurts made from cow's milk using different production process during storage period at 5 C

Product cads	Microorganisms cfu/gr	Trials	Storage period (day)							Dead Bact. %
			1	7	14	21	28	35	42	
CA	<i>S. thermophilus</i>	1	5.5E+08	5.3E+08	4.4E+08	2.2E+08	1.0E+08	6.0E+07	4.0E+07	92.73
		2	4.9E+08	5.5E+08	3.8E+08	1.5E+08	9.0E+07	8.0E+07	7.0E+07	85.71
		Mean	5.2E+08	5.4E+08	4.1E+08	1.9E+08	9.5E+07	7.0E+07	5.5E+07	89.42
	<i>L. bulgaricus</i>	1	4.3E+08	4.1E+08	2.4E+08	1.1E+08	5.0E+07	1.3E+07	1.1E+07	97.44
		2	2.9E+08	3.6E+08	2.1E+08	7.0E+07	9.0E+07	7.2E+07	6.0E+07	79.31
		Mean	3.6E+08	3.9E+08	2.3E+08	9.0E+07	7.0E+07	4.3E+07	3.6E+07	90.14
	Total count	1	9.6E+08	8.5E+08	7.0E+08	3.8E+08	2.5E+08	9.3E+07	6.1E+07	93.65
		2	7.8E+08	6.2E+08	5.7E+08	3.2E+08	1.8E+08	1.4E+08	1.1E+08	85.90
		Mean	8.7E+08	7.4E+08	6.4E+08	3.5E+08	2.2E+08	1.2E+08	8.6E+07	90.17
CB	<i>S. thermophilus</i>	1	5.7E+08	3.2E+08	2.1E+08	1.8E+08	9.6E+07	7.9E+07	5.6E+07	90.18
		2	5.0E+08	4.8E+08	3.2E+08	2.3E+08	1.7E+08	1.1E+08	7.4E+07	85.20
		Mean	5.4E+08	4.0E+08	2.7E+08	2.1E+08	1.3E+08	9.7E+07	6.5E+07	87.85
	<i>L. bulgaricus</i>	1	3.1E+08	2.3E+08	2.2E+08	1.5E+08	1.1E+08	9.5E+07	6.5E+07	79.03
		2	4.5E+08	3.7E+08	2.5E+08	1.8E+08	1.0E+08	8.5E+07	5.3E+07	88.22
		Mean	3.8E+08	3.0E+08	2.4E+08	1.7E+08	1.0E+08	9.0E+07	5.9E+07	84.47
	Total count	1	8.8E+08	6.3E+08	4.3E+08	3.2E+08	1.8E+08	1.7E+08	1.2E+08	86.25
		2	9.1E+08	8.1E+08	5.7E+08	4.3E+08	2.9E+08	2.0E+08	1.3E+08	86.04
		Mean	9.0E+08	7.2E+08	5.0E+08	3.8E+08	2.4E+08	1.9E+08	1.2E+08	86.15
CC	<i>S. thermophilus</i>	1	6.2E+08	5.9E+08	4.5E+08	4.2E+08	2.5E+08	1.8E+08	1.0E+08	83.39
		2	4.9E+08	3.5E+08	2.7E+08	2.5E+08	1.6E+08	9.7E+07	7.2E+07	85.31
		Mean	5.6E+08	4.7E+08	3.6E+08	3.4E+08	2.0E+08	1.4E+08	8.8E+07	84.23
	<i>L. bulgaricus</i>	1	2.1E+08	2.0E+08	1.6E+08	1.5E+08	1.3E+08	1.0E+08	8.4E+07	60.00
		2	3.1E+08	2.3E+08	1.8E+08	1.2E+08	9.6E+07	7.5E+07	5.5E+07	82.26
		Mean	2.6E+08	2.2E+08	1.7E+08	1.4E+08	1.1E+08	8.8E+07	7.0E+07	73.27
	Total count	1	8.6E+08	7.9E+08	6.1E+08	5.4E+08	3.8E+08	2.7E+08	1.9E+08	78.26
		2	7.5E+08	5.8E+08	4.5E+08	3.7E+08	2.5E+08	1.7E+08	1.3E+08	83.07
		Mean	8.1E+08	6.9E+08	5.3E+08	4.6E+08	3.2E+08	2.2E+08	1.6E+08	80.50
CD	<i>S. thermophilus</i>	1	1.1E+09	6.9E+08	3.3E+08	2.2E+08	1.2E+08	1.1E+08	1.0E+08	90.91
		2	9.8E+08	7.2E+08	6.1E+08	4.1E+08	2.0E+08	9.2E+07	8.0E+07	91.84
		Mean	1.0E+09	7.1E+08	4.7E+08	3.2E+08	1.6E+08	1.0E+08	9.0E+07	91.35
	<i>L. bulgaricus</i>	1	8.1E+08	7.0E+08	5.6E+08	3.7E+08	2.5E+08	1.2E+08	9.2E+07	88.64
		2	7.0E+08	5.0E+08	4.0E+08	2.8E+08	1.7E+08	1.1E+08	8.9E+07	87.29
		Mean	7.6E+08	6.0E+08	4.8E+08	3.3E+08	2.1E+08	1.2E+08	9.1E+07	88.01
	Total count	1	2.0E+09	1.6E+09	1.0E+09	8.9E+08	5.7E+08	3.6E+08	2.1E+08	89.50
		2	1.6E+09	1.2E+09	9.6E+08	6.9E+08	3.7E+08	2.0E+08	1.7E+08	89.44
		Mean	1.8E+09	1.4E+09	9.8E+08	7.9E+08	4.7E+08	2.8E+08	1.9E+08	89.47
CE	<i>S. thermophilus</i>	1	1.9E+09	1.5E+09	7.7E+08	5.5E+08	3.9E+08	2.5E+08	1.9E+08	90.00
		2	1.2E+09	9.0E+08	6.9E+08	4.7E+08	2.7E+08	1.5E+08	9.0E+07	92.50
		Mean	1.6E+09	1.2E+09	7.3E+08	5.1E+08	3.3E+08	2.0E+08	1.4E+08	90.97
	<i>L. bulgaricus</i>	1	7.0E+08	5.0E+08	5.1E+08	3.9E+08	1.7E+08	1.5E+08	1.2E+08	83.00
		2	8.0E+08	5.0E+08	4.3E+08	3.6E+08	2.9E+08	1.7E+08	1.3E+08	84.25
		Mean	7.5E+08	5.0E+08	4.7E+08	3.8E+08	2.3E+08	1.6E+08	1.2E+08	83.67
	Total count	1	2.3E+09	1.9E+09	1.4E+09	9.7E+08	6.5E+08	4.0E+08	3.1E+08	86.57
		2	1.9E+09	1.3E+09	1.0E+09	8.0E+08	5.6E+08	3.2E+08	2.2E+08	88.63
		Mean	2.1E+09	1.6E+09	1.2E+09	8.9E+08	6.1E+08	3.6E+08	2.6E+08	87.50
CF	<i>S. thermophilus</i>	1	1.0E+09	8.0E+08	8.0E+08	3.5E+08	2.6E+08	2.0E+08	1.9E+08	81.00
		2	1.3E+09	7.5E+08	6.0E+08	3.3E+08	3.3E+08	2.4E+08	9.0E+07	93.08
		Mean	1.2E+09	7.8E+08	7.0E+08	3.4E+08	3.0E+08	2.2E+08	1.4E+08	87.83
	<i>L. bulgaricus</i>	1	9.0E+08	8.0E+08	6.0E+08	4.3E+08	2.4E+08	1.8E+08	1.2E+08	86.67
		2	8.5E+08	6.5E+08	4.3E+08	2.7E+08	1.6E+08	1.5E+08	8.0E+07	90.59
		Mean	8.8E+08	7.3E+08	5.2E+08	3.5E+08	2.0E+08	1.7E+08	1.0E+08	88.57
	Total count	1	1.8E+09	1.6E+09	1.3E+09	9.1E+08	6.1E+08	3.8E+08	3.1E+08	82.78
		2	2.0E+09	1.5E+09	1.2E+09	8.5E+08	5.3E+08	3.9E+08	2.1E+08	89.50
		Mean	1.9E+09	1.6E+09	1.3E+09	8.8E+08	5.7E+08	3.9E+08	2.6E+08	86.32
CG	<i>S. thermophilus</i>	1	4.8E+08	3.0E+08	2.6E+08	2.0E+08	1.4E+08	1.3E+08	1.2E+08	75.00
		2	3.7E+08	3.6E+08	3.3E+08	3.0E+08	2.4E+08	1.5E+08	1.0E+08	72.97
		Mean	4.3E+08	3.3E+08	3.0E+08	2.5E+08	1.9E+08	1.4E+08	1.1E+08	74.12
	<i>L. bulgaricus</i>	1	1.9E+08	1.8E+08	2.0E+08	1.6E+08	1.5E+08	1.2E+08	7.0E+07	63.16
		2	4.3E+08	2.5E+08	2.2E+08	1.9E+08	1.4E+08	1.0E+08	9.0E+07	79.07
		Mean	3.1E+08	2.2E+08	2.1E+08	1.8E+08	1.5E+08	1.1E+08	8.0E+07	74.19
	Total count	1	6.6E+08	5.8E+08	4.6E+08	3.9E+08	2.9E+08	2.4E+08	2.1E+08	68.18
		2	8.2E+08	6.8E+08	5.5E+08	4.5E+08	3.6E+08	2.6E+08	1.8E+08	78.05
		Mean	7.4E+08	6.3E+08	5.1E+08	4.2E+08	3.3E+08	2.5E+08	2.0E+08	73.65

Appendix VII - 2 Some microbiological changes in set and stirred style thick yogurts made from ewe's milk using different production process during storage period at 5 C

Product cods	Microorganisms cfu/g	Trial	Storage period (Day)							Dead Bact. %
			1	7	14	21	28	35	42	
EA	<i>S. thermophilus</i>	1	6.1E+08	5.7E+08	5.4E+08	4.5E+08	3.0E+08	7.0E+07	5.9E+07	90.33
		2	6.9E+08	6.2E+08	6.4E+08	3.7E+08	2.2E+08	9.1E+07	7.3E+07	89.42
		Mean	6.5E+08	6.0E+08	5.9E+08	4.1E+08	2.6E+08	8.1E+07	6.6E+07	89.85
	<i>L. bulgaricus</i>	1	3.0E+08	2.1E+08	1.7E+08	1.0E+08	8.7E+07	7.5E+07	6.7E+07	77.67
		2	3.5E+08	2.6E+08	2.2E+08	1.7E+08	1.1E+08	9.2E+07	7.4E+07	78.86
		Mean	3.3E+08	2.4E+08	2.0E+08	1.4E+08	9.9E+07	8.4E+07	7.1E+07	78.31
	Total count	1	9.3E+08	7.7E+08	7.2E+08	5.7E+08	3.9E+08	2.0E+08	1.3E+08	86.45
		2	1.1E+09	9.0E+08	8.3E+08	6.0E+08	3.3E+08	2.4E+08	1.9E+08	82.73
		Mean	1.0E+09	8.4E+08	7.8E+08	5.9E+08	3.6E+08	2.2E+08	1.6E+08	84.43
EB	<i>S. thermophilus</i>	1	6.5E+08	6.2E+08	5.5E+08	4.3E+08	3.8E+08	1.8E+08	1.2E+08	81.54
		2	7.6E+08	6.9E+08	4.3E+08	3.8E+08	3.0E+08	1.1E+08	9.5E+07	87.50
		Mean	7.1E+08	6.6E+08	4.9E+08	4.1E+08	3.4E+08	1.4E+08	1.1E+08	84.75
	<i>L. bulgaricus</i>	1	2.3E+08	1.9E+08	1.1E+08	1.1E+08	9.3E+07	9.1E+07	5.0E+07	78.26
		2	3.3E+08	2.8E+08	2.3E+08	2.1E+08	1.8E+08	1.2E+08	1.1E+08	66.67
		Mean	2.8E+08	2.4E+08	1.7E+08	1.6E+08	1.4E+08	1.1E+08	8.0E+07	71.43
	Total count	1	8.9E+08	8.2E+08	7.1E+08	5.4E+08	4.7E+08	3.1E+08	2.1E+08	76.40
		2	1.0E+09	9.7E+08	6.8E+08	5.9E+08	4.8E+08	2.4E+08	1.9E+08	81.00
		Mean	9.5E+08	9.0E+08	7.0E+08	5.7E+08	4.8E+08	2.8E+08	2.0E+08	78.84
EC	<i>S. thermophilus</i>	1	6.8E+08	6.6E+08	6.0E+08	5.9E+08	4.3E+08	1.0E+08	9.0E+07	86.76
		2	7.4E+08	6.9E+08	5.8E+08	4.9E+08	4.8E+08	1.4E+08	9.5E+07	87.16
		Mean	7.1E+08	6.8E+08	5.9E+08	5.4E+08	4.6E+08	1.2E+08	9.3E+07	86.97
	<i>L. bulgaricus</i>	1	3.2E+08	2.9E+08	2.5E+08	2.4E+08	1.8E+08	1.0E+08	9.6E+07	70.00
		2	2.6E+08	2.8E+08	2.1E+08	1.9E+08	2.0E+08	2.3E+08	1.0E+08	61.54
		Mean	2.9E+08	2.9E+08	2.3E+08	2.2E+08	1.9E+08	1.7E+08	9.8E+07	66.21
	Total count	1	1.1E+09	9.6E+08	8.6E+08	8.3E+08	6.4E+08	3.0E+08	1.9E+08	83.09
		2	1.0E+09	9.5E+08	7.9E+08	6.8E+08	6.8E+08	3.7E+08	2.0E+08	80.50
		Mean	1.1E+09	9.6E+08	8.3E+08	7.6E+08	6.6E+08	3.4E+08	1.9E+08	81.86
ED	<i>S. thermophilus</i>	1	6.5E+08	5.7E+08	5.7E+08	4.1E+08	3.8E+08	3.9E+08	2.1E+08	67.69
		2	7.4E+08	7.4E+08	6.4E+08	5.7E+08	4.0E+08	3.5E+08	2.9E+08	60.81
		Mean	7.0E+08	6.6E+08	6.1E+08	4.9E+08	3.9E+08	3.7E+08	2.5E+08	64.03
	<i>L. bulgaricus</i>	1	3.3E+08	3.1E+08	2.9E+08	2.3E+08	1.9E+08	1.7E+08	8.0E+07	75.76
		2	4.7E+08	3.6E+08	2.3E+08	1.6E+08	1.7E+08	1.3E+08	1.5E+08	68.09
		Mean	4.0E+08	3.4E+08	2.6E+08	2.0E+08	1.8E+08	1.5E+08	1.2E+08	71.25
	Total count	1	9.7E+08	8.8E+08	8.6E+08	6.4E+08	5.7E+08	5.6E+08	2.9E+08	70.10
		2	1.3E+09	1.1E+09	8.9E+08	7.3E+08	6.0E+08	5.1E+08	4.4E+08	66.15
		Mean	1.1E+09	9.9E+08	8.8E+08	6.9E+08	5.9E+08	5.4E+08	3.7E+08	67.84
EE	<i>S. thermophilus</i>	1	7.8E+08	7.7E+08	6.8E+08	4.1E+08	3.7E+08	2.6E+08	2.4E+08	69.49
		2	6.6E+08	6.7E+08	5.5E+08	5.1E+08	4.7E+08	3.2E+08	2.2E+08	67.12
		Mean	7.2E+08	7.2E+08	6.2E+08	4.6E+08	4.2E+08	2.9E+08	2.3E+08	68.40
	<i>L. bulgaricus</i>	1	4.5E+08	3.3E+08	3.4E+08	3.0E+08	2.7E+08	1.9E+08	1.6E+08	64.00
		2	3.8E+08	2.7E+08	2.4E+08	1.8E+08	1.7E+08	1.6E+08	1.4E+08	63.16
		Mean	4.2E+08	3.0E+08	2.9E+08	2.4E+08	2.2E+08	1.8E+08	1.5E+08	63.61
	Total count	1	1.3E+09	1.1E+09	9.7E+08	7.4E+08	6.3E+08	4.5E+08	3.5E+08	73.08
		2	9.9E+08	9.4E+08	7.9E+08	6.5E+08	6.0E+08	4.8E+08	2.7E+08	72.73
		Mean	1.1E+09	1.0E+09	8.8E+08	7.0E+08	6.2E+08	4.6E+08	3.1E+08	72.93
EF	<i>S. thermophilus</i>	1	1.5E+09	1.1E+09	9.5E+08	8.9E+08	6.4E+08	6.0E+08	3.1E+08	79.33
		2	3.1E+09	2.8E+09	1.7E+09	1.1E+09	9.5E+08	7.6E+08	3.9E+08	87.42
		Mean	2.3E+09	2.0E+09	1.3E+09	1.0E+09	7.9E+08	6.8E+08	3.5E+08	84.78
	<i>L. bulgaricus</i>	1	9.3E+08	5.1E+08	3.8E+08	3.0E+08	2.7E+08	2.8E+08	2.0E+08	78.49
		2	6.7E+08	4.2E+08	3.4E+08	2.7E+08	2.5E+08	2.2E+08	2.6E+08	61.19
		Mean	8.0E+08	4.7E+08	3.6E+08	2.9E+08	2.6E+08	2.5E+08	2.3E+08	71.25
	Total count	1	2.5E+09	1.7E+09	1.3E+09	1.0E+09	9.1E+08	8.8E+08	5.1E+08	79.60
		2	3.7E+09	3.1E+09	1.8E+09	1.2E+09	9.8E+08	8.7E+08	6.5E+08	82.43
		Mean	3.1E+09	2.4E+09	1.6E+09	1.1E+09	9.5E+08	8.8E+08	5.8E+08	81.29
EG	<i>S. thermophilus</i>	1	6.9E+08	5.7E+08	4.6E+08	4.0E+08	3.1E+08	2.2E+08	1.9E+08	72.46
		2	4.2E+08	4.3E+08	4.2E+08	3.4E+08	2.7E+08	1.6E+08	1.5E+08	64.29
		Mean	5.6E+08	5.0E+08	4.4E+08	3.7E+08	2.9E+08	1.9E+08	1.7E+08	69.37
	<i>L. bulgaricus</i>	1	3.6E+08	2.4E+08	2.4E+08	2.5E+08	2.2E+08	1.8E+08	1.5E+08	58.33
		2	3.9E+08	2.1E+08	2.0E+08	1.7E+08	1.3E+08	1.0E+08	6.0E+07	84.62
		Mean	3.8E+08	2.3E+08	2.2E+08	2.1E+08	1.8E+08	1.4E+08	1.1E+08	72.00
	Total count	1	1.0E+09	8.2E+08	7.1E+08	6.5E+08	5.1E+08	3.9E+08	3.2E+08	68.00
		2	9.0E+08	6.5E+08	6.0E+08	5.1E+08	4.1E+08	2.6E+08	1.9E+08	78.89
		Mean	9.5E+08	7.4E+08	6.6E+08	5.8E+08	4.6E+08	3.3E+08	2.6E+08	73.16

Time (min)	Volume (mL)			Volume (mL)							Volume (mL)			% Drained Whey
	No-vacuum I	% Drained Whey		(5 in. Hg vacuum)				% Drained Whey	(10 in. Hg vacuum)				% Drained Whey	
				I	II	III	Mean		I	II	III	Mean		
5	32	6.40		137	145	148	143.33	28.67	167	170	185	174.00	34.80	
10	49	9.80		191	195	194	193.33	38.67	216	224	235	225.00	45.00	
15	65	13.00		229	230	231	230.00	46.00	256	260	270	262.00	52.40	
20	76	15.20		254	257	258	256.33	51.27	282	285	290	285.67	57.13	
25	88	17.60		279	277	282	279.33	55.87	301	303	307	303.67	60.73	
30	100	20.00		299	295	298	297.33	59.47	314	316	321	317.00	63.40	
35	108	21.60		315	308	312	311.67	62.33	325	329	332	328.67	65.73	
40	116	23.20		328	322	323	324.33	64.87	335	343	342	340.00	68.00	
45	124	24.80		339	332	334	335.00	67.00	345	356	353	351.33	70.27	
50	131	26.20		348	340	343	343.67	68.73						
55	138	27.60		357	348	351	352.00	70.40						
60	145	29.00		363	356	358	359.00	71.80						

Appendix VIII - 1 Whey drainage from cow's milk yogurt by using cotton filter medium

Time (min)	Volume (mL)		Volume (mL)					Volume (mL)				% Drained Whey
	No-vacuum	% Drained	(5 in. Hg vacuum)				(10 in. Hg vacuum)					
			I	II	III	Mean	Whey	I	II	III	Mean	
5	19	3.80	119	108	103	110.00	22.00	194	200	190	194.67	38.93
10	32	6.40	174	163	153	163.33	32.67	248	260	254	254.00	50.80
15	44	8.80	209	204	198	203.67	40.73	294	305	300	299.67	59.93
20	55	11.00	243	236	235	238.00	47.60	319	325	322	322.00	64.40
25	64	12.80	270	259	262	263.67	52.73	340	346	336	340.67	68.13
30	73	14.60	278	284	293	285.00	57.00	356	360	351	355.67	71.13
35	81	16.20	318	304	312	311.33	62.27					
40	89	17.80	333	320	327	326.67	65.33					
45	96.5	19.30	343	335	340	339.33	67.87					
50	103	20.60	352	349	352	351.00	70.20					
55	109	21.80										
60	115	23.00										

Appendix VIII - 2 Whey drainage from cow's milk yogurt by using synthetic fiba filter medium

Time (min)	Volume (mL)				% Drained	Volume (mL)				% Drained	Volume (mL)				% Drained	
	(No- vacuum)			Whey		(5 in.Hg vacuum)			Whey		(10 in.Hg vacuum)			Whey		
	I	II	Mean			I	II	III			Mean	I	II			Mean
5	19	12	15.50	3.10	58	53	60	57.00	11.40	103	105	104.00	20.80			
10	29	21	25.00	5.00	79	72	80	77.00	15.40	136	140	138.00	27.60			
15	36	28	32.00	6.40	95	86	92	91.00	18.20	157	159	158.00	31.60			
20	41	34	37.50	7.50	108	100	115	107.67	21.53	174	174	174.00	34.80			
25	46	39	42.50	8.50	118	111	122	117.00	23.40	189	190	189.50	37.90			
30	51	44	47.50	9.50	128	122	132	127.33	25.47	200	205	202.50	40.50			
35	56	48	52.00	10.40	136	133	140	136.33	27.27	216	219	217.50	43.50			
40	61	53	57.00	11.40	146	141	150	145.67	29.13	222	225	223.50	44.70			
45	65	58	61.50	12.30	155	150	155	153.33	30.67	229	231	230.00	46.00			
50	69	61	65.00	13.00	161	156	162	159.67	31.93	236	237	236.50	47.30			
55	72	64	68.00	13.60	169	163	167	166.33	33.27	243	245	244.00	48.80			
60	75	68	71.50	14.30	177	167	173	172.33	34.47	250	250	250.00	50.00			
65	78	71	74.50	14.90	182	173	178	177.67	35.53	255	253	254.00	50.80			
70	81	75	78.00	15.60	187	177	183	182.33	36.47	259	256	257.50	51.50			
75	84	79	81.50	16.30	191	182	187	186.67	37.33	263	259	261.00	52.20			
80	87	82	84.50	16.90	196	186	192	191.33	38.27	267	261	264.00	52.80			
85	90	85	87.50	17.50	201	189	197	195.67	39.13	270	264	267.00	53.40			
90	93	88	90.50	18.10	206	193	200	199.67	39.93	274	267	270.50	54.10			

Appendix VIII - 3 Whey drainage from ewe's milk yogurt by using cotton fiba filter medium.

Time (min)	Volume (mL)		Volume (mL)				Volume (mL)			
	No - vacuum	% Drained	(5ins Hg vacuum)			% Drained	(10 ins. Hg vacuum)			
			I	II	Mean		I	II	Mean	
5	4	0.80	46	48	47.00	9.40	114	126	120.00	24.00
10	10	2.00	91	90	90.50	18.10	214	205	209.50	41.90
15	13.5	2.70	106	104	105.00	21.00	245	220	232.50	46.50
20	17	3.40	120	115	117.50	23.50	275	234	254.50	50.90
25	20	4.00	132	127	129.50	25.90	290	250	270.00	54.00
30	23	4.60	146	131	138.50	27.70				
35	26	5.20	155	137	146.00	29.20				
40	29.5	5.90	164	145	154.50	30.90				
45	32	6.40	171	150	160.50	32.10				
50	35	7.00	176	154	165.00	33.00				
55	37.5	7.50	184	159	171.50	34.30				
60	40	8.00	189	163	176.00	35.20				
65	42.5	8.50	194	167	180.50	36.10				
70	45	9.00	199	170	184.50	36.90				
75	47	9.40	203	173	188.00	37.60				
80	49	9.80	207	176	191.50	38.30				
85			212	179	195.50	39.10				

Appendix VIII - 4 Whey drainage from ewe's milk yogurt by using synthetic fiba filter medium.

Appendix XI-1 Published paper.

Abstract

The historical background and basic manufacturing methods for yogurt and related products in Turkey are summarized. These products are standard set and stirred type natural yogurt, torba or süzme yogurt (concentrated yogurt), "Ayran", "Çökelek" (cooked yogurt), "Silivri"-type yogurt, "Lor", "Çacik" and "Kurut" (dry cooked yogurt).

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