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THE UNIVERSITY OF ALBERTA

Studies to improve the shelf-life and consumer acceptance of
quarg

by

Tarsem Singh Sohal

A THESIS

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES AND RESEARCH
IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE
OF MASTER OF SCIENCE

IN

FOOD PROCESSING

DEPARTMENT OF FOOD SCIENCE

EDMONTON, ALBERTA

Spring, 1986

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The undersigned certify that they have read, and recommend to the Faculty of Graduate Studies and Research, for acceptance, a thesis entitled **Studies to improve the shelf-life and consumer acceptance of quarg** submitted by **Tarsem Singh Sohal** in partial fulfilment of the requirements for the degree of **MASTER OF SCIENCE in FOOD PROCESSING**.

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ABSTRACT

Quarg is a soft, unripened fresh cheese. The product available on Alberta market showed frequent shelf life problems related to bitterness. The bitterness was detectable after 1-2 days of storage and became unacceptable after 14 days of storage. This study was initiated to investigate the possible causes of bitterness development in quarg during storage.

Quarg was produced at the University of Alberta on a laboratory scale following the standard commercial procedure. The effect of microbial contaminants, rennet level, culture composition and culture strain were evaluated to study the potential causes for the bitterness in quarg.

Rennet level was found to be the critical factor in the development of bitter flavor. When the rennet level was reduced from the commercial practice of 3876 units/1000 kg of milk to 388 units/1000 kg of milk, an acceptable product with an extended shelf life was obtained. The results of storage study indicated low level of psychrotrophs, coliforms, yeasts and molds, suggesting that the bitterness was not the outcome of microbial contaminants. The nature of the starter culture did not play an important role in bitterness development during storage. This study showed that quarg produced with a reduced level of rennet could be stored at refrigerated temperature (4-5°C) for up to 9 weeks without bitterness development.

A household study was conducted to assess the consumer acceptance of plain and fruit quarg and to project the future market potential of this relatively new dairy product in Alberta. The statistical analysis of consumer acceptance study data indicated respondents' preferential choice for fruit quarg over the unflavoured product. This study also revealed that both plain and fruit quarg have good potential to become a regular part of the diet in Alberta and possibly other areas of North America.

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Table of Contents

Chapter	Page
1. INTRODUCTION	1
1.1 Speciality Cheese Production in Canada	1
1.2 Objectives of the Study	2
2. REVIEW OF LITERATURE	6
2.1 Quarg	6
2.2 The Manufacture of Quarg	7
2.3 Technological Developments in the Manufacture of Quarg	13
2.3.1 Retention of Whey Proteins by Heat Treatment	14
2.3.2 Retention of Whey Proteins by Ultrafiltration	15
2.3.3 Other Methods of Quarg Manufacture	17
2.4 Shelf Life of Quarg	18
2.5 Microflora of Quarg	23
2.6 Bitterness Development in Cheese	27
2.6.1 Influence of Rennet	28
2.6.2 Influence of Starter Bacteria	32
2.6.3 Influence of Psychrotrophic Organisms	35
2.7 Nutritional Value of Quarg	36
3. MATERIALS AND METHODS	41
3.1 Experimental Approach	41
3.2 Materials	41
3.2.1 Milk	42
3.2.2 Starter Culture	42
3.2.3 Rennet	42

3.2.4	Fruit Pulp	43
3.2.5	Microbiological Media	43
3.2.6	Chemicals†	43
3.3	Preparation of Mother Culture	43
3.4	Preparation of Rennet Solution	44
3.5	Laboratory Production of Quarg	44
3.6	Sampling of Quarg	49
3.7	Microbiological Analyses	49
3.7.1	Psychrotrophic Count	49
3.7.2	Coliform Count	49
3.7.3	Yeast and Mold Count	50
3.8	Chemical Analyses	50
3.8.1	Moisture content	50
3.8.2	Butterfat	50
3.8.3	Protein	51
3.8.4	Titratable Acidity	51
3.8.5	Determination of pH	51
3.9	Sensory Evaluation of Quarg	52
3.9.1	Preliminary Study	52
3.9.2	Panel Selection and Training	53
3.9.3	Sensory Evaluation of Stored Quarg	54
3.10	Consumer Acceptance Study	55
3.10.1	Household Selection Procedure	55
3.10.2	Preparation of Quarg Samples	56
3.10.3	Survey Method and Questionnaire	56
4.	RESULTS AND DISCUSSION	58
4.1	Preliminary Study	58

4.2	Effect of Rennet Level on Bitterness in Quarg	66
4.3	Effect of Starter Culture on Bitterness in Quarg	79
4.4	Commercial Production of Quarg	89
4.5	Yield of Quarg	95
4.6	Consumer Acceptance Study	96
4.6.1	Survey Response	96
4.6.2	Opinion on Plain and Fruit Quarg	96
4.6.3	Market Potential of Plain and Fruit Quarg	100
4.6.4	General Comments from Respondents	102
5.	CONCLUSIONS AND RECOMMENDATIONS	109
5.1	Summary of Results	109
5.2	Future Research Needs	111
6.	BIBLIOGRAPHY	113
	APPENDIX 1 : TRIANGLE TEST FORM	130
	APPENDIX 2 : SENSORY EVALUATION OF QUARG	131
	APPENDIX 3 : NEW DAIRY PRODUCT SURVEY	132
	APPENDIX 4 : SENSORY EVALUATION DATA OF QUARG	133

List of Tables

Table		Page
2.1	Composition of quarg produced from whole and skim milk	8
2.2	Nutritional value of different dairy products	37
4.1	Composition of quarg produced on a laboratory scale and commercially with 3876 units rennet per 1000 kg milk and 1% Starter Culture (Flora Danica)	59
4.2	Changes in the microflora, pH and lactic acid in quarg during storage at 5°C. The quarg was produced commercially using 1% starter culture (Flora Danica) and 3876 units rennet per 1000 kg of milk	60
4.3	Changes in the microflora, pH and lactic acid in quarg during storage at 10°C. The quarg was produced commercially using 1% starter culture (Flora Danica) and 3876 units rennet per 1000 kg of milk	61
4.4	Changes in the microflora, pH and lactic acid in quarg during storage at 5°C. The quarg was produced using 1% starter culture (Flora Danica) and 3876 units rennet per 1000 kg of milk	62
4.5	Changes in the microflora, pH and lactic acid in quarg during storage at 10°C. The quarg was produced using 1% starter culture (Flora Danica) and 3876 units rennet per 1000 kg of milk	63
4.6	Effect of storage at two different temperatures (5° and 10°C) on bitterness development in quarg produced on a laboratory scale and commercially using 1% starter culture (Flora Danica) and 3876 units rennet per 1000 Kg milk	64
4.7	Composition of Quarg produced with 1% starter culture (Flora Danica) and different levels of rennet	67

Table	Page
4.8 Changes in the microflora, pH and lactic acid in quarg during storage at 7°C. The quarg was produced using 1% Flora Danica and 0 unit rennet per 1000 kg of milk	69
4.9 Changes in the microflora, pH and lactic acid in quarg during storage at 7°C. The quarg was produced using 1% Flora Danica and 388 units rennet per 1000 kg of milk	70
4.10 Changes in the microflora, pH and lactic acid in quarg during storage at 7°C. The quarg was produced using 1% Flora Danica and 775 units rennet per 1000 kg of milk	71
4.11 Changes in the microflora, pH and lactic acid in quarg during storage at 7°C. The quarg was produced using 1% Flora Danica and 1550 units rennet per 1000 kg of milk	72
4.12 Changes in the microflora, pH and lactic acid in quarg during storage at 7°C. The quarg was produced using 1% Flora Danica and 3876 units rennet per 1000 kg of milk	73
4.13 Effect of five different rennet levels on bitterness development in quarg during storage. Starter culture used was 1% Flora Danica	74
4.14 Analysis of variance on sensory data of quarg produced using five rennet levels, 1% starter culture (Flora Danica) and stored at 7°C for 4 Weeks	75
4.15 Comparison of Means for five rennet levels used in the production of quarg with 1% Flora Danica	76
4.16 Composition of quarg produced with 388 units of rennet per 1000 kg of milk and four different type of starter culture at 1% level	80
4.17 Changes in the microflora, pH and lactic acid in quarg during storage at 7°C. The quarg was produced using 1% Flora Danica and 388 units rennet per 1000 kg of milk	82

4.18	Changes in the microflora, pH and lactic acid in quarg during storage at 7°C. The quarg was produced using 1% Flora Danica without <i>S. diacetylactis</i> and 388 units rennet per 1000 kg of milk	83
4.19	Changes in the microflora, pH and lactic acid in quarg during storage at 7°C. The quarg was produced using 1% single strain culture (# 134) and 388 units rennet per 1000 kg of milk	84
4.20	Changes in the microflora, pH and lactic acid in quarg during storage at 7°C. The quarg was produced using 1% single strain culture (# 584) and 388 units rennet per 1000 kg of milk	85
4.21	Effect of four different starter cultures on bitterness development in quarg during storage. Rennet level used was 388 units per 1000 kg of milk	86
4.22	Analysis of variance on sensory data of quarg produced using four different starter cultures, 388 units rennet per 1000 kg of milk and stored at 7°C for 4 weeks	87
4.23	Comparison of means for four different starter cultures used in the production of quarg with 388 units rennet per 1000 kg of milk	88
4.24	Composition of quarg produced commercially with 388 units rennet per 1000 kg milk and 1% starter culture (Flora Danica)	91
4.25	Changes in the microflora, pH and lactic acid in quarg during storage at 7°C. The quarg was produced commercially using 1% Flora Danica and 388 units rennet per 1000 kg of milk	92
4.26	Sensory evaluation data of commercial quarg produced with 388 units rennet per 1000 kg of milk and 1% starter culture (Flora Danica) and stored at 7°C	93

Table	Page
4.27 Analysis of variance on sensory data of quarg produced commercially using 388 units rennet per 1000 kg of milk, 1% starter culture (Flora Danica) and stored at 7°C for 4 weeks.....	94
4.28 Statistical analysis of consumer acceptance survey data	101
4.29 Consumption pattern of quarg by respondents.	106
A-4.1 Sensory evaluation data of fresh quarg produced with 1% Flora Danica and different levels of rennet	133
A-4.2 Sensory evaluation data of quarg after 1 week of storage. The quarg was produced with 1% Flora Danica and different levels of rennet	134
A-4.3 Sensory evaluation data of quarg after 2 weeks of storage. The quarg was produced with 1% Flora Danica and different levels of rennet	135
A-4.4 Sensory evaluation data of quarg after 3 weeks of storage. The quarg was produced with 1% Flora Danica and different levels of rennet	136
A-4.5 Sensory evaluation data of quarg after 4 weeks of storage. The quarg was produced with 1% Flora Danica and different levels of rennet	137
A-4.6 Sensory evaluation data of fresh quarg. The quarg was produced with 388 units rennet per 1000 kg milk and four different starter cultures	138
A-4.7 Sensory evaluation data of quarg after 1 week of storage. The quarg was produced with 388 units rennet per 1000 kg milk and four different starter cultures	139
A-4.8 Sensory evaluation data of quarg after 2 weeks of storage. The quarg was produced with 388 units rennet per 1000 kg milk and four different starter cultures	140

Table

Page

A-4.9	Sensory evaluation data of quarg after 3 weeks of storage. The quarg was produced with 388 units rennet per 1000 kg milk and four different starter cultures	141
A-4.10	Sensory evaluation data of quarg after 3 weeks of storage. The quarg was produced with 388 units rennet per 1000 kg milk and four different starter cultures	142

List of Figures

Figure	Page
1.1 Production, disappearance, import and export of specialty cheeses in Canada during 1972-1983	3
1.2 Per capita consumption of specialty cheeses in Canada during 1972-1983	4
3.1a Quarg Producing Assembly (a and b).....	45
3.1b Quarg Producing Assembly (c and d).....	46
3.2 Flow-diagram of quarg production	48
4.1 Respondents opinion on appearance of plain and fruit quarg	97
4.2 Respondents opinion on flavour of plain and fruit quarg	98
4.3 Respondents opinion on overall quality of plain and fruit quarg	99
4.4 Respondents previous exposure to quarg	103
4.5 Frequency of respondents consumption of quarg	104
4.6 Respondents willingness to buy plain and fruit quarg	105

1. INTRODUCTION

Quarg is a soft, unripened fresh cheese. In Canada, it is classified as a specialty cheese and can be described (Kroger, 1980) as milky white or faintly yellowish in colour. Body and texture are homogeneously soft and mildly supple or elastic. Quarg originated in Germany where it is more popular than yogurt (Mann, 1978). Quarg has the potential of becoming a regular part of the diet in every North American household.

Quarg is one of the specialty cheeses being manufactured in Alberta. Earlier, the product had a limited shelf-life of two weeks or less. Microbial contamination, especially yeasts and molds, and development of bitterness were two areas of concern. The limited shelf-life restricted the product to be sold within the province whereas the product has considerable export potential as the commercial plant in Alberta is the only one making quarg in Western Canada. However, in Europe where quarg is more popular, the problems of extended shelf-life are not of great consequence since the product is consumed within 5-7 days. Consequently little research has been carried out in the area of shelf-life problems.

1.1 Speciality Cheese Production in Canada

In Canada most of the specialty cheeses (such as Brick, Limburger, Gouda, Edam, Colby, Monterey Jack, Quarg and Cream cheese) are imported from Europe and USA. The domestic

production of specialty cheese demonstrated continued growth during last ten years as shown in Figure 1.1. According to Agriculture Canada (1985) statistics the domestic production of all specialty cheeses exceeded 94.5 kT during 1984-85, which represents an increase of 8.2% over 1983-84 production. However, the imports remained at the global quota level of 20 kT. As evident from Figure 1.2, the per capita consumption of specialty cheeses increased from 1.39 kg to 4.09 kg during last ten years. The policy of the Alberta Dairy Division, Department of Agriculture is to encourage increased production of variety cheeses in Alberta in order to curb the import of such dairy products.

1.2 Objectives of the Study

Search of literature revealed that very limited work has been done on bitterness in quarg, which appeared to be the major quality defect affecting the shelf-life of this product. It was therefore decided to investigate the possible causes of the short shelf-life and the bitterness development in quarg during storage. Also, there was interest to find reasons for the low consumption pattern of this highly nutritious dairy product in Canada when compared with many European countries. Thus the specific objectives of this study were:

1. To investigate the effects of microbial flora, rennet level, culture composition and culture strain on the shelf life of quarg, including microbial growth and

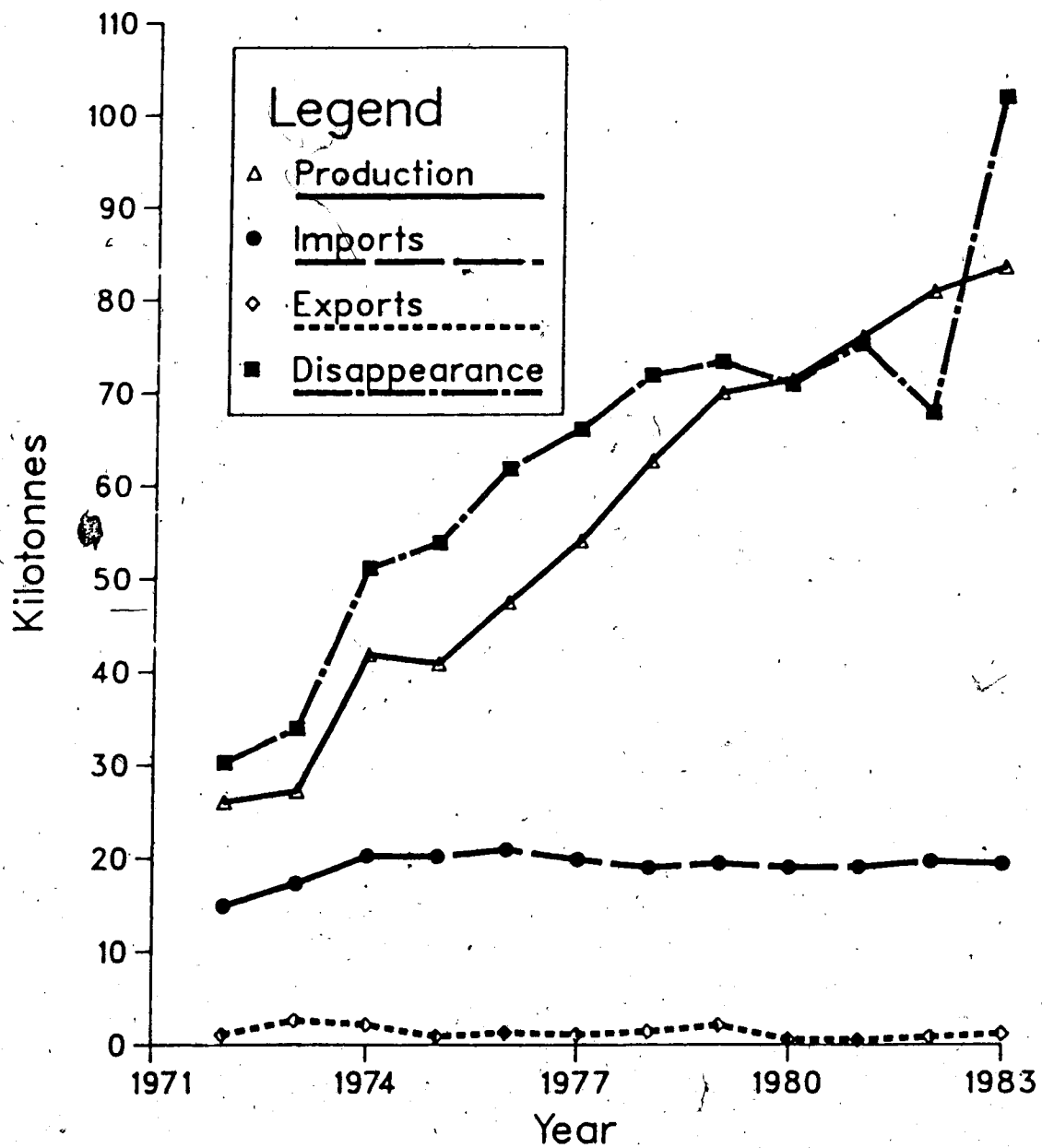


Figure 1.1 Production, disappearance, import and export of specialty cheeses in Canada during 1972-1983. (Source: Agriculture Canada, 1984).

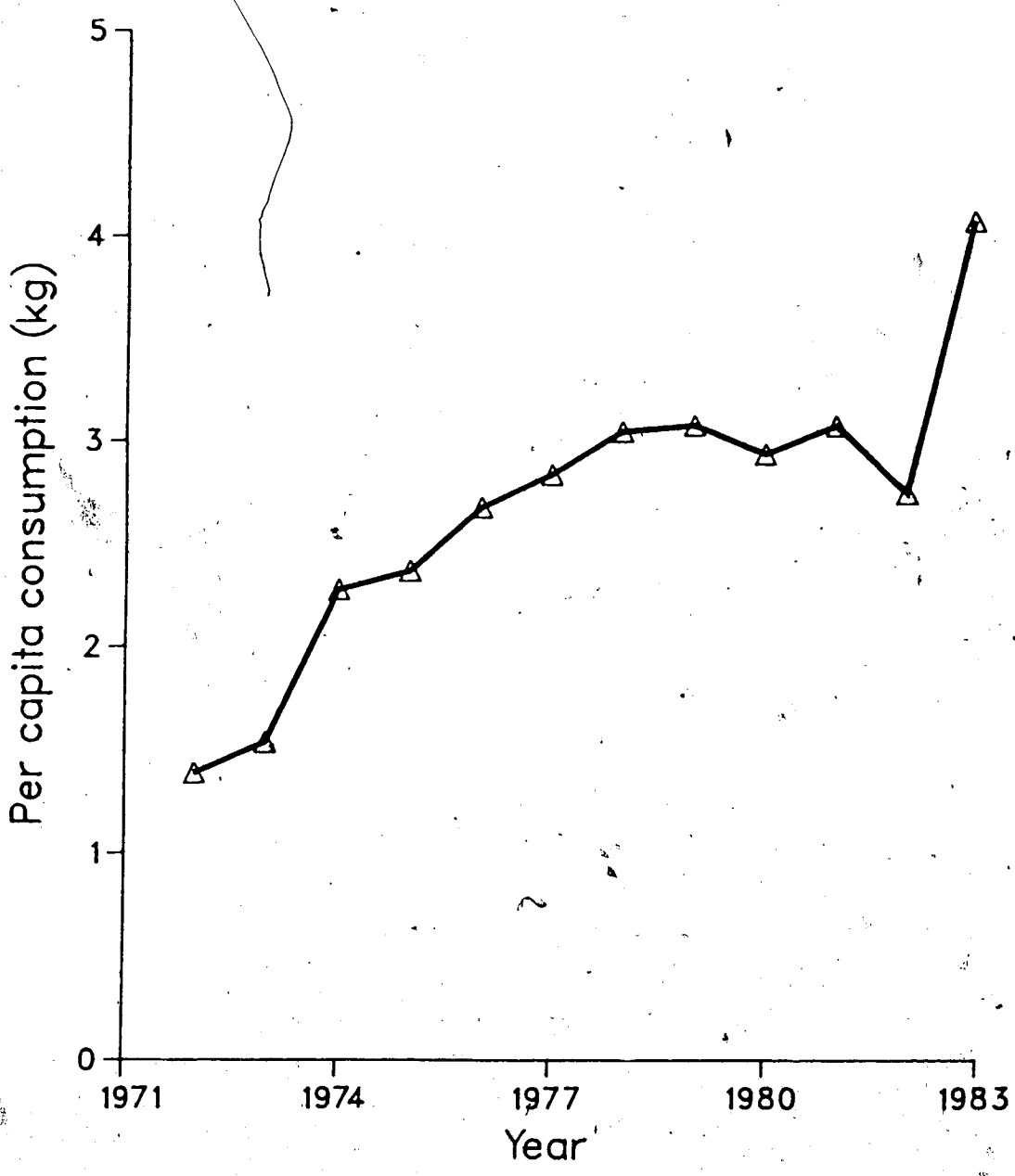


Figure 1.2 Per capita consumption of specialty cheeses, in Canada during 1972-1983. (Source: Agriculture Canada, 1984).

bitter flavour development.

2. To recommend changes in the current industrial process which would result in quality improvement and shelf-life extension without impairment of yield.
3. To study the market potential of plain and fruit quarg, using a consumer acceptance study to ascertain the future marketing trend of this relatively new dairy product in North America.

2. REVIEW OF LITERATURE

2.1 Quarg

Quarg (also Quark, Tvaroh or Twarog) is a dairy product popular in Central Europe. It is smooth like cream cheese, spreadable and usually made with a buttermilk culture rather than cottage cheese culture. Quarg contains more acid than cottage cheese and has smooth body and texture. The colour of quarg is white or slightly yellowish. Quarg should possess good spreadability without any appearance of water (or whey), dryness or graininess. The flavour must be clean and may be slightly acidic. This versatile product is used by Europeans as a simple dairy spread or it may be seasoned and flavoured for variety. It is also used in salads, on baked potatoes, on snack biscuits, and in baked foods such as cheese cake (Claydon *et al.* 1972). Other products found in European markets include whipped quarg of porous consistency, quarg flavoured with horse-radish, red paprika, onions and other vegetables, vanilla or fruit flavoured sweetened dessert quargs with 10% fat in the dry matter, or products made by whipping quarg with added fruit, often combined with jellies and whipped cream (Lang, 1981; Jelen, personal communication).

Quarg is essentially an acid coagulated, flocculated casein with a high moisture content (70-80%). It is produced from the milk mainly by lactic acid coagulation, followed by addition of rennet and finally separation of the whey.

Composition is variable and depends on the composition of the vat milk. Chemical composition of quarg made from full-fat and skim milk is shown in Table 2.1. Commercially, quarg is available in Europe with different fat levels (Kroger, 1980).

2.2 The Manufacture of Quarg

The basic technology of quarg production is described by Kroger (1980). Good quality pasteurized skim milk, cooled to 20-22°C, is mixed with a 1-2% bulk starter culture. Rennet is added 60-90 minutes after culture addition at the rate of 3-5 g dry preparation per 100 litre milk or 1 to 2 mL of a 1:10,000 rennet solution per 100 litre milk. The pH is about 6.3 at this stage. Rennet enhances the protein stabilization, but an excessive amount causes off-flavours (Kroger, 1980). The low temperature quarg manufacturing method has always been preferred, although incubation at high temperature (25-30°C) has also been practised. Milk temperature and amount of starter culture should be adjusted so that a pH of 4.6-4.7 is reached after 16-18 hours. This would represent 0.50-0.54% acidity expressed as lactic acid. Coagulation begins after 6-8 hours of incubation at a pH 5.5 or about 0.41% acidity.

Traditionally, the subsequent whey separation is achieved by cutting the curd into a mass of particles of 10-15 cm (4-6 inches) in diameter and by scooping the curd mass into bags or onto cloth lined drain tables. This method

Table 2.1 Composition of quarg produced from whole and skim milk.

Components	Skim ¹ Milk	Whole ² Milk
	(%)	(%)
Total solids	17.48	26.98
Protein	12.10	9.87
Butterfat	Traces	13.40

¹From Puhap and Gallman (1981). Ultrafiltration in the manufacture of quarg.

²Analysis of commercial quarg available in Alberta (Kadis, personal communication).

has severe limitations when the daily milk volume is higher than 10,000 litres.

In the late 1950's first attempt was made to produce quarg with a separator. The removal of the whey from the coagulum by means of a specially constructed quarg separator was successful and this method of quarg manufacture has been used increasingly ever since. The first quarg separator began operating in 1961 in the dairy town of Bielefeld, West Germany (Kroger, 1980)

Two major companies manufacture quarg equipment: Westfalia Separator A.G., Oelde, West Germany (in North America represented by Centrico, inc.) and Alfa-Laval, Lund, Sweden. According to information supplied by these two companies, Westfalia Separator claims that it has supplied 90% of all the quarg separators operating in Germany and that the market share is as high in other countries. In 1977, 233 Westfalia quarg separators were producing 1,000,000 kg quarg every day (Kroger, 1980)

Modern methods of quarg manufacture are highly automated and controlled. They include equipment not in existence 25 years ago. New problems such as moisture control, chemical cleaning-in-place operations, and yield optimization had to be solved.

According to Lang (1980) the commercial manufacturing processes generally consist of ripening the skim milk with a starter and coagulation with a small amount of rennet, mostly in vertical tanks of 5,000 litres or more. The

coagulated milk is well stirred and passed on to the separator. The function of the separator in this case differs substantially from that of an ordinary milk separator. Separation of the curd of higher specific gravity from the lighter whey occurs, with the heavier curd being forced outwards towards the periphery of the bowl and discharged through a number of nozzles (0.4-0.8 mm in diameter) into a circular trough surrounding the bowl. The lighter whey moves inwards and is discharged through openings in the top of the bowl. The curd is cooled to about 4°C by passage through a tubular or plate cooler using chilled water. The separated curd is then mixed with cream to the desired fat content.

In a large-scale production, this may be done continuously, with the curd being measured volumetrically; the measured value may govern a metering pump for the addition of the required amount of cream. Other ingredients, such as fruit, flavouring substances or fruit juices may also be added in appropriate quantities. It is, however, essential that the solids content of the curd and fat content of the cream be maintained constant as far as possible. There is no need to homogenize the product manufactured by this process.

Optionally, the curd-whey mixture from the coagulation tanks may be pumped by a positive pump through a heat exchanger to be heated to about 60°C to destroy most of the microflora and to extend the shelf-life of the product. This

process referred to as thermization is becoming popular especially in countries with poor microbiological quality of milk (Jelen, personal communication).

Other methods of quarg manufacture are outlined by Lang (1980). One method involves the use of a rectangular coagulation tank with a special 'filtering' cover rotatable along its longitudinal axis. When the milk coagulates, the special 'filtering' cover (provided with a filter cloth) is attached and, after a predetermined time the tank begins to rotate, causing breaking up of the coagulum and drainage of the whey through the cloth in the cover. Finally, the cover is removed and the tank is rotated to collect the curd in a mobile container placed below. Another system consists essentially of two vats of semi-circular cross-section of the same shape, but of slightly different dimensions, placed on top of each other. First, the milk is coagulated in the larger container and then, after a predetermined interval, the smaller (perforated) container is immersed into the coagulum, pressing out the whey, by its weight, through the perforations in its walls. The whey accumulates in it and is pumped out. After the desired degree of pressing, the perforated container is lifted pneumatically and the pressed quarg is removed through an opening in the bottom into a mobile vessel below.

A report from the German Federal Republic (Lang, 1980) gives details of the operation of a modern quarg production line employing a new "Thermo-process" design in a dairy

plant at Wetzlar, which processes annually some 25 million kg milk into a variety of products including quarg, cocoa drink, several types of cultured milks, cream and butter.

The process starts with high temperature pasteurization of skim milk with concurrent denaturation of whey proteins. The milk is then cooled to the renneting temperature and transferred to the coagulation tank where it is inoculated with a "Biograde" culture (specially developed for use with this process of quarg manufacture), followed by the addition of rennet and the milk is then held overnight. The holding of the heated milk in the soured form before separation causes contraction of casein particles with enclosed and adsorbed whey proteins, thus enabling the normally difficult-to-separate whey proteins to be recovered.

Next day, the coagulated skim milk is pumped into a surge tank, provided with a stirrer, where it is subjected to another heat treatment at pH 4.5. The additional heat treatment causes most of the precipitated whey proteins to be retained in the quarg. The coagulated milk is cooled and separated in a Westfalia quarg separator. The separator is supplied with sterile air to avoid the possibility of recontamination and the cooling air for the motor is fed in via a duct directly from the outside and is discharged to outside again in a closed channel.

The separated curd is discharged into a covered hopper and is pumped through a moisture meter and a mixer into a tank. The moisture content is measured continuously and

recorded. The tank is supplied with sterile air to prevent airborne contamination. Positive displacement pumps feed the quarg into a packaging machine.

Quarg may be made into an aerated product by incorporating whipped cream and various additives such as sugar, flavours and gelatin as the stabilizer (Llewellyn, 1983). The process involves preparation and heating of the stabilizer and flavouring components dissolved in water. Measured quantities of this solution are then continuously blended into quarg. At the same time, whipping cream is foamed continuously and mixed with the quarg in a helical conveyor, and the product is packaged. The product contains 30 percent fat in the dry matter.

2.3 Technological Developments in the Manufacture of Quarg

Recent advances in quarg manufacture are the introduction of the curd separator, coagulation of milk in closed tanks, thermization (heat treatment) of the coagulated milk before separation, cooling of the product in tubular or plate heat-exchangers and continuous product pasteurization in heat exchangers. The introduction of new curd separators doubled the capacity of quarg production per hour (Lang, 1980). These developments are mainly geared towards the improvement of shelf-life and retention of whey proteins in the product. Various researchers have used different cultures to improve the shelf-life of quarg. In recent years most of the developments have been in the area

of ultrafiltration to retain the whey proteins in the product.

2.3.1 Retention of Whey Proteins by Heat Treatment

According to Lang (1980), the economy of quarg production may be determined to a large extent by the amount of whey protein retention in the product. Methods by which this can be achieved are the centri-whey process of protein precipitation in whey, the "Thermo-process" for precipitating the protein in skim milk before coagulation, and ultrafiltration of milk for concentrating casein and whey protein simultaneously. These methods substantially improved the yield of quarg, in terms of skim milk required per kg of product with 17.5% total solids, in comparison to the conventional method.

In the Centri-whey process, whey from previous quarg production is heated to 96°C, held for about 20 minutes to precipitate the protein and then passed through a self desludging separator to obtain a protein concentrate with 15-18% total solids and 10-12% protein. The whey proteins are precipitated by severe heating before they are incorporated into the products. The recovery of the coagulated proteins in the concentrate is claimed to be 90%. This concentrate is added to the cheese milk, to pass together with the casein into the finished product. The concentrate can also be used in other products such as desserts and low fat spreads (Lang, 1980). In the

"Thermo-process" approach, skim milk is heated to 95-98°C and held for 2.5 min at that temperature for the retention of whey proteins in the quarg (Dolle, 1978).

Zall (1981) carried out an experiment to study the effect of heat-treatment and storing of milk on the farm prior to making quarg. He found that quarg yield was not increased significantly when the product was made with heated/stored milk. The milk was heated to 70-90°C and stored in a refrigerated vessel for 7 days. It was found that this heat treatment reduced psychrotrophs, stabilised acid degree value and prolonged the delicate flavour of milk. The quality and shelf-life of quarg were not impaired.

2.3.2 Retention of Whey Proteins by Ultrafiltration

According to Puhan and Gallman (1981), the major advantage of using ultrafiltration (UF) is that UF has the ability to retain all the milk proteins. This results in higher yield and an improved casein:whey protein ratio of 4:1 which corresponds to that in milk, making the product more valuable from a nutritional point of view as the protein efficiency ratio (PER) of whey proteins is higher than casein. According to Puhan and Gallman's method (1981) pasteurized skim milk was inoculated with a mesophilic starter and incubated at 23°C for seven hours. The pH dropped to 5.7-5.9 after this period of incubation. It was then concentrated by ultrafiltration to 12% protein content at 20-25°C and fermentation was continued until the pH

dropped to 4.55. The fermentation was not affected by the ultrafiltration process. The texture of the quarg thus produced was firm and grainy and homogenization was necessary if pasty texture was required. In evaluation of sensory properties, no difference was noted in comparison to conventional quarg.

Hensen and Bret (1976a,b) used the ultrafiltration process to concentrate milk and to remove mineral salts with or without removal of lactose. Fresh milk was pasteurized at 72°C for 15 sec and the cream was separated. Skim milk thus obtained was ultrafiltered to obtain a protein rich concentrate. The quarg was made from this protein-rich (up to 15% protein) concentrate by using a conventional method. The coagulum was whipped to provide quarg with overrun of 200%. This method of quarg making improved the viscosity, protein ratio and yield.

Using ultrafiltration, Gungerich (1981) improved the traditional product not only in terms of efficiency but also with respect to nutrition because of the retention of whey proteins in the product, as whey proteins have higher nutritional value compared to casein (Renner, 1983). Another method he used consisted of thermizing the entire amount of skim milk in order to reduce the amount of whey, which was removed in the quarg separator. In this way, protein content in whey was reduced to 0.52% and the amount of milk required per kg of quarg was reduced from 4.6 to 4.1 kg. A product of this type has a shelf-life of 21 days and, in general, a

satisfactory quality.

2.3.3 Other Methods of Quarg Manufacture

The manufacture of quarg in Poland, as described by Poznanski *et al.* (1974), consisted of precipitating both the whey proteins and casein. Whey proteins were interacted with casein and the complex was separated from milk by a suitable technological method. It was claimed that the above process enables the recovery of 96-97% of all proteins present in the milk and the yield was increased by 12-15%. The product had a shelf-life of 5 days at a temperature below 10°C.

Lotter *et al.* (1975) made quarg by acidifying milk at 15-30°C to bring down the pH to 4.95-5.35. Acidogen and proteolytic enzymes were then added to the warm acidified milk, which was kept for four hours until curd was formed. Acid cheese curd production was improved by using this process.

Tomka *et al.* (1978) used an electrically driven perforated drum to drain whey from quarg curd. This method of whey separation saved time (rotary drum 4-10 min. compared to 60-150 min. in a conventional drainage cart), space and heavy work. No adverse effects were observed in regard to fat, loss of protein, yield and sensory quality of the quarg.

Lipatov *et al.* (1978) studied the effects of pressure in milk homogenization and of coagulation temperature on the continuous production of quarg. Increase in homogenization

pressure reduced cheese firmness and the structure became flaky. The coagulation temperature above 38°C affected cheese structure and syneresis substantially. The body consistency of quarg became coarse and rubbery.

Ott *et al.* (1978) described a process of quarg manufacturing from skim milk. Skim milk was heat treated at 90-95°C for 2-3 min and cooled to 28-30°C. Pasteurized skim milk was then inoculated with a starter culture and rennet and incubated for 16 hours. When the pH dropped to 4.5 the coagulum was heated to 60°C and held at that temperature for 1-2 min. The coagulum was then cooled to 40°C and centrifuged in a curd separator. The curd was further cooled to 6-8°C and vigorously mixed with 3% starter culture and a calculated amount of cultured cream. The product so obtained was then packaged. The results showed that the whey from the separator contains approximately 0.4% less total solids than whey from quarg produced conventionally. The increased yield of 10% has also been claimed when the "Thermo-process" and Westfalia quarg whey separator were used due to retention of whey proteins.

2.4 Shelf Life of Quarg

Quarg has a limited shelf-life due to development of bitterness and the growth of contaminating microorganisms during storage. Studies have been carried out with the aim to prolong the storage life. Some of the methods tried were heat treatment, use of chemical preservatives and use of

different microbial cultures.

Frank (1963) studied the significance of reinfection in quarg manufacture. The development of non-acidifying bacteria, coliform, yeasts and molds in quarg stored at 4-20°C for up to 18 days was examined. No significant changes occurred at 4°C for up to 8 days, whereas at 10°C the mold count increased 355-fold and yeast count about 50-fold with the products exhibiting a strong yeasty-fermented off-flavour. At 15° or 20°C the populations increased more than 1,000-fold on the 4th day and rapid spoilage occurred. It was recommended, therefore, that the storage temperature should not be higher than 4-6°C.

Yurin (1966) examined the microbiological quality of milk and dairy products after various storage periods in polyethylene containers. Pasteurized milk, sour cream, high-fat quarg and dried milk were stored in low-density polyethylene film of 50 or 100 μm thickness and kept at $3\pm 1^\circ\text{C}$ for 6-48 h (liquid milk), 1-14 days (cream or quarg), or 1-6 months (dried milk). The quarg samples showed undesirable changes after 7 days.

Studies by Ailsa *et al.* (1969) on manufacture of quarg by acid coagulation of milk indicated that quarg can be stored for 15 days at both 5°C and 7°C before showing increase in numbers of presumptive spoilage organisms including yeast and mold. However, no sign of coliform contamination was observed even after 19 days of storage.

The preservation of dairy products with chemicals was examined by Weiss (1970). Non-pasteurized skim-milk quarg was treated with 0, 0.015, 0.022, 0.033 and 0.050% potassium sorbate and stored for 40 days at 8°C and room temperature. This showed that at 8°C the preservative virtually eliminated mold contamination and greatly enhanced the overall shelf life of the quarg.

Krcal (1970) studied the effect of CO₂ and N₂ on the growth of spoilage microorganisms in quarg. The study was conducted in laboratory, pilot plant and commercial conditions. Both CO₂ and N₂ restricted the growth of some predominant microorganisms in quarg during storage under laboratory conditions. In storage, under pilot plant and commercial conditions, the beneficial effect of either atmosphere persisted for 4-6 months. As N₂ or CO₂ inhibited the growth of aerobic microflora on the surface, the acidity of the product decreased slightly and the pH rose moderately, resulting in a marked reduction in soluble nitrogenous products of protein breakdown. The end result was a longer shelf life, increased yield and a better quality quarg. It would not be possible to maintain a clear N₂ atmosphere because of a large volume of CO₂ developed during quarg making and storage (as a result of microbial action). Also, CO₂ is more bacteriostatic than N₂. It would, therefore, be preferable to use CO₂.

Schulz and Thomasow (1970) experimented with sorbic acid to preserve quarg. It was found that the addition of

0.07% sorbic acid improved the keeping quality of quarg by one week when stored at 15-20°C. In an attempt to control the growth of yeast and mold on the surface of quarg, good results were obtained by using a parchment paper cover treated with potassium sorbate (dry 0.2g/dm²).

Braun (1972), described a method of increasing the shelf life of yogurt, kefir and quarg. These products containing additives of vegetable origin were filled into containers with lids. Prior to closing them, lids were held spaced above the top edge of the container, CO₂ was injected from the side through the space into the container, displacing the air inside and then the lid was applied. The CO₂ becomes partially dissolved in the product and was found to prevent yeast and mold growth and confer to the products a shelf life of several months without affecting the flavour.

Manus (1973) modified the quarg manufacturing process by inoculating milk containing 1.3-3.6% fat with a high acid producing starter (*Streptococcus lactis*, *Streptococcus cremoris*, or a yogurt culture) prior to addition of rennet. When the pH was about 4.3 the coagulum was heated to 60-65°C for 20 minutes and then cooled to 32°C before separation in a curd separator. The separated whey was pasteurized and the mixture of pasteurized whey, salt and stabilizer was added to the curd in a suitable quantity to reduce the fat content to 4-10%. The product with 21-22% total solids was very smooth and spreadable and had a greatly extended storage

life.

Babella and Szabo (1974) reported the use of thermophilic yogurt culture (*Streptococcus thermophilus* and *Lactobacillus bulgaricus*) instead of using a conventional starter. By exposing the curd to different heating temperatures and times (65, 70 or 75 °C over 10, 60 and 180 sec) they could improve the sensory quality of quarg. However, rapid heating was essential to ensure product quality. The pasteurized product could be stored at room temperature (20°C) for 14 days or for a month under refrigerated conditions (7°C).

Data obtained in a one-month laboratory study on the keeping quality of different types of quarg were given in a study by Thamm (1977) of 70 quarg samples (25 from skim-milk, 22 with 20% fat, 17 with 40% fat and 6 herb quargs) stored at 10°C. Of these, 47-64% exhibited slight sensory defects after 10 days; after 15 days the incidence of slight defects increased to 50-80% and pronounced defects appeared in 13-26% of the samples. When the storage temperature was reduced to 6°C, only 5-17% of the samples were slightly defective after 20 days of storage.

A study by Holdt (1978) revealed that plain quarg processed by heating at 85°C after incubation, possessed a storage life of about 2-3 weeks at 8-10°C. Quarg with additives such as fruit, vegetables, spices and stabilizers had a shelf-life of more than 4 weeks, whereas quarg containing liquid additives like fruit juice, or stabilizers

and whipped with the incorporation of N_2 and aseptically packaged could be stored for 2-3 months.

Beerwerth (1979) found that, despite constant improvements in plant hygiene and reduction in coliform titres, many samples of quarg exhibited considerable sensory defects. These defects resulted largely from yeast and mold contamination and too long periods of guaranteed keeping quality. Over 21% of samples with yeast and/or mold counts of 10^4 - 10^5 CFU/g and 96% of samples with more than 10^5 CFU/g were not of market quality. Rapid growth of the spoilage microflora (more than 10,000 CFU/g) was observed approximately 1 week before the guaranteed period. Sensory defects were observed in 50% of samples stored at 10°C .

2.5 Microflora of Quarg

Quarg is a high moisture, medium acid dairy product. It provides a favourable environment for the growth and proliferation of microbes, particularly yeasts and molds. Several studies have been carried out to determine the microflora present in quarg.

Lyshcheva (1968) examined the contaminating microflora of full-fat quarg. Cultures were isolated from 150 samples of fresh, stored and frozen quarg. *Micrococcus albidus*, *Micrococcus radiatus*, *Staphylococcus aureus* and *Micrococcus albocereus* were identified in all cases; they were actively lipolytic but not proteolytic. Spore-forming yeasts isolated were *Saccharomyces lactis*, *Saccharomyces ribis*;

non-sporeforming yeasts included *Torulopsis kefir*, *Torulopsis uvae*, *Torula candida*, *Candida krusei*, *Candida catenulata*. All the yeasts were lipolytic, only *Saccharomyces lactis* and *Saccharomyces ribis* were proteolytic. Bacilli isolated included *Bacillus cereus*, *Bacillus megaterium*, *Bacillus angulans*, *Bacillus albolactis*, *Bacillus mesentericus*. All were actively proteolytic. Molds isolated were *Penicillium commune*, *Penicillium roqueforti*, *Aspergillus glaucus*, *Aspergillus clavop*, *Mucor mucedo* and *Mucor racemosus*, all were lipolytic.

• Mudretsova-Viss and Zav'yalova (1968) examined changes in the microflora of quarg during different storage temperatures. Full-fat quarg manufactured from renneted and starter-treated pasteurized milk was stored at 10, 4-5, 0-2, -10, -12 and -18 °C in wooden boxes containing 5 kg packs. The surface layer and the interior of the quarg were examined bacteriologically and chemically for up to 31 days. Within the temperature range 0-10°C, spoilage occurred mainly in the surface layer. This was caused by *Geotrichum candidum*, *Torulopsis* and *Candida* species, and to some extent by acetic acid bacteria. Limits of storage without appreciable deterioration were 2-3 days at 10°C, 7-9 days at 4-5°C and 25-27 days at 0-2°C.

Muzikar (1970), examined the incidence and importance of yeasts and related microflora in quarg (tvaroh) desserts. Eighty seven samples of the dessert (prepared by adding sugar, fruit syrup and jam) were examined by disintegration

of 10 g of the sample in 90 mL of physiological saline solution, with final dilution of 1:10,000 using 0.5 mL inoculum on malt agar with 3 days incubation at 25°C. 1×10^3 - 5×10^5 yeast CFU/g were isolated from 15 samples; 5×10^4 - 1×10^5 yeast CFU/g from 27 samples; 1×10^4 - 5×10^6 yeast CFU/g from 31 samples and 5×10^3 - 1×10^6 yeast CFU/g from 14 samples. *Geotrichum candidum* was almost always isolated together with other yeasts but in lesser numbers. The most prevalent yeasts isolated were *Saccharomyces fragilis*, *Saccharomyces cerevisiae* and *Saccharomyces lactis*. *Saccharomyces fragilis* and *Saccharomyces lactis* ferment lactose and result in sensory changes of the product with formation of gas. The study concluded that strict microbiological control of the basic material, i.e. curd used in the manufacture of these deserts, was required.

Mudretsova-Viss and Zav'yalova (1970) examined the residual microflora of pasteurized milk used for quarg making and found that the predominant bacteria were *Streptococcus bovis*, *Streptococcus thermophilus* and *Streptococcus faecalis*. Occasionally thermophilic lactobacilli and proteolytic bacteria were also encountered. *Escherichia coli* and thermophilic lactobacilli also developed during quarg manufacture. Yeast flora of freshly made quarg consisted chiefly of asporogenous aerobic forms of *Torulopsis* and *Candida* species.

Aleksieva and Mirkov (1977) produced quarg experimentally contaminated with enterococci (*Streptococcus*

faecalis and *Streptococcus faecium*) or coliforms (mixture of *Escherichia coli* and *Aerobacter* species). A *Streptococcus lactis*, *Streptococcus diacetylactis*, *Streptococcus cremoris* starter was used; the fermentation was carried out for 14-18 hours at 28°C. During fermentation, when titratable acidity increased, the numbers of enterococci increased 21-48 fold and the numbers of coliforms increased 17-450 fold. The cut coagulum was drained for 4-5 hours at 11-15°C, homogenized, packaged in nylon film in 250-300 g portions and stored at 8-10 °C for 14 days or at 18-23°C for 6 days. During these steps of manufacture the number of enterococci increased to 95-114 fold and coliforms to 770-2800 fold in numbers.

In a search for the causes of the "over-acidification" defect, soured milk, yogurt and sour milk quarg samples were examined by Kielwein and Melling (1978). These samples were examined for titratable acidity, pH, lactic acid content, enterobacteria, yeasts and molds, *Lactobacillus bifementans* and sensory properties. The only notable difference in composition of sensorically normal and over-acidified sample was the amount of acid-tasting D-lactic acid. Initial contamination with *Pseudomonas* species and other proteolytic microorganisms and secondary contamination with *Lactobacillus bifementans* resulted in appreciable production of D-lactic acid which was considered to be responsible for the above defect.

In a study performed by Tolle (1979), the microflora of quarg was analyzed. The pH averaged 4.6 varying between 4.3 and 4.7. Starter organisms predominated, followed by gram positive saprophytes. A total of 17% of the samples showed the presence of coli-aerogenes bacteria, but the number of enterobacteriaceae was low (1%).

Kielwein (1981) tested 151 samples of fermented milk and discovered that many were contaminated with enterobacteria, enterococci, yeasts or molds. Enterobacteria were found more often in quarg than in yogurt, Kefir or cultured milk products. Of the samples tested, over two-thirds displayed the flavour defect described as "high-acidity". Determination of pH, titratable acidity and lactic acid content revealed that quarg samples with this defect contained significantly more D-lactic acid than normal samples. It was suspected that this increase in D-lactic acid was due to *Lactobacillus bifementans*. It is thought that *Lactobacillus bifementans* occurs in protein deposits in dairy equipment and contaminates the milk after pasteurization. It has been found that its production of D-lactic acid is stimulated by metabolites of *Pseudomonas fluorescens* and other proteolytic bacteria.

2.6 Bitterness Development in Cheese

Development of bitter flavour in quarg is one of the major factors that affects shelf-life and consumer acceptability. Although knowledge is evolving steadily on

bitter flavour in hard cheese, there is hardly any work being done to study the bitterness problem of quarg and other soft unripened cheeses. Study of bitter flavour in quarg and other cheeses is difficult because of the great variety of technological parameters and the extremely complex system of enzymes, substrates and microorganisms present. However, numerous studies have been conducted on bitterness development in gouda and cheddar cheese.

The bitter defect in hard cheese is attributable to the accumulation of bitter tasting peptides formed from casein components during ripening (Harwalkar and Elliot, 1971; Raadsveld, 1953; Richardson and Creamer, 1973). Since a high average hydrophobicity appears to be a common property of bitter peptides (Kleter and Visser, 1977), strongly hydrophobic caseins (Mercier *et al.*, 1972) are extremely susceptible to liberation of bitter peptides on hydrolysis. Visser *et al.* (1983) reported that β -casein, particularly its C-terminal region, forms a principal source of bitter peptides in cheese. The specific nature and amount of milk coagulating agents, type of starter, and final pH of cheese are generally considered to be contributing factors to the development of bitter flavour (Edwards and Košikowski, 1983).

2.6.1 Influence of Rennet

Cheese prepared with rennet or rennet substitutes may become mildly or intensely bitter. This bitterness results

from the proteolytic production of bitter peptides during ripening and storage. Proteolytic activity in cheese is due largely to rennet and starter or contaminating microorganisms. Bitter cheese is more likely to be produced with rapid acid producing culture than slower acid producing strains. Czulak (1959) found bitter flavour most frequently in cheese having a pH less than 5.0 in the first week. He suggested that accumulation of bitter-tasting peptides might be due to increased activity of rennet at low pH levels. Rennet presumably hydrolyses casein to high molecular weight peptides, which are further degraded by the bacteria or rennet to smaller, sometimes bitter peptides. Unless these bitter peptides are further metabolized by the starter organisms, they will accumulate and will produce detectable bitter flavour (Lawrence *et al.*, 1978)

It has been known for a long time that rennet is able to produce bitter peptides from casein. Gouda cheese made with an excess of rennet was often found to become bitter (Stadhouders, 1960). Milk, to which starter culture and rennet had been added, acquired an extremely bitter taste after a short incubation time (Stadhouders, 1962). The studies of Reiter *et al.* (1967) and Visser (1977a) showed that residual rennet in cheese can produce bitter peptides. They conducted experiments with starter-free cheeses, whereby the acid formation was stimulated by the addition of δ -gluconic acid lactone. Bitter peptides were produced in these products. This was also the case when cheese was made

in which a certain amount of rennet, i.e., a quantity normal for Gouda cheese, was retained. When 60% of the normal amount was retained (normal for cheddar cheese) a slightly bitter flavour developed (Visser, 1977a).

In a study by Edwards and Kosikowski (1983), bitter peptides were isolated from cheddar cheese rendered bitter by milk coagulating agents. Bitter peptides produced differed in such properties as elution volume from Sephadex gel, migration on silica gel, and amino acid composition. Bitter peptides contained relatively large amounts of aliphatic, acidic and hydroxy amino acids, but small amounts of basic and aromatic amino acids. The most consistent factors were relatively large amounts of glutamic acid, proline, leucine, valine and the ratio of aliphatic to acidic amino acid (0.8 to 1.3). One would be inclined to deduce from these findings that rennet is capable of direct production of bitter peptides in cheese.

Lawrence *et al.* (1972) prepared cheddar cheese with different amounts of rennet. By reducing the amount considerably below the normal value of 0.22 mL standard rennet per liter cheese milk, the bitterness of the cheeses prepared with 'bitter' starters such as *Streptococcus cremoris* Hp was decreased. No bitterness could be evoked by an increase of two or three times the normal amount of rennet when using 'non-bitter' starters such as *Streptococcus cremoris* AM. From these findings, it was concluded that the major involvement of calf rennet is not

in forming bitter peptides directly, but rather to provide non-bitter peptides which are subsequently degraded to bitter ones by 'bitter' starters. However, the fact that non bitter cheddar cheese can be produced, even when a three fold excess of rennet is used in combination with 'non-bitter' starter, might be also due to the relatively low amount of rennet retained in this kind of cheese (Stadhouders and Hup, 1975). Gouda cheese, when produced under these circumstances ('non-bitter' starters *Streptococcus cremoris* AM, and a three fold excess of rennet) appears to be very bitter (Visser, 1977). Apparently, rennet on its own can produce bitter peptides in cheese. Visser (1977) reported that in normal cheese, the action of rennet clearly stimulates the starter bacteria to accumulate amino acids and low molecular weight peptides. The amount of residual rennet in cheese had a distinct influence on accumulation of soluble nitrogen compounds but not of amino acids. The production of soluble nitrogen was positively correlated with the amount of residual rennet in cheese. Decreasing the amount of rennet used for "bitter"-starter cheeses in parallel experiments resulted in significantly less intense bitterness. Visser (1977) also showed that rennet alone has the potency to produce bitter peptides if high amounts were retained in the cheeses.

The importance of the amount of rennet retained in the cheese for the development of bitter flavour was emphasized by Stadhouders and Hup (1975) and Stadhouders *et al.* (1977).

This amount is not only governed by the amount and type of rennet added to the cheese milk, but also by the manner in which the curd is washed and drained, by the pH of the milk and curd during cheese making, and in particular by the cooking temperature. The lower the pH and cooking temperature, the higher is the amount of rennet retained in the cheese.

2.6.2 Influence of Starter Bacteria

According to Lawrence and Pearce (1968) cheese starters fall into three main groups: (i) "slow" strains (slow acid producers), which give no off-flavours under normal conditions; (ii) "fast" strains (fast acid producers), which have a tendency under normal conditions to produce bitterness; (iii) starters, containing mainly *Streptococcus lactis* strains, which tend to produce varying degrees of fruitiness as well as bitterness. The starters in the last two groups are those which are comparatively "fast" and are therefore the most popular in commercial application. The widely held belief that only *Streptococcus cremoris* strains cause bitterness is not correct since many "fast" strains of *Streptococcus lactis* give bitter cheese (Fryer, 1969).

Emmons *et al.* (1962) were the first group in Canada to carry out a systematic investigation of the effect of the conditions of manufacture on the formation of bitterness in cheese and found that the main factor was the strain of starter organism used. They claimed that some starter

strains which produced bitterness were not greatly affected by pH, but others were affected significantly.

Lowrie *et al.* (1972b) have demonstrated that starter bacteria have a direct role in bitterness development in cheddar cheese, since the use of high cooking temperatures leading to relatively low cell densities reduced bitterness development. A lower cell density in the cheese will result in reduced levels of both cell wall-associated starter proteinase (Thomas *et al.*, 1974; Exterkate, 1975) and intercellular proteinase and peptidases (Exterkate, 1975; Mou *et al.*, 1975). It is presumed that starter bacteria lyse during cheese ripening and their intercellular enzymes are released into the curd matrix. Mills and Thomas (1981) reported that the cell wall-associated proteinase plays a role in the formation of bitter peptides.

Gordon and Speck (1965) have established that certain starter bacteria are capable of forming bitter peptides. In another study it was observed that certain strains known as 'non-bitter' do not develop very well if cooking temperatures of 36-39°C are used, and consequently reach a lower final number in the cheese than do certain strains known as 'bitter' ones (Martley and Lawrence, 1972; Lowrie *et al.*, 1972a; Lowrie and Lawrence, 1972). Visser (1976) prepared rennet-free cheese with 'bitter' strains and found that the cheeses turned out to be bitter. In a similar study Visser (1977) observed that the bitterness development was more intense when cheeses were aseptically made with the

same starter strains and with the normal amounts of rennet.

These cumulative effects may indicate a concerted but separate action of rennet and certain starter bacteria in the formation of bitter peptides in cheese.

The fact that the intensity of bitter flavour can diminish during ripening (Stadhouders and Hup, 1975; Stadhouders, 1978) indicates that certain strains of starter bacteria have the ability to degrade bitter peptides. Since normal cheese containing both rennet and starter bacteria is generally not bitter, despite the fact that rennet is capable of forming bitter peptides, degradation of these peptides in non-bitter cheese by starter bacteria and/or their enzymes is one obvious explanation (Stadhouders *et al.* 1983).

A Study of Lowrie and Lawrence (1972) indicated that the degradation of casein by rennet produced a pool of high molecular weight non-bitter peptides, some of which were hydrolysed by proteases of starter *Streptococci* to low molecular weight bitter peptides. These bitter peptides were further degraded to non-bitter peptides and amino acids by peptidases of starter *Streptococci*. Visser *et al.* (1983) have shown that degradation of bitter peptides occurs by the action of proteolytic enzymes from the cytoplasmic membrane of bacterial cells, either in concert with enzymes from the cytoplasm, or without their aid.

Kaneko and Yoneda (1974) studied the cause of bitterness development in yeast ripened cheese. The cheese

was prepared with 2.5% *Streptococcus cremoris* and 2.5% *Streptococcus lactis*, together with a lactose fermenting yeast (5% *Saccharomyces fragilis*). The cheese was found to be very prone to bitter flavour development. However, using a non-bitter *Lactobacillus* strain the authors were successful in making yeast-ripened cheese without any bitterness.

Visser *et al.* (1983) have studied the effect of salt content and ripening temperature on the development of bitter flavour. Salt strongly influenced the net result of formation and degradation of bitter peptides by bacterial cells in gouda cheese. The higher the salt content, the lower the bitter flavour score (Visser *et al.*, 1983; Stadhouders, 1962; Tuckey and Ruehi, 1940; Lawrence and Gilles, 1969). Stadhouders (1975) studied the effect of ripening temperature on bitterness in gouda cheese and found that cheese ripened at 16°C had a somewhat more bitter flavour than cheese ripened at 6°C.

2.6.3 Influence of Psychrotrophic Organisms

Psychrotrophic organisms or their enzymes excreted in raw milk can give rise to various defects in cheese. While these organisms usually do not survive pasteurization, the lipases and proteases they produce do. These enzymes are carried into cheese and in excess could produce bitterness and rancidity. Moskowitz (1980) and Ohren and Tuckey (1969) observed that good quality cheeses were made from milk

having a psychrotrophic count between 1×10^3 - 2×10^4 CFU/mL and lower. Hattowska (1970) reported that psychrotrophic growth in milk before pasteurization and cheese making produces enzymes that inhibited acid and flavour development in cheese. Psychrotrophs produce off-flavours and odors during growth in stored, refrigerated raw milk which carry over into the finished product. The flavours produced by psychrotrophic bacteria in milk and dairy products are bitter, fruity, putrid, rancid and acidic (Mikolajcik, 1979). In another study, Juffs (1974) used protease from *Pseudomonas* species as coagulants of milk for cheddar cheese manufacture. It was found that the body of the cheese made with protease as coagulant was soft because whey expulsion was reduced and proteolytic activity decreased with the increase in the acidity. These cheeses were found to have a bitter and unclean flavour.

2.7 Nutritional Value of Quarg

Quarg is a soft, unripened fresh cheese. It is nutritious and easily digestible. The nutritional value of quarg is presented and compared with other dairy products in Table 2.2.

According to regulations in West Germany (Czulak and Hammond, 1973), skim milk quarg should contain not less than 18% (w/w) of milk solids. Fat enriched varieties have also become popular; some products being marketed may contain 10%, 20% or 40% of fat in dry matter. Whether low-fat or

Table 2.2 Nutritional value of different dairy products (per
100 g)

Products	Kilojoules	Protein (g)	Carbohydrates (g)	Fat (g)
Milk	270	3.3	4.9	3.6
Yogurt	286	5	7	2
Full-fat Quarg	508	11	4	7
Cottage cheese	437	12	3	5
Cream cheese	1487	8	3	35
Sour cream	559	3	8	8
Mayonnaise	2999	trace	trace	79
Margarine	3024	trace	trace	81
Butter	3276	3.2	3.2	82

Source: Renner (1983)

fat-enriched, quarg made in the traditional way by syneresis of acid-coagulated casein has only about twice the solids content of milk and has lost many valuable milk components such as whey proteins, lactose and water-soluble vitamins and minerals.

Kruglova and Bubnova (1962) conducted studies on the possible enrichment of quarg with vitamin B₁₂. Special cultures of propionic and lactic acid bacteria in equal proportions (5% total inoculum) were used in quarg production for vitamin B₁₂ enrichment of the product. The average content of vitamin B₁₂ in 3 batches of quarg, each from 1,000 Kg milk, was 130.1 µg/kg versus 12.3 µg/Kg for the control quarg made with normal starter. The flavour and consistency of the enriched quarg were as good as those of the control product. On storage at 0.5°C, the vitamin B₁₂ content of the experimental quarg declined with time, by 22% after 7 days and by 34% after 2 weeks, while that of the control quarg showed some increase.

Lipatov *et al.* (1974) produced quarg by direct acidification of milk with lactic acid. No difference in protein, P, Ca contents and nutritive value was observed when compared with quarg manufactured by the conventional batch method.

Korolczuk, *et al.* (1975) examined the effect of heat treatment of milk on the nutritional value of quarg. Quarg samples were taken from commercial production, where the milk was pasteurized for 15 sec at 75°C, as well as from

experimental production, where the milk was heated either for 20 min at 65°C or for 5 min at 95°C. The protein was precipitated by acidification using either microbiological or chemical agents (hydrochloric acid or lactic acid at pH 4.6 and 35-40°C), or by rennet and CaCl₂ (0.2% at 95°C).

Protein retention in quarg produced from milk heated at 75° or 65°C was at the 77-79% level, increasing to 88-91% at 95°C. This was caused by the transfer to the quarg of about 90% of the β -lactoglobulin and 60% of the α -lactalbumin. The main change in the amino acid composition was an increase in the content of cystine to 1.3g/16g N in quarg from high-heat milk compared with 0.7g/16g N from low-heat milk. Riboflavin retention with microbial acidification of the milk was at 35% level, compared to 23% for chemical precipitation of the protein regardless of the temperature and time of heating. The production of quarg from high heat milk should be increased because of the higher protein retention and higher nutritional value. The PER value of α -lactalbumin and β -lactoglobulin are 4.0 and 3.5 respectively, whereas that of casein is 2.5 (Renner, 1983).

Review of literature revealed that very limited research has been carried out on bitterness development during storage and on the shelf-life problem of quarg. Most of the literature articles cited are from Europe where the problems of bitterness and extended shelf-life are not of great concern since quarg is not stored more than 5-7 days after production and is consumed fresh. Some of the research

papers are in the form of patents while others are in languages other than English which makes it difficult to obtain detail information on various aspects of quarg. Quarg is a relatively new dairy product in Canada and other areas of North America and as such very little data on production, storage, consumption and other aspects of quarg are available. It was therefore felt necessary to undertake this study in order to generate research data on some of the important aspects of quarg which affects its storage and marketing in this part of the world.

3: MATERIALS AND METHODS

3.1 Experimental Approach

This study was divided into four major sections :

1. A preliminary study to compare the development of bitterness in quarg produced both commercially and on a laboratory scale. This was also done to indicate the possible causes of bitterness development in quarg during storage.
2. A laboratory investigation of the effect of rennet level on bitterness development in quarg, including confirmation of the major findings by industrial trials.
3. A laboratory comparison of four different starter cultures relative to bitterness development in quarg.
4. A consumer acceptance study of plain and fruit quarg to evaluate consumer reaction and to predict market potential of this relatively new dairy product in Canada.

3.2 Materials

Various dairy supplies used in production and analyses of quarg were obtained from local industrial sources. Most of the laboratory processing experiments were carried out at the Department of Food Science, University of Alberta.

3.2.1 Milk

Homogenized and pasteurized (72°C, 22 sec) milk was obtained from Lucerne Foods Ltd., Edmonton as needed. All batches were analysed by the Alberta Agriculture Food Laboratory. The average butter fat content of the milk was 3.45%, the average standard plate count (SPC) was less than 33 CFU/mL, coliform count was less than 10 CFU/mL and the antibiotic test of milk was negative.

3.2.2 Starter Culture

Freeze dried lactic culture (Flora Danica) was obtained from Horan-Lally Co. Ltd., Mississauga, Ontario. The starter culture consisted of *Streptococcus lactis*, *Streptococcus cremoris* and *Streptococcus diacetilactis* as indicated by the supplier. Other cultures used were Flora Danica without *S. diacetilactis* and single strain cultures (#134 and #584) of *S. cremoris*.

3.2.3 Rennet

Single strength pure calf-rennet was obtained from Horan-Lally Co. Ltd., Mississauga, Ontario. The rennet was sent for the estimation of its activity to Dr. R.J. Brown at Department of Nutrition and Food Science, Utah State University, Logan 84322, USA. The rennet activity was estimated to be 88 rennin units per mL. The method employed for the activity estimation is described in the paper by

Longman Building, 6905-116 Street, Edmonton T6H 2N2

McMahon and Brown (1982).

3.2.4 Fruit Pulp

"Tropical fruit" pulp (mixture of papaya, coconut, pineapple, and guava) used for making fruit quarg in the consumer acceptance study, was obtained from Lucerne Food Ltd., Edmonton.

3.2.5 Microbiological Media

The media used for microbiological analyses, obtained from Difco Laboratories, Detroit, Michigan, USA, were :

1. Standard Plate Count (SPC) Agar, for total bacterial count.
2. Violet Red Bile (VRB) Agar, for coliform count.
3. Potato Dextrose (PD) Agar, for yeast and mold count.

3.2.6 Chemicals

Chemicals such as Tartaric acid, Sodium hydroxide, Phenolphthalein indicator and Sulfuric acid used in chemical analyses were obtained from Fisher Scientific Company, Fair Lawn, New Jersey 07410, USA.

3.3 Preparation of Mother Culture

Freeze dried lactic culture (Flora Danica), supplied by Horan-Lally, was used to make the mother culture used in the laboratory production of quarg. The culture was grown in 10% reconstituted skim milk. The milk was heated at $90 \pm 2^\circ\text{C}$ for

one hour and cooled to 22°C before inoculating with the freeze dried culture. The culture was set in 500 mL Erlenmeyer flask containing 200 mL of reconstituted skim milk.

The flask, containing milk and lactic culture, was incubated at 22±1°C for 16 hours until the pH dropped to 4.8-5.0. This culture was immediately transferred to a refrigerator unless used directly to make quarg. The above process of reinoculation of culture was repeated three to four times before using it for the production of quarg. The amount of culture used as an inoculum was 1% of milk in each batch of quarg made in the laboratory.

3.4 Preparation of Rennet Solution

Required quantities (0, 388, 775, 1550 and 3876 units per 1000 kg) of the single strength calf rennet were diluted 40 times with water before adding to the milk.

3.5 Laboratory Production of Quarg

The assembly used for producing quarg on a laboratory scale consisted of a thermostat (supplied by M/S Blue M Electric Company, Blue Island, Illinois, USA), two stainless steel containers of 10 liter capacity, a pH meter and a recorder. The pH meter used was a Fisher 520 model. Cheese cloth was used to separate the whey from the curd. The entire assembly is shown in Figures 3.1 a, b, c, d.

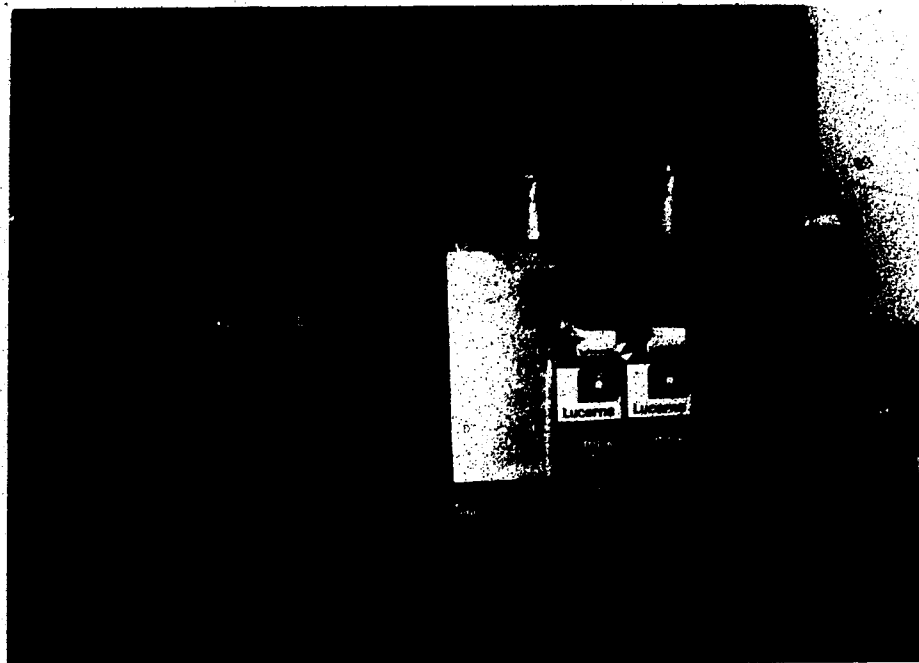


Figure 3.1 a-d Quarg Producing Assembly



Figure 3.1 (contd.) Quarg Producing Assembly

Quarg samples produced commercially by a local cheese plant were used for preliminary and consumer acceptance studies. For the rest of the experiments, quarg was produced at the Department of Food Science, University of Alberta according to the procedure shown in Figure 3.2.

All the vessels, utensils, draining pail and cheese cloth used in the quarg production were sanitized with live steam. Homogenized, pasteurized milk obtained from Lucerne Foods Ltd. in Pure Pak cartons was transferred to the stainless steel container and was heated to about 30°C. This was then inoculated with 1% starter culture, prepared before, and was incubated at 30±1°C in a thermostatically controlled waterbath. An appropriate rennet solution was added after 1 hour of incubation and the milk was held at 30°C for 8-9 hours till the pH dropped to 4.5.

The whey was drained by using a perforated plastic pail covered with three layers of cheese cloth. The coagulum was transferred from the stainless steel containers to the cheese cloth with a stainless steel spoon to avoid any physical damage to the curd. The whey was drained at room temperature (22±1°C). The curd was mixed manually using a stainless steel spoon and packed in Whirl Pack (500 mL) plastic bags. These bags were then stored at predetermined temperatures for the shelf-life study. Four liter quantities of milk were used for each batch of quarg made in the laboratory.

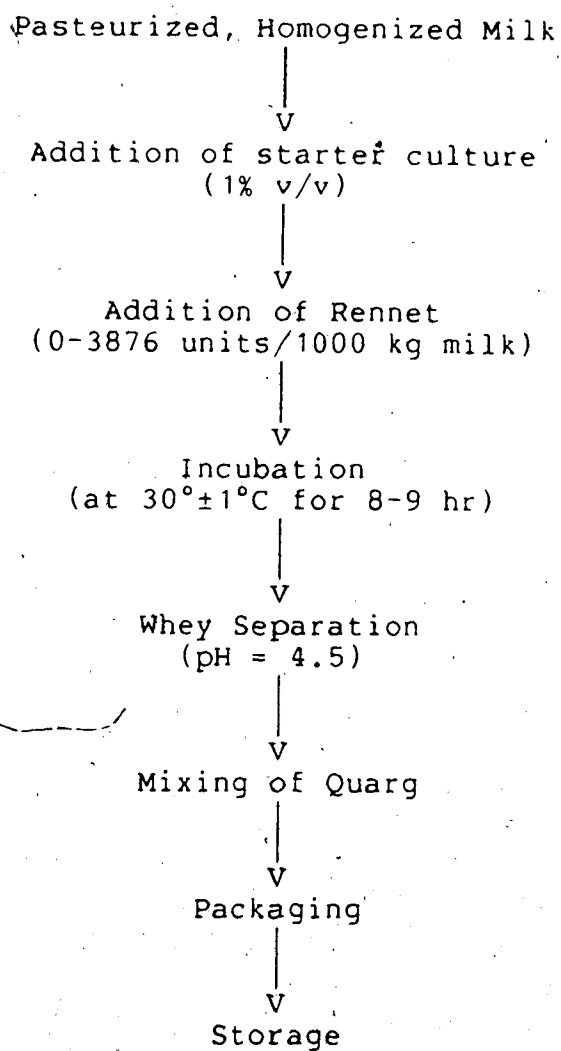


Figure 3.2 Flow-diagram of quarg production.

3.6 Sampling of Quarg

Quarg samples were mixed homogeneously by manual manipulation of the Whirl Pak bag. Test samples were obtained with a sterile spatula (sterilized by dipping in alcohol followed by flaming) and were used for microbiological and chemical analyses. The fresh (zero time) samples were obtained immediately after production in the laboratory.

3.7 Microbiological Analyses

Conventional pour plate techniques used for the microbiological analyses (APHA, 1978) were as follows:

3.7.1 Psychrotrophic Count

Psychrotrophs were estimated by plating the quarg samples with Difco plate count Agar and incubating at 7°C for 10 days.

3.7.2 Coliform Count

Coliforms were enumerated by plating the quarg samples with Difco violet red bile Agar and incubating at 32°C for 24±1 hour. The solidified poured violet red bile Agar plates were overlaid with an additional 3 to 4 mL of plating medium before incubation to prevent surface colony formation.

3.7.3 Yeast and Mold Count

Poured plates of Difco potato dextrose agar, acidified to pH 3.5 with tartaric acid were prepared with quarg and incubated at room temperature ($22\pm 1^\circ\text{C}$) for three to five days.

3.8 Chemical Analyses

The various chemical analyses carried out included estimation of moisture, fat, protein, titratable acidity and pH.

3.8.1 Moisture content

Moisture content of quarg was estimated following the AOAC procedure # 16.233 (AOAC, 1980). Duplicate samples of 2.5-3.0 g were predried for 10-15 minutes on a steam bath and then placed in a forced air oven at 100°C for three hours. The samples were then cooled and weighed. The amount of moisture was determined by subtracting the weight of total solid from the weight of the sample and converting to percentages.

3.8.2 Butterfat

Fat estimation was done by the Babcock procedure. Nine grams of sample was weighed in a 50 mL beaker and diluted with 2 mL of distilled water. Fifteen mL of concentrated sulphuric acid was added to the diluted sample and mixed with a glass rod. The mixture was transferred to a 20%

Babcock cream bottle and the beaker rinsed with an additional 3 mL concentrated sulphuric acid. Thereafter, the usual procedure for the babcock test was followed as described in APHA (1978).

3.8.3 Protein

Protein in quarg was estimated by measuring total nitrogen following the Kjeldahl method (AOAC # 16.245, 1980). The total protein content of quarg was determined by multiplying the percent nitrogen with a factor of 6.38.

3.8.4 Titratable Acidity

Estimation of titratable acidity was carried out following the AOAC procedure # 16.023 (AOAC, 1980). Ten grams of quarg sample and 80 mL of water were mixed in a beaker with a glass rod and titrated with 0.1N sodium hydroxide solution. Phenolphthalein solution (1%) was used as an indicator. The end point was reached when the colour changed from milky white to faint pink. The titratable acidity expressed as percent lactic acid is given by

$$\% \text{ Lactic Acid} = \text{mLs NaOH} \times 0.09$$

3.8.5 Determination of pH

Fifteen to twenty grams of quarg were placed in a 50 mL beaker. The pH meter (Fisher model # 520) was adjusted using standard pH solution (lower standard of pH 4 and upper standard of pH 7) and pH of quarg sample was measured and

recorded.

3.9 Sensory Evaluation of Quarg

3.9.1 Preliminary Study

The purpose of this study was to compare the development of bitterness in quarg produced commercially and on a laboratory scale and also to ascertain the possible causes of bitterness development during storage. Quarg was produced at an industrial cheese plant and at the Department of Food Science, University of Alberta. In this study, the triangle test (Larmond, 1977) was employed to identify the odd sample in quarg produced commercially and on a laboratory scale. The degree of difference (slight, moderate, much, extreme) was noted by panelists on a standard form (Appendix 1). This was an untrained panel consisting of men and women, randomly selected from staff and student members of the Department of Food Science, University of Alberta. These panelists were further screened and trained for sensory analyses carried out in subsequent investigations.

All sensory evaluations took place in individual testing booths illuminated with incandescent light. Morning sessions (10:00-11:30) were used.

Quarg samples were prepared immediately after being removed from the refrigerator and were served to the panelists with a spoon in polyethelene sample cups. The test

samples were identified by three-digit random numbers and the participants were instructed as to the order in which the quarg samples were to be tasted. Tap water was provided for mouth rinsing.

3.9.2 Panel Selection and Training

The panel screening and selection for subsequent sensory analyses were based on the ability of the panelists to recognize bitterness. This was examined by presenting each candidate with five quarg samples (slightly bitter, bitter, very bitter, extremely bitter, and a good fresh sample of quarg [not bitter] as evaluated subjectively by the author and an experienced dairy judge). These quarg samples (excluding the fresh sample) were prepared by storing fresh quarg at 7°C for different periods of time until bitter taste developed. These samples were presented to the panelists who were asked to recognize and record the degree of bitterness in randomly coded samples on the ballot provided (Appendix 2).

The selected panelists were trained to improve their ability to detect bitter taste and to discriminate among intensities of bitterness. They were provided with bitter water samples prepared with quinine sulphate. The appropriate solutions were made from 0.02 g of quinine sulphate in one liter of water. Four different water samples (slightly bitter, bitter, very bitter and extremely bitter) were presented to the panelists to taste and improve their

sensation of bitterness. These samples were then stored in the cooler for the panelists to try on their own whenever they felt the need to retest their taste buds. Group sessions were held with the purposes of agreeing upon definitions, clarifying the mechanics of testing procedures and sharing cues in perception of flavour parameters.

Six candidates were selected on the basis of their performance in panel screening and training. This selected panel of six judges evaluated the quarg samples on a bitterness scale of 1-6 in subsequent sensory analysis sessions.

3.9.3 Sensory Evaluation of Stored Quarg

The six selected panelists were students and staff of the Department of Food Science. All screening sessions and subsequent taste testing sessions were conducted in individual booths in an air conditioned room. Test session judgements were made under incandescent light.

The arrangement of the sensory evaluation room, and coding procedure of the samples were as described before. Duplicate samples were presented to each of the panelists who were then asked to grade the samples according to the intensity of bitterness. The taste panel form provided to the panelists was the same as used in the training sessions as shown in Appendix 2. Sensory data on bitterness were collected at weekly intervals over a period of four weeks. Analysis of variance and comparison of means by Tukey's test

were performed on the sensory data using the University computer facilities with standard statistical package.

3.10 Consumer Acceptance Study

A consumer study was carried out to assess the acceptability and consumer preference for fruit or plain quarg in the City of Edmonton. A representative sample of consumers was used in this study by delivering the quarg in person to randomly selected households.

3.10.1 Household Selection Procedure

The sample of consumers was selected using the Henderson's directory of households for Edmonton. Forty numbers were taken from a random number table and the addresses corresponding to those particular numbered locations on the same numbered page were used as starting points for clusters of 15 alternate households.

The cluster method of sampling was chosen as being most economical and logistically feasible. According to Emory (1976), a 30% return in most mail-in surveys is considered satisfactory. Accordingly, a sample size of 600 was arbitrarily selected with allowance for non-response, so that the actual sample would be close to 200. This would constitute a 33% return.

3.10.2 Preparation of Quarg Samples

The plain and fruit quarg used in the consumer survey were commercially made by a local cheese plant in Alberta. The method used in making the flavoured cheese was the same as for plain quarg, except that tropical fruit pulp (used normally in yogurt) was added to the plain quarg at a rate of 20% w/w of quarg and mixed prior to packaging. The quarg samples of 125 g were packed in plastic tubs. The containers were plain white without any label or instructions.

3.10.3 Survey Method and Questionnaire

Samples of both plain and fruit quarg were distributed to the randomly selected households in the city along with a questionnaire (Appendix 3) which was to be completed and mailed back to the Department of Food Science, University of Alberta in a stamped, self-addressed envelope. The householders were advised to taste the products, grade them and return the questionnaire after answering the given questions. They were told to use the samples as they wished and report when and how they tried the products. They were also asked to report whether they had tried similar products before and whether they would buy the products if they were available at a reasonable price. Each individual member of the household was asked to express his or her liking for the product on a six point scale and mail back the questionnaire to the Department of Food Science. They were given two weeks to reply, but the questionnaires were received until the

fourth week. The results of the consumer study were tabulated and analysed statistically.

4. RESULTS AND DISCUSSION

4.1 Preliminary Study

Quarg was obtained from a local commercial cheese processing plant in Alberta in an attempt to identify the possible causes of quality deterioration in the product. At the same time, quarg was also produced on a laboratory scale in the Department of Food Science, University of Alberta, simulating the commercial manufacturing process (Figure 3.2). The rennet level used in the laboratory production was the same as in the industry, i.e., 3876 units per 1000 kg of milk with 1% starter culture (Flora Danica). The chemical composition of the quarg produced and of that obtained from the local cheese plant is shown in Table 4.1.

The quarg samples (commercial and laboratory produced) were stored at two different temperatures (5°C and 10°C) for 8 weeks. These two temperatures of storage were selected on the basis of 5°C being the normal refrigeration temperature expected in retail outlets and 10°C as a possible extreme deviation from normal refrigeration temperature. The stored samples were analysed at intervals of 0, 3, 5, 6, 7 and 8 weeks to assess the changes in the predominant microflora, pH and lactic acid content (Tables 4.2, 4.3, 4.4 and 4.5).

Sensory analyses were carried out to assess the bitterness development over a period of six weeks. The results are shown in Table 4.6 which indicates that 7-8 panelists out of 10 could identify the odd (bitter) sample

Table 4.1 Composition of 'quarg' produced on a laboratory scale and commercially using 3876 units rennet per 1000 kg milk and 1% starter culture (Flora Danica).

Components	Laboratory Scale	Commercial
	(%)	(%)
Moisture	73.66 ± 0.70	73.02 ± 0.54
Lactic Acid	1.10 ± 0.03	1.19 ± 0.02
Protein	10.03 ± 0.20	9.87 ± 0.18
Butterfat	12.35 ± 0.29	13.20 ± 0.42
pH.	4.43	4.40

Above figures are average ± standard deviation of 6 replicates performed on 3 samples

Table 4.2 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 5°C. The quarg was produced commercially using 1% starter culture (Flora Danica) and 3876 units rennet per 1000 kg of milk.

Storage time Weeks	PBC/mL ¹	Presumptive Coliform/mL	Yeasts and molds/mL	pH	Lactic acid (%)
0	1.2x10 ⁴ (3.0x10 ³ -5.9x10 ⁴)	<10	3.5x10 ⁷ (1.5x10 ⁷ -7.6x10 ⁷)	4.44	1.19
3	3.6x10 ⁴ (3.0x10 ³ -1.5x10 ⁵)	<10	6.5x10 ⁷ (1.0x10 ⁷ -2.1x10 ⁸)	4.41	1.15
5	1.3x10 ⁴ (3.0x10 ³ -4.7x10 ⁴)	<10	6.8x10 ⁷ (1.0x10 ⁷ -1.8x10 ⁸)	4.36	1.16
6	4.7x10 ⁴ (3.0x10 ³ -2.0x10 ⁵)	<10	1.9x10 ⁸ (1.0x10 ⁷ -2.1x10 ⁸)	4.42	1.24
7	4.9x10 ⁴ (3.0x10 ³ -2.3x10 ⁵)	<10	1.9x10 ⁸ (1.0x10 ⁷ -9.4x10 ⁸)	4.31	1.31
8	3.2x10 ⁴ (3.0x10 ³ -1.5x10 ⁵)	<10	1.1x10 ⁸ (1.0x10 ⁷ -4.8x10 ⁸)	4.27	1.58

¹ PBC values in parentheses indicate range of counts from three samples.

² PBC = Psychrotrophic bacterial count.

Table 4.3 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 10°C. The quarg was produced commercially using 1% starter culture (Flora Danica) and 3876 units rennet per 1000 kg of milk.

Storage time Weeks	PBC/mL	Presumptive Coliform/mL	Yeasts and molds/mL	pH	Lactic acid (%)
0	1.2x10 ⁴ (3.0x10 ³ -5.9x10 ⁴)	<10	3.5x10 ⁷ (1.5x10 ⁷ -7.6x10 ⁷)	4.44	1.19
3	3.1x10 ⁵ (3.0x10 ³ -1.3x10 ⁵)	<10	1.3x10 ⁸ (2.4x10 ⁷ -6.0x10 ⁸)	4.40	1.25
5	3.7x10 ⁵ (3.0x10 ³ -1.4x10 ⁵)	<10	3.3x10 ⁸ (1.0x10 ⁸ -1.2x10 ⁹)	4.32	1.33
6	5.4x10 ⁵ (3.0x10 ³ -1.3x10 ⁵)	<10	3.0x10 ⁸ (1.0x10 ⁸ -9.1x10 ⁸)	4.17	1.55
7	2.4x10 ⁶ (3.0x10 ³ -9.1x10 ⁶)	<10	1.3x10 ⁹ (3.0x10 ⁷ -2.9x10 ⁹)	4.12	1.57
8	----				

* Samples discarded due to microbial spoilage.

All values in parentheses indicate range of counts from three samples.

Table 4.4 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 5°C. The quarg was produced on a laboratory scale using 1% starter culture (Flora Danica) and 3876 units rennet per 1000 kg of milk.

Storage time Weeks	PBC/mL	Presumptive Coliforms/mL	Yeasts and molds/mL	pH	Lactic acid (%)
0	1.2x10 ⁷ (3.0x10 ⁷ -3.0x10 ⁷)	<10	<10	4.43	0.95
3	4.0x10 ⁷ (3.0x10 ⁷ -5.6x10 ⁷)	<10	3.3x10 ⁷ (1.0x10 ⁷ -9.0x10 ⁷)	4.21	1.24
5	1.4x10 ⁷ (3.2x10 ⁷ -3.2x10 ⁷)	<10	2.0x10 ⁷ (1.0x10 ⁷ -4.0x10 ⁷)	4.21	1.27
6	2.4x10 ⁷ (4.1x10 ⁷ -6.1x10 ⁷)	<10	1.3x10 ⁷ (1.0x10 ⁷ -3.6x10 ⁷)	4.20	1.18
8	4.6x10 ⁷ (2.2x10 ⁷ -8.0x10 ⁷)	<10	3.0x10 ⁷ (1.0x10 ⁷ -7.0x10 ⁷)	4.20	1.22

All values in parentheses indicate range of counts from three samples.

Table 4.5 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 10°C. The quarg was produced on a laboratory scale using 1% starter culture (Flora Danica) and 3876 units rennet per 4000 kg of milk.

Storage time Weeks	PBC/mL	Presumptive coliform/mL	Yeasts and molds/mL	pH	Lactic acid (%)
0	1.2x10 ⁷ (3.0x10 ⁶ -3.0x10 ⁷)	<10	<10	4.43	0.95
3	2.5x10 ⁶ (1.9x10 ⁶ -3.7x10 ⁶)	<10	4.7x10 ¹ (1.0x10 ¹ -1.6x10 ¹)	4.23	1.30
5	9.7x10 ⁵ (0-9.7x10 ⁵)	<10	<10	4.18	1.51
6	1.2x10 ⁶ (0-1.2x10 ⁶)	<10	<10	4.21	1.50
8	----				

* Samples discarded due to microbial spoilage

All values in parentheses indicate range of counts from three samples.

Table 4.6 Effect of storage at two different temperatures (5° and 10°C) on bitterness development in quarg produced on a laboratory scale and commercially using 1% starter culture (Flora Danica) and 3876 units rennet per 1000 kg milk.

Storage time (weeks)	Number of Panelists' (out of 10) that could detect bitterness in quarg produced			
	On Laboratory Scale		Commercially	
	5°C	10°C	5°C	10°C
0	0	0	1	1
3	7	10	8	10
6		---*	7	---*

* Samples were discarded due to microbial spoilage

'Untrained panel

of quarg stored for 3 weeks at 5°C, whereas all the panelists could detect bitterness in samples stored at 10°C. In the triangle test, 7 correct judgements from 10 panelists are significant at the 5% level and 8 correct judgements out of 10 panelists are significant at the 1% level (Larmond, 1977). Therefore, bitterness development in quarg was significant after 3 weeks of storage at 5°C and highly significant in samples stored at 10°C after 3 weeks of storage. Bitterness in samples stored for 6 weeks at 5°C were highly significant.

The yeast and mold counts of commercial quarg samples stored at the two temperatures (5° and 10°C) were considerably higher than in the samples produced in the laboratory. The psychrotrophic counts (PBC) in both cases at two different temperatures were, almost identical, in the order of 10⁴/mL and the coliform count was always below 10/mL (Tables 4.2-4.5).

It was evident from these findings that microbial contaminants such as yeasts, molds and psychrotrophs may have been responsible for bitterness development in the quarg during storage. Other potential causes of this quality defect could have been rennet level, culture composition and culture strain (Edwards and Kosikowski, 1983). Further studies were carried out to determine which of the above may be the main factors for bitterness development in the quarg during storage.

A survey of refrigeration temperatures at different retail outlets conducted by the author indicated that the storage temperature for dairy products varied between 5°C to 9°C. It was therefore decided to carry out further storage studies at an average temperature of 7°C.

4.2 Effect of Rennet Level on Bitterness in Quarg

It has been established by several researchers that rennet is able to produce bitter peptides by hydrolysing casein which is extremely sensitive to bitter flavour development on proteolysis. In general, it is thought that rennet is mainly responsible for the breakdown of para-casein to the larger peptides, while starter bacteria liberate smaller peptides and amino acids.

In order to unravel the action of rennet on the development of bitterness in quarg, varying amounts of rennet were used in parallel quarg making experiments with 1% starter culture (Flora Danica). Five batches of quarg in duplicate were produced in the laboratory using 0, 388, 775, 1550 and 3876 units rennet per 1000 kg of milk and 1% starter culture (Flora Danica). The chemical composition of each batch of quarg produced is shown in Table 4.7. Following production, quarg samples were stored at 7°C for 4 weeks and were analysed at weekly intervals to follow the effect of storage on predominant microorganisms, pH and lactic acid content.

Table 4.7 Composition of anarg produced with 1% starter culture (Flora Danica) and different levels of rennet.

Components	rennet level (units/1000 kg milk)				
	388	775	1550		
	0		3876		
Moisture	78.90 ± 0.24	77.60 ± 0.32	73.71 ± 0.12	73.76 ± 0.56	76.18 ± 0.47
Lactic Acid	1.08 ± 0.03	1.16 ± 0.04	1.16 ± 0.01	1.18 ± 0.03	1.16 ± 0.02
Protein	6.33 ± 0.04	7.51 ± 0.32	9.10 ± 0.13	8.91 ± 0.11	7.45 ± 0.56
Butterfat	8.85 ± 0.97	10.70 ± 0.27	12.30 ± 0.77	12.75 ± 0.44	11.23 ± 0.34
pH	4.32	4.28	4.24	4.25	4.4
yield (%)	18.05	21.90	25.50	24.65	28.42

The above figures are average ± standard deviation of 4 replicates performed on 2 samples

The results of the storage study (Tables 4.8-4.12) indicate low level of psychrotrophs, yeasts and molds, suggesting that the bitterness development in quarg was not the outcome of microbial contaminants. This observation is in agreement with Moskowitz (1980) and Ohren and Tuckey (1969) who suggested that good quality cheese can be produced from milk having a psychrotrophic count between 1×10^3 - 2×10^4 CFU/mL and lower.

The stored samples were scored at weekly intervals by a panel of six trained judges on a bitterness scale of 1 to 6 and their individual scores are shown in the Appendix 4. Table 4.13 shows the mean bitterness score of the six panelists; as seen, the score decreased as the amount of rennet used for making quarg was reduced from 3876 units to 0 unit per 1000 kg of milk. This decrease in bitterness score might have resulted from a reduced level of rennet retained in quarg when a lower amount was used in quarg making, confirming the findings of Stadhouders and Hup (1975). The analysis of variance was performed on the sensory data and the result is presented in Table 4.14 which shows a highly significant difference in bitterness development during storage in quarg produced using five different rennet levels. A comparison of means (0-4 weeks of storage) for different rennet levels was carried out using Tukey's test and the results in Table 4.15 indicate that bitterness development in quarg produced with 0 unit and 388 units rennet per 1000 kg of milk differed significantly from

Table 4.8 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 7°C. The quarg was produced on a laboratory scale using 1% starter culture (Flora Danica) and 0 mL rennet per 1000 kg of milk.

Storage time (Weeks)	PBC/mL	Presumptive Colifag ^o /mL	Yeasts and molds/mL	pH	Lactic acid
0	3x10 ⁷ (3.0x10 ⁷ -3.0x10 ⁷)	<10	<10	4.30	1.08
1	1.3x10 ⁸ (3.8x10 ⁷ -2.6x10 ⁸)	<10	<10	4.22	1.03
2	2.1x10 ⁸ (1.8x10 ⁸ -2.4x10 ⁸)	<10	<10	4.21	1.03
3	1.1x10 ⁸ (6.0x10 ⁷ -1.6x10 ⁸)	<10	3.5x10 ¹ (1.0x10 ¹ -6.0x10 ¹)	4.23	1.04
4	1.8x10 ⁸ (3.2x10 ⁷ -3.3x10 ⁸)	<10	1.5x10 ¹ (1.0x10 ¹ -2.0x10 ¹)	4.17	1.06

All values in parentheses indicate range of counts from three samples.

Table 4.9 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 7°C. The quarg was produced on a laboratory scale using 1% starter culture (Flora Danica) and 388 units rennet per 1000 kg of milk.

Storage time (Weeks)	PBC/mL	Presumptive Coliform/mL	Yeasts and molds/mL	pH	Lactic acid (%)
0	4.0×10^7	<10	<10	4.28	1.15
1	(3.0×10^7 - 4.9×10^7) 5×10^7	<10	<10	4.23	1.06
2	(2.5×10^7 - 7.6×10^7) 2.6×10^8	<10	<10	4.22	1.08
3	(1.5×10^7 - 5.0×10^7) 4.0×10^7	<10	<10	4.24	1.10
4	(1.0×10^7 - 7.0×10^7) 1.4×10^7	<10	<10	4.24	1.13
	(3.0×10^7 - 2.5×10^7)				

All values in parentheses indicate range of counts from three samples.

Table 4.10 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 7°C. The quarg was produced on a laboratory scale using 1% starter culture (Flora Danica) and 775 units rennet per 1000 kg of milk.

Storage time (Weeks)	PBC/mL	Presumptive Coliform/mL	Yeasts and molds/mL	pH	Lactic acid (%)
0	3.0x10 ⁷ (3.0x10 ⁷ -3.0x10 ⁷)	<10	<10	4.23	1.16
1	2.0x10 ⁷ (1.4x10 ⁷ -2.6x10 ⁷)	<10	<10	4.19	1.17
2	3.0x10 ⁷ (3.0x10 ⁷ -3.0x10 ⁷)	<10	<10	4.19	1.25
3	3.0x10 ⁷ (3.0x10 ⁷ -3.0x10 ⁷)	<10	<10	4.19	1.24
4	3.0x10 ⁷ (3.0x10 ⁷ -3.0x10 ⁷)	<10	<10	4.19	1.26

All values in parentheses indicate range of counts from three samples.

Table 4.11 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 7°C. The quarg was produced on a laboratory scale using 1% starter culture (Flora Danica) and 1550 units rennet per 1000 kg of milk.

Storage time (Weeks)	PBC/mL	Presumptive Coliform/mL	Yeasts and molds/mL	pH	Lactic acid (%)
0	3.0×10^7 (3.0×10^7 - 3.0×10^7)	<10	<10	4.25	1.17
1	2.7×10^7 (2.3×10^7 - 3.1×10^7)	<10	<10	4.20	1.22
2	3.0×10^7 (3.0×10^7 - 3.0×10^7)	<10	<10	4.15	1.23
3	3.0×10^7 (3.0×10^7 - 3.0×10^7)	<10	<10	4.18	1.24
4	3.0×10^7 (3.0×10^7 - 3.0×10^7)	<10	<10	4.18	1.23

All values in parentheses indicate range of counts from three samples.

Table 4.12 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 7°C. The quarg was produced on a laboratory scale using 1% starter culture (Flora Danica) and 3876 units rennet per 1000 kg of milk.

Storage time (Weeks)	PBC/mL	Presumptive Coliform/mL	Yeasts and molds/mL	pH	Lactic acid (%)
0	3×10^7	<10	<10	4.44	1.16
1	(3.0×10^7 - 3.0×10^7) 3.1×10^7	<10	<10	4.27	1.06
2	(1.0×10^7 - 5.2×10^7) 5.6×10^7	<10	<10	4.29	1.08
3	(2.2×10^7 - 1.1×10^8) 4.7×10^7	<10	3.5×10^7	4.24	1.11
4	(2.9×10^7 - 9.2×10^7) 5.1×10^7	<10	(1.0×10^7 - 7.0×10^7) <10	4.24	1.13
	(1.2×10^7 - 9.0×10^7)				

All values in parentheses indicate range of counts from three samples.

Table 4.13 Effect of five different rennet levels on bitterness development in quarg during storage. Starter culture used was 1% Flora Danica.

Rennet Level (units/1000 kg milk)	Average Sensory Score on Bitterness in Quarg at					Mean Score
	0 week	1 week	2 weeks	3 weeks	4 weeks	
0.00	1.00 ± 0.00	1.00 ± 0.00	1.08 ± 0.29	1.08 ± 0.29	1.25 ± 0.45	1.08
4.41	1.08 ± 0.29	1.17 ± 0.39	1.17 ± 0.39	1.17 ± 0.39	1.67 ± 0.65	1.25
8.81	1.17 ± 0.39	1.25 ± 0.45	1.50 ± 0.67	1.33 ± 0.49	2.00 ± 0.60	1.45
17.62	1.00 ± 0.00	1.67 ± 0.65	1.67 ± 0.65	2.17 ± 0.83	3.17 ± 0.93	1.93
44.05	1.25 ± 0.45	2.42 ± 1.24	2.83 ± 0.83	4.50 ± 0.90	4.92 ± 0.79	3.18

Above figures are average score ± standard deviation of duplicate sample graded by six panelist on a bitterness scale of 1 to 6 (1=not bitter, 6=extremely bitter).

Table 4.14 Analysis of variance on sensory data of quarg produced using five rennet levels, 1% starter culture (Flora panica) and stored at 7°C for 4 weeks.

Source of Variation	Degrees of Freedom	Total Sum of Squares	Mean Sum of Squares	F Value
Rennet level (RL)	4	172.08	43.02	39.83**
Replicates within rennet level (REP)	5	5.4	1.08	
Storage time (ST)	4	78.18	19.55	26.07**
RL by ST	16	71.55	4.47	5.98**
REP by REP	20	14.93	0.75	
Judges (J)	5	4.84	0.97	3.34**
J by RL	20	9.56	0.48	1.66*
J by REP	25	7.20	0.29	
J by ST	20	7.26	0.36	1.16
RL by RL by ST	80	19.01	0.24	0.77
Error	100	31.47	0.31	
Total	299	421.48		

* Significant at 5% level
 ** Significant at 10% level

Table 4.15 Comparison of means using Tukey's test for five rennet levels used in the production of quarg with 1% starter culture (Flora Danica) and stored for 0-4 weeks at 7°C

Sequence of Means				
1	2	3	4	5
1.08	1.25	1.45	1.93	3.18

Table of Difference					
	1	2	3	4	5
5	2.10	1.93	1.73	1.25	0.00
4	0.85	0.68	0.48	0.00	
3	0.37	0.20	0.00		
2	0.17	0.00			
1	0.00				

Table of Test Values					
	1	2	3	4	5
5	0.2861	0.2861	0.2861	0.2861	
4	0.2861	0.2861	0.2861		
3	0.2861	0.2861			
2	0.2861				
1					

1 2 3 4 5

that produced with 775 units, 1550 units and 3876 units per 1000 kg of milk.

These findings may be explained in the light of what is currently known of the role of rennet in bitterness development in hard cheese. Lawrence *et al.* (1972) have shown that the bitterness in cheddar cheese can be lowered by reducing the amount of rennet used per liter of milk. Stadhouders and Hup (1975) and Stadhouders *et al.* (1977) have correlated bitterness development with the amount of rennet retained in the cheddar cheese after cheese making. This amount was governed by the manner in which the curd was washed and drained, the pH of milk and curd during cheese making, amount and type of rennet and the cooking temperature. The lower the pH and cooking temperature, the higher was the amount of the rennet retained. They also reported that cheddar cheese having lower rennet content did not develop bitterness during ripening. Lowrie *et al.* (1972b) however, contradicted this, and stated that rennet would not contribute directly to the formation of bitter peptides but would provide non-bitter peptides of high molecular weights. Stadhouders (1975) ascribed a more active role to rennet for development of bitterness in Gouda cheese.

Rennet is the dominating agent for the production of peptides of high, medium and low molecular weights. Various authors have shown rennin to be capable of liberating bitter peptides from casein components in model systems (Pelissier

and Manchon, 1976; Visser *et al.*, 1975). Visser *et al.* (1983) demonstrated from model studies that rennet generates bitter peptides by hydrolysing α_1 -, β -, and para- κ -casein. Visser and de Groat-Mostert (1977) have reported that rennet in a normal concentration, without the presence of starter bacteria, was able to accomplish the same degradation of α_1 -casein and nearly the same of β -casein as found in normal aseptic cheeses. From these findings, it is evident that rennet could be responsible for the development of bitter flavour in quarg, and the results in Tables 4.13 to 4.15 show that the bitterness development during storage was prevented by using lower levels of rennet. This reduction in bitterness could possibly be explained by the low level of high molecular weight peptides produced by rennet. According to Lawrence *et al.* (1978), high molecular weight peptides are degraded by the culture bacteria to bitter peptides during storage. If the concentration of the precursor (high molecular weight peptides) was low, the culture bacteria could not produce detectable bitterness.

Bitterness in quarg was at a minimum when no rennet was used but the yield was poor when compared to the quarg produced with different amounts of rennet (Table 4.7). This reduced yield could be due to loss of protein and butterfat during draining of whey as a result of weak curd structure. Further, the curd did not possess desirable texture and smoothness. It was, therefore, decided to use 388 units rennet per 1000 kg of milk in subsequent studies. Although

yield was not as high as when higher levels of rennet were used in quarg making, the bitterness development during storage was minimized with 388 units of rennet per 1000 kg of milk.

4.3 Effect of Starter Culture on Bitterness in Quarg

Starter bacteria are known to have a role in bitterness development in cheese. Several authors (Gordon and Speck, 1965; Harwalkar and Seitz, 1971; Stadhouders, 1974) found certain strains of *Streptococcus cremoris* to produce considerable amounts of bitter peptides in milk without the addition of rennet. Stadhouders (1974) reported that in model experiments, bitter peptides were liberated from β -casein by "bitter" starter, in contrast to "non-bitter" strains. According to Lowrie and Lawrence (1972), the occurrence of bitterness in cheese depends upon the ability of starter bacteria to multiply to high cell densities under the manufacturing conditions used.

Thus a study was designed to find whether the type of starter culture used could have a role in the development of bitterness in quarg. In our study four batches of quarg in duplicate were produced with four different types of starter culture, keeping the rennet level at 388 units per 1000 kg of milk. The types of starter culture used and the composition of the quarg produced are shown in Table 4.16. The quarg samples were stored for four weeks at 7°C to evaluate the effect of starter culture on bitterness

Table 4.18 Composition of quarg produced with 388 units of rennet per 1000 kg of milk and four different type of starter culture at 1% level

Components	starter Culture			
	Flora Danica		Streptococcus cremoris	
	with <i>S. diacetilactis</i>	without <i>S. diacetilactis</i>	(#134)	Single Strain (#584)
Moisture	73.26 ± 0.88	72.01 ± 0.26	74.90 ± 0.38	73.95 ± 0.26
Lactic Acid	1.06 ± 0.02	1.17 ± 0.02	1.03 ± 0.04	1.12 ± 0.01
Protein	7.50 ± 0.21	7.67 ± 0.22	7.70 ± 0.19	7.89 ± 0.19
Butterfat	10.80 ± 0.18	10.15 ± 0.23	10.60 ± 0.41	10.68 ± 0.29
pH	4.29	4.20	4.29	4.15
Yield (%)	21.90	21.20	22.52	21.24

The above figures are average of 4 replicates on 2 Samples

development, changes in the predominant microorganisms, pH and lactic acid content. The results are presented in Tables 4.17, 4.18, 4.19 and 4.20. Psychrotrophic, Coliform, Yeast and Mold counts did not differ significantly for different starters used, and the counts were low and similar to those observed with different levels of rennet. These low counts suggest that the bitterness in quarg was not due to microbial contaminants.

The average bitterness scores of quarg produced with four different starter cultures and stored for a period of four weeks are presented in Table 4.21. Although the analysis of variance performed on the sensory data (Table 4.22) indicates a significant difference among the four starters used, the average bitterness scores for these starters are low even after four weeks of storage. The highly significant difference (Table 4.22) among starters is obviously due to the high relative difference in bitterness scores between Flora Danica (with and without *S. diacetilactis*) and single strain cultures (#134 and #584) of *S. cremoris*. A comparison of means for the four weeks of storage for different starter cultures was carried out using Tukey's test. Table 4.23 shows that Flora Danica with and without *Streptococcus diacetilactis* showed similar pattern in bitterness score as was observed in quarg made with 1% Flora Danica and 388 units rennet per 1000 kg of milk. But the bitterness scores were lower in quarg made with single strain (#134 and #584) cultures of *Streptococcus cremoris*.

Table 4.17 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 7°C. The quarg was produced on a laboratory scale using 1% starter culture (Flora Danica with *S. diacetilactis*) and 388 units rennet per 1000 kg of milk.

Storage time (weeks)	PBC/mL	Presumptive Coliform/mL	Yeasts and molds/mL	pH	Lactic acid (%)
0	2.4x10 ⁷ (3.0x10 ⁷ -4.5x10 ⁷)	<10	<10	4.29	1.06
1	2.3x10 ⁷ (2.0x10 ⁷ -3.5x10 ⁷)	<10	<10	4.23	1.09
2	5.4x10 ⁷ (1.8x10 ⁷ -8.9x10 ⁷)	<10	<10	4.27	1.13
3	3.7x10 ⁷ (3.3x10 ⁷ -4.0x10 ⁷)	<10	<10	4.29	1.15
4	3.9x10 ⁷ (3.0x10 ⁷ -4.7x10 ⁷)	<10	<10	4.26	1.17

All values in parentheses indicate range of counts from three samples.

Table 4.18 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 7°C. The quarg was produced on a laboratory scale using 1% starter culture (Flora Danica without *S. diacetilactis*) and 388 units rennet per 1000 kg of milk.

Storage time (Weeks)	PBC/mL	Presumptive Coliform/mL	Yeasts and molds/mL	pH	Lactic acid (%)
0	6.2x10 ⁷ (3.0x10 ⁷ -1.2x10 ⁸)	<10	2.5x10 ⁷ (2.0x10 ⁷ -3.0x10 ⁷)	4.20	1.17
1	1.6x10 ⁷ (1.2x10 ⁷ -2.0x10 ⁷)	<10	<10	4.13	1.29
2	4.0x10 ⁷ (2.2x10 ⁷ -5.8x10 ⁷)	<10	5.5x10 ⁷	4.15	1.27
3	5.5x10 ⁷ (2.0x10 ⁷ -9.0x10 ⁷)	<10	<10	4.22	1.22
4	6.2x10 ⁷ (4.4x10 ⁷ -8.0x10 ⁷)	<10	<10	4.12	1.27
8	3.2x10 ⁷ (3.0x10 ⁷ -1.5x10 ⁸)	<10	1.1x10 ⁷ (1.0x10 ⁷ -4.8x10 ⁷)	4.27	1.58

All values in parentheses indicate range of counts from three samples.

Table 4.19 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 7°C. The quarg was produced on a laboratory scale using 1% starter culture (*S. cremoris* Single Strain # 134) and 388 units rennet per 1000 kg of milk.

Storage time (Weeks)	PBC/mL	Presumptive Coliform/mL	Yeasts and molds/mL	pH	Lactic acid (%)
0	2.6x10 ³ (3.0x10 ¹ -4.8x10 ³)	<10	<10	4.19	1.03
1	2.6x10 ³ (2.0x10 ³ -3.2x10 ³)	<10	8.5x10 ¹ (1.0x10 ¹ -1.6x10 ²)	4.17	1.00
2	4.0x10 ³ (7.6x10 ¹ -8.0x10 ³)	<10	2.0x10 ¹ (1.0x10 ¹ -3.0x10 ¹)	4.15	1.13
3	1.7x10 ³ (2.7x10 ¹ -3.2x10 ³)	<10	1.7x10 ¹ (3.6x10 ¹ -3.0x10 ¹)	4.20	1.08
4	3.6x10 ³ (3.8x10 ³ -6.9x10 ³)	<10	1.6x10 ¹ (1.5x10 ¹ -3.2x10 ¹)	4.13	1.31

All values in parentheses indicate range of counts from three samples.

Table 4.20 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 7°C. The quarg was produced on a laboratory scale using 1% starter culture (*S. cremoris* Single Strain # 584) and 388 units rennet per 1000 kg of milk.

Storage time (Weeks)	PBC/mL	Presumptive Coliform/mL	Yeasts and molds/mL	pH	Lactic acid (%)
0	6.2x10 ⁷ (5.0x10 ⁷ -7.3x10 ⁷)	<10	<10	4.15	1.12
1	3.3x10 ⁷ (3.0x10 ⁷ -3.5x10 ⁷)	<10	<10	4.11	
2	2.3x10 ⁷ (1.3x10 ⁷ -3.2x10 ⁷)	<10	<10	4.12	1.16
3	1.9x10 ⁷ (1.3x10 ⁷ -3.8x10 ⁷)	<10	<10	4.19	1.13
4	1.8x10 ⁷ (3.7x10 ⁷ -3.6x10 ⁸)	<10	<10	4.23	1.12

All values in parentheses indicate range of counts from three samples.

Table 4.21 Effect of four different starter cultures on bitterness development in quarg during storage. Rennet level used was 388 units per 1000 kg of milk.

Starter Culture	Average sensory score on bitterness in quarg at					Mean Score
	0 week	1 week	2 weeks	3 weeks	4 weeks	
Flora Danica	1.08 ± 0.29	1.17 ± 0.39	1.17 ± 0.39	1.33 ± 0.49	1.00 ± 0.60	1.35
Flora Danica without <i>S. diacetylactis</i>	1.25 ± 0.45	1.33 ± 0.49	1.00 ± 0.00	1.67 ± 0.65	1.92 ± 0.67	1.43
<i>S. cremoris</i> Single Strain #134	1.00 ± 0.00	1.17 ± 0.39	1.00 ± 0.00	1.00 ± 0.00	1.08 ± 0.29	1.05
<i>S. cremoris</i> Single Strain #584	1.00 ± 0.00	1.08 ± 0.29	1.08 ± 0.29	1.08 ± 0.29	1.08 ± 0.29	1.07

Above figures are average score ± standard deviation of duplicate samples judged by six panallist on a bitterness scale of 1 to 6 (1=not bitter, 6=extremely bitter).

Table 4.22 Analysis of variance on sensory data of quarg produced using four different starter cultures, 388 units rennet per 1000 kg of milk and stored at 7°C for 4 weeks.

Source of Variation	Degrees of Freedom	Total Sum of Squares	Mean Sum of Squares	F Value
Starter culture(SC)	3	6.88	2.29	38.17**
Replicates within starter Culture(REP)	4	0.23	0.06	
Storage Time (ST)	4	6.60	1.65	8.68**
SC by ST	12	6.70	0.56	2.95*
ST by REP	16	3.10	0.19	
Judges (J)	5	2.80	0.56	4.67**
J by SC	15	4.37	0.29	2.42*
J by REP	20	2.37	0.12	
J by ST	20	1.45	0.07	0.50
J by SC by ST	60	6.05	0.10	0.71
Error	80	11.30	0.14	
Total		51.85		

Table 4.23 Comparison of means using Tukey's test for four starter cultures used in the production of quarg with 388 units rennet per 1000 kg of milk.

Sequence of Means

3	4	1	2
1.05	1.07	1.35	1.43

Table of Difference

	3	4	1	2
2	0.38	0.36	0.08	0.00
1	0.30	0.28	0.00	
4	0.02	0.00		
3	0.00			

Table of Test Values

	3	4	1	2
2	0.1807	0.1807	0.1807	
1	0.1807	0.1807		
4	0.1807			

3 4 1 2

The low bitterness scores for single strain cultures (Table 4.21) indicate that these cultures were selected from "non-bitter" strains of *Streptococcus cremoris*. Martley and Lawrence (1972) observed that the "non-bitter" strains exhibited lower proteolytic activity. Lowrie *et al.* (1972b) reported that the "non-bitter" strains do not reach high populations characteristic of "bitter" *Streptococci*. According to Lawrence and Gilles (1969), "slow" starter never gives bitter cheese, even when added in high amounts to the cheese vat so as to give a high rate of acid production. These observations suggest that the degradation of high molecular peptides is limited when "non-bitter" strains are used and low molecular bitter peptides may not then be produced to an extent to cause detectable bitterness. Thus, the concentration of bitter peptides might not have exceeded the threshold at which bitterness could be detected in quarg made with single strain cultures (#134 and #584) of *Streptococcus cremoris*. Although the nature of starter culture may play an important role in production of bitter peptides from the non-bitter ones, the level of rennet appears to be the most critical factor in development of bitterness in quarg during storage.

4.4 Commercial Production of Quarg

The laboratory study indicated that the quality defect due to bitterness can be minimized by using a rennet level of 388 units per 1000 kg of milk with 1% starter culture

(Flora Danica). Quarg samples were produced commercially in a local cheese plant following the procedure shown in Figure 3.2 to evaluate the results of the laboratory study. Table 4.24 shows the chemical composition of the quarg produced. The results of the storage study of commercial quarg stored at 7°C for four weeks are presented in Table 4.25. The psychrotroph and coliform counts were low, as was observed in the laboratory study. However, yeast and mold count was higher. This may have been due to improper sanitation resulting in contamination during commercial production. The quarg samples were evaluated every week by the same panel of six trained judges and their scores are presented in Table 4.26, which shows that bitterness could not be detected after 4 weeks of storage. Dr. E. Renner, a Visiting Professor of Dairy Technology from Justus-Liebig University, West Germany tasted the product after 10 weeks of storage at 5°C and could not detect bitterness. He compared the samples examined to a typical commercial quarg available in West Germany. The analysis of variance carried out on the sensory data (Table 4.27) did not show significant difference in bitterness scores for quarg samples stored for a period of four weeks. It appears that good quality commercial quarg can be produced by using a rennet level of 388 units per 1000 kg of milk with 1% starter culture (Flora Danica).

Table 4.24 Composition of quarg produced commercially with 388 units rennet per 1000 kg milk and 1% starter culture (Flora Danica)

Components	Amount (%)
Moisture	72.20 ± 0.32
Lactic Acid	1.36 ± 0.02
Protein	9.88 ± 0.12
Butterfat	14.35 ± 0.22

pH	4.43
Yield (%)	22.50

Above figures are average ± standard deviation of 6 replicates performed on 3 samples

Table 4.25 Changes in the predominant types of microorganisms, pH and lactic acid in quarg during storage at 7°C. The quarg was produced commercially using 1% starter culture (Flora Danica) and 388 units rennet per 1000 kg of milk.

Storage Time (Weeks)	PBC/mL	Presumptive Coliform/mL	Yeasts and Molds/mL	pH	Lactic Acid %
0	1.7x10 ⁷ (3.0x10 ⁷ -3.0x10 ⁷)	<10	1.7x10 ⁷ (1.0x10 ⁷ -3.3x10 ⁷)	4.50	1.36
1	1.7x10 ⁷ (3.0x10 ⁷ -3.0x10 ⁷)	<10	1.2x10 ⁷ (1.2x10 ⁷ -1.2x10 ⁷)	4.53	1.37
2	3.0x10 ⁷ (3.0x10 ⁷ -3.0x10 ⁷)	<10	1.2x10 ⁷ (1.0x10 ⁷ -2.3x10 ⁷)	4.48	1.36
3	3.0x10 ⁷ (3.0x10 ⁷ -3.0x10 ⁷)	<10	1.5x10 ⁷ (1.0x10 ⁷ -3.0x10 ⁷)	4.43	1.35
4	3.0x10 ⁷ (3.0x10 ⁷ -3.0x10 ⁷)	<10	5.0x10 ⁷ (1.0x10 ⁷ -8.0x10 ⁷)	4.50	1.37

All values in parentheses indicate range of counts from three samples.

Table 4.26 Sensory evaluation data of commercial quarg produced with 388 units rennet per 1000 kg of milk and 1% starter culture (Flora Danica) and stored at 7°C.

Judges	Storage Time (weeks)				
	0	1	2	3	4
1	1.0	2.0	2.0	1.0	2.0
	1.0	2.0	1.0	1.0	1.0
2	1.0	1.0	1.0	2.0	1.0
	1.0	1.0	1.0	1.0	1.0
3	1.0	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	2.0	1.0
4	1.0	1.0	2.0	1.0	2.0
	1.0	1.0	2.0	2.0	1.0
5	1.0	1.0	1.0	2.0	1.0
	1.0	2.0	1.0	1.0	1.0
6	1.0	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0	1.0
Mean	1.00	1.08	1.17	1.00	1.25

Table 4.27 Analysis of Variance on Sensory Data of Ouarg Produced Commercially Using 388 units rennet per 1000 kg of milk, 1% starter Culture (Flora Danica) and Stored at 7°C for 4 Weeks.

Source of Variation	Degrees of Freedom	Total Sum of Squares	Mean Sum of Squares	F Value
Storage Time (ST)	4	0.77	0.19	1.90
Replicates within Storage Time (REP)	5	0.50	0.10	
Judges (J)	5	1.40	0.28	2.00
ST by J	20	3.43	0.17	1.21
Error	25	3.50	0.14	
Total		9.60		

4.5 Yield of Quarg

The yield, an important determinant of effectiveness of a commercial process, is defined as the amount of quarg produced from a given amount of milk and is directly related to moisture content of the finished product. Yield is affected by the insoluble fractions of milk, viz. fat, casein and some minerals, whereas the soluble components such as whey proteins, lactose and certain minerals do not affect yield considerably, since most of these components are lost in the whey during draining. Our laboratory study on the production of quarg with five different levels of rennet indicated that the yield of quarg improved with increased levels of rennet (Table 4.7). This might be due to a higher degree of recovery of fat and casein by the curd during quarg making. Thus, it is evident that the higher levels of rennet have a positive influence on the yield of quarg. Although the yield was improved with a higher level of rennet, it resulted in development of bitterness during storage, limiting the shelf-life of the products to two weeks at refrigerated temperature. It was, therefore, decided to use a rennet level of 388 units per 1000 kg of milk, which gave yield of 21.9% in the laboratory study. Further, it was observed that different starter cultures used in the laboratory study have no influence on the yield of quarg (Table 4.16). Quarg was produced commercially using 388 units of rennet per 1000 kg of milk and 1% of starter culture (Flora Danica) and the yield was observed to be

22.5%. It is apparent that good quality quarg can be produced with an extended shelf-life without jeopardising the yield as indicated in initial trials. The value of 22.5% is considered to be a good compromise between yield and good product quality with an extended shelf-life.

4.6 Consumer Acceptance Study

Consumption of quarg in Alberta is very low compared to many European countries. It was therefore considered useful to assess the consumer reaction to plain and fruit quarg and to project the future marketing potential of this dairy product which is relatively new in Alberta and in Canada. Accordingly, a consumer acceptance study was conducted in the City of Edmonton.

4.6.1 Survey Response

Plain and fruit (tropical fruit pulp used in yogurt) quarg samples were distributed to 596 households along with a questionnaire (Appendix 3). Of these, 223 households responded by mailing back the forms. This constitutes 37.4% return, considered satisfactory in most mail-in surveys (Emory, 1976).

4.6.2 Opinion on Plain and Fruit Quarg

The opinions of respondents on appearance, flavour and overall quality of plain and fruit quarg are presented in Figures 4.1, 4.2 and 4.3. These figures indicate a unimodal,

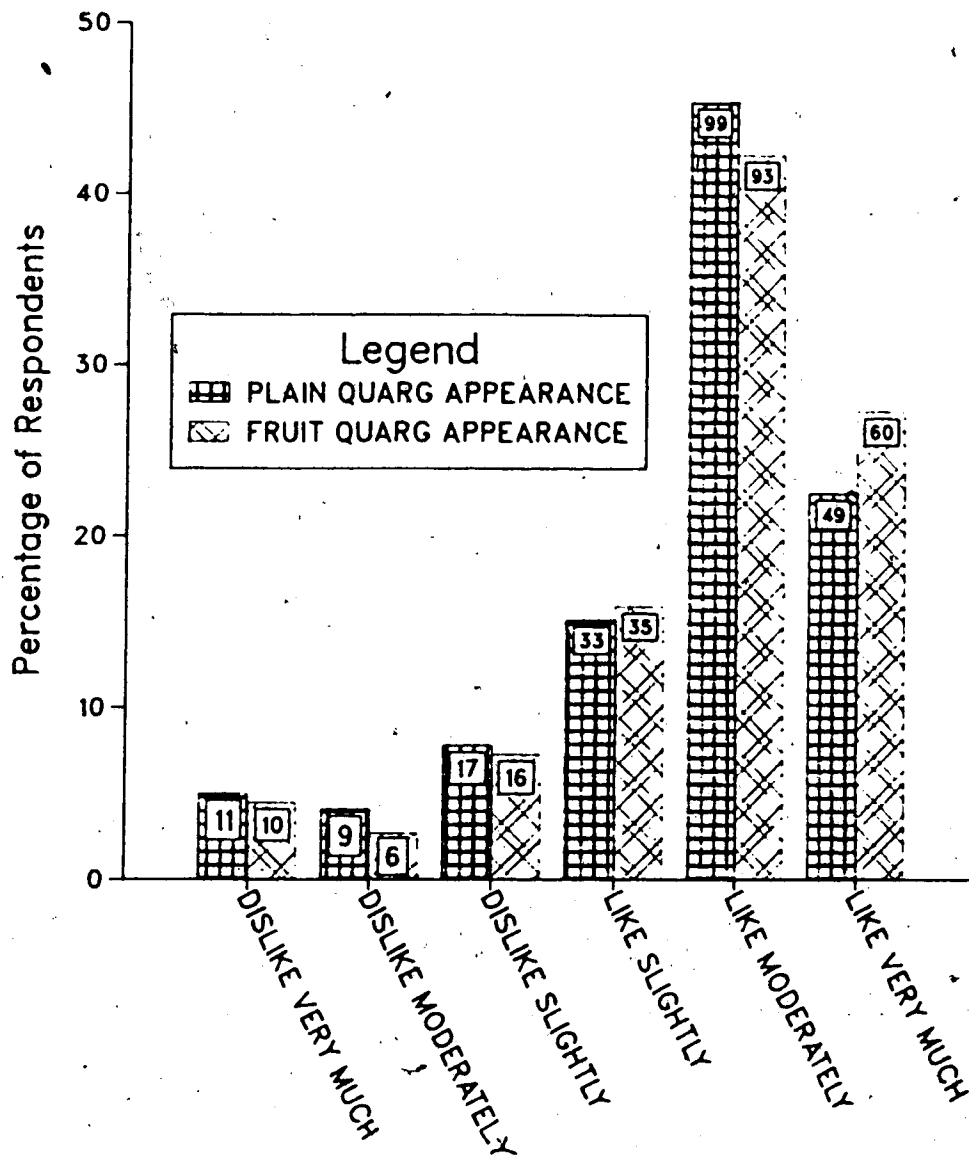


Figure 4.1 Respondents' opinion on appearance of plain and fruit quarg. The numbers at the top of bars indicate totals of respondents in each category.

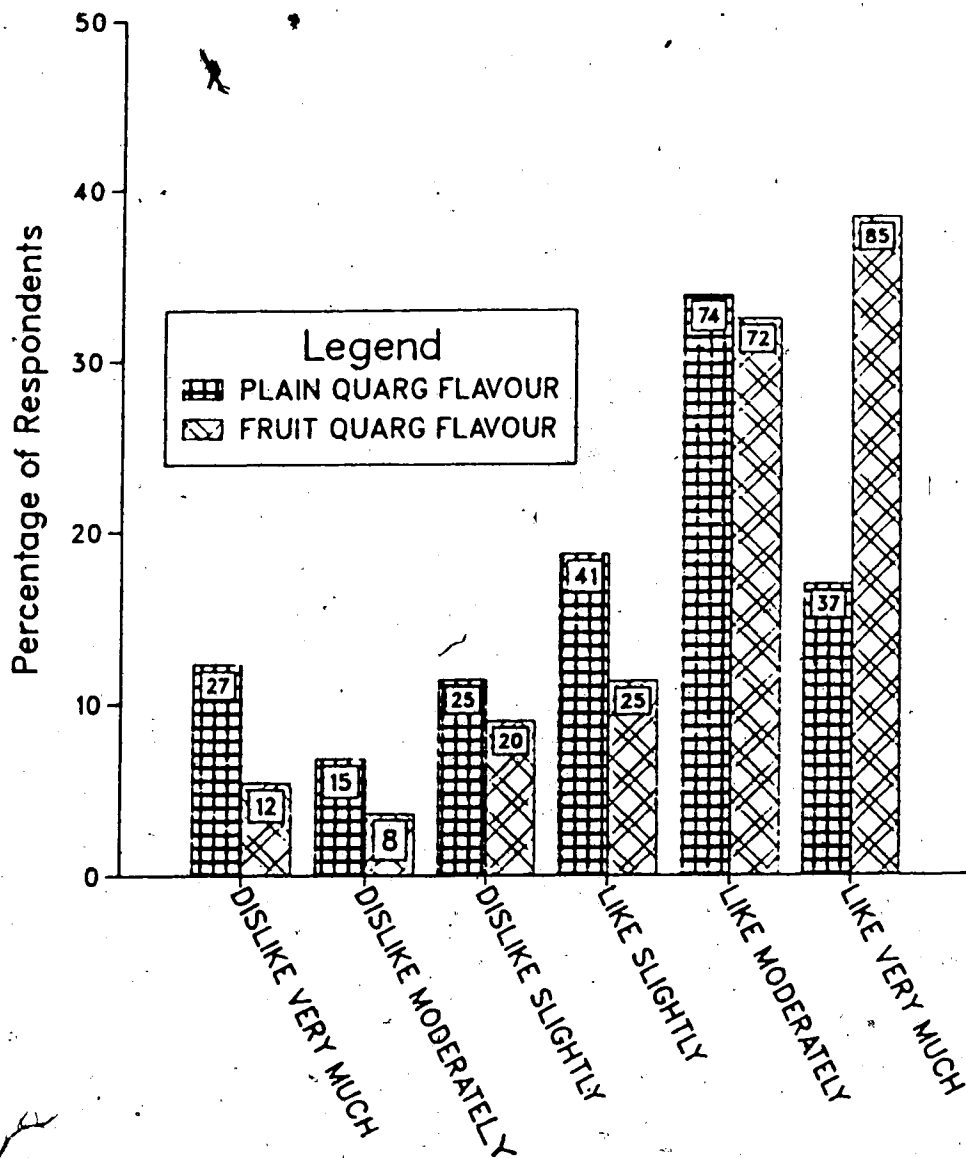


Figure 4.2 Respondents' opinion on flavour of plain and fruit quarg. The numbers at the top of bars indicate totals of respondents in each category.

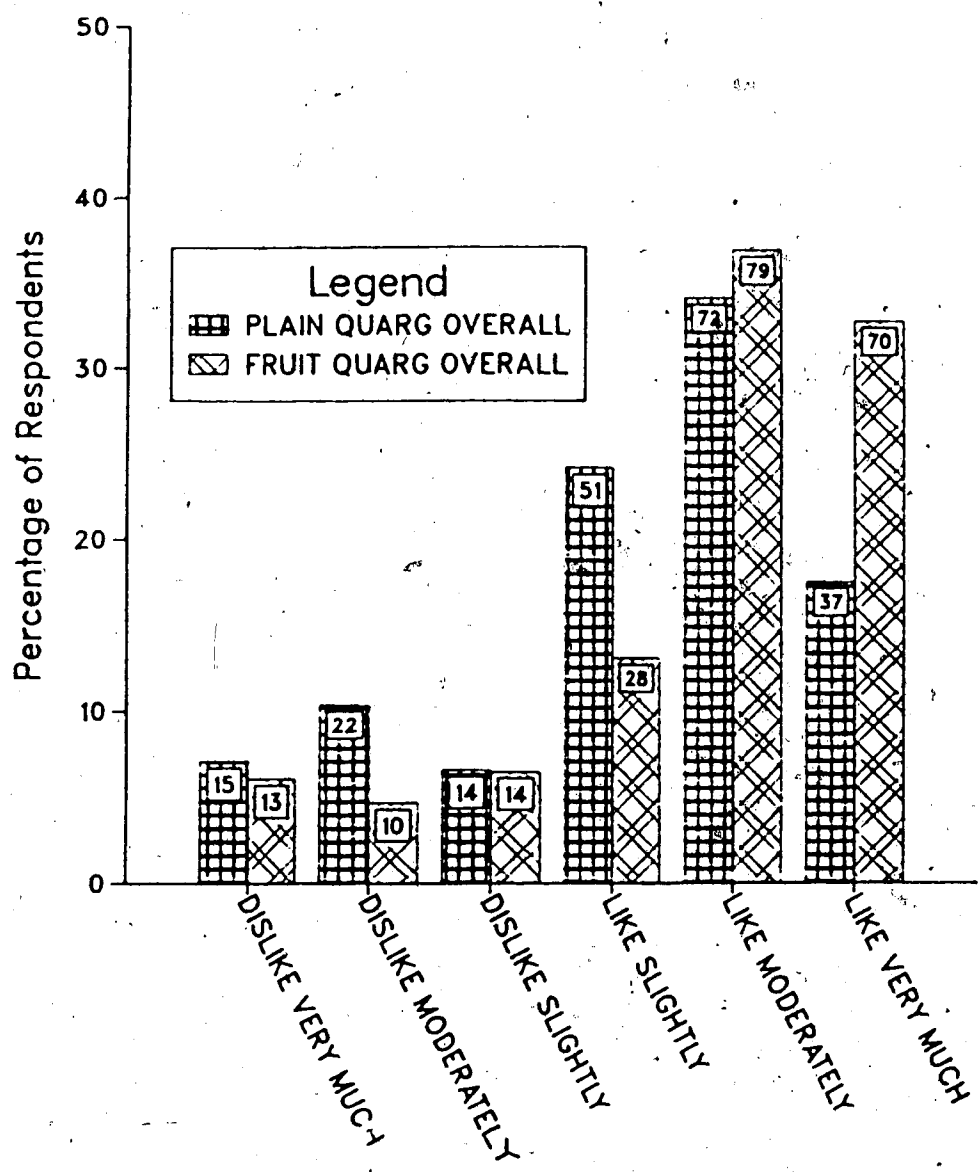


Figure 4.3 Respondents' opinion on overall quality of plain and fruit quarg. The numbers at the top of bars indicate totals of respondents in each category.

negatively skewed distribution of respondents' opinion, with a peak at "like moderately" for appearance and overall quality of both plain and fruit quarg. The peaks for flavour are at "like moderately" for plain quarg and at "like very much" for fruit quarg which reflects consumers preference for the flavour of fruit quarg over that of plain quarg. It is also evident (Figures 4.1, 4.2 and 4.3) that the percentage of respondents expressing favourably ("like moderately" and "like very much") for plain quarg is more than 50% (67.9% for appearance, 50.7 for flavour and 51.6% for overall quality) and about 70% for fruit quarg (69.6% for appearance, 70.7% for flavour and 69.6% for overall quality). The statistical analysis of the consumer acceptance study data is presented in Table 4.28 which clearly indicates consumers' preferential choice for fruit quarg over that of plain quarg. The t-values between plain and fruit quarg for all the quality attributes (appearance, flavour and overall) were significant at 5% level and the mean score for fruit quarg was always higher than that for plain quarg. Thus, it can be concluded that the respondents showed a significant preference for fruit quarg over the unflavoured product.

4.6.3 Market Potential of Plain and Fruit Quarg

The overall result of the survey was very encouraging with respect to the market potential of this relatively new dairy product and indicates that both plain and fruit quarg

Table 4.28 Statistical analysis of consumer acceptance survey data.

Quality Parameter	Quang	Mean	Standard Deviation	Standard Error	T-Value	Degree of Freedom
Appearance	Plain	4.5899	1.310	0.089		
	Fruit	4.6912	1.274	0.086	-1.90*	216
Flavour	Plain	4.0548	1.587	0.107		
	Fruit	4.7489	1.416	0.096	-6.89**	218
Overall	Plain	4.2038	1.461	0.101		
	Fruit	4.6635	1.423	0.098	-5.13**	210

* Significant at 5% level

** Significant at 10% level

have a good potential to become regular part of the diet in North American households. Respondents' previous exposure to the product and their willingness to buy it are shown in Figures 4.4, 4.5 and 4.6.

A majority of the respondents (60.5% for plain quarg and 74.0% for fruit quarg) have expressed their interest in buying the product (often and sometimes), even though only 23.2% of them had tasted the product previously. Figure 4.6 indicates a unimodal distribution of respondents' willingness to buy the product with a single peak at "sometimes". This reflects a high degree of acceptability of the product, even though the previous exposure of the respondents to the product was very low, as shown in Figures 4.5 and 4.6.

The consumption pattern of the quarg is presented in Table 4.29 which reveals that 29.9% of the respondents ate the product at lunch and 24.9% at other times. It appears that more than 50% of the respondents preferred to eat quarg as a snackfood or part of lunch.

4.6.4 General Comments from Respondents

In addition to the acceptance scores, the respondents also gave numerous comments on flavour, texture, overall quality and mode of use of plain and fruit quarg.

1. Flavour: Majority of the respondents liked the flavour of fruit quarg. Some of them have also given favourable comments on plain quarg flavour. A few have commented

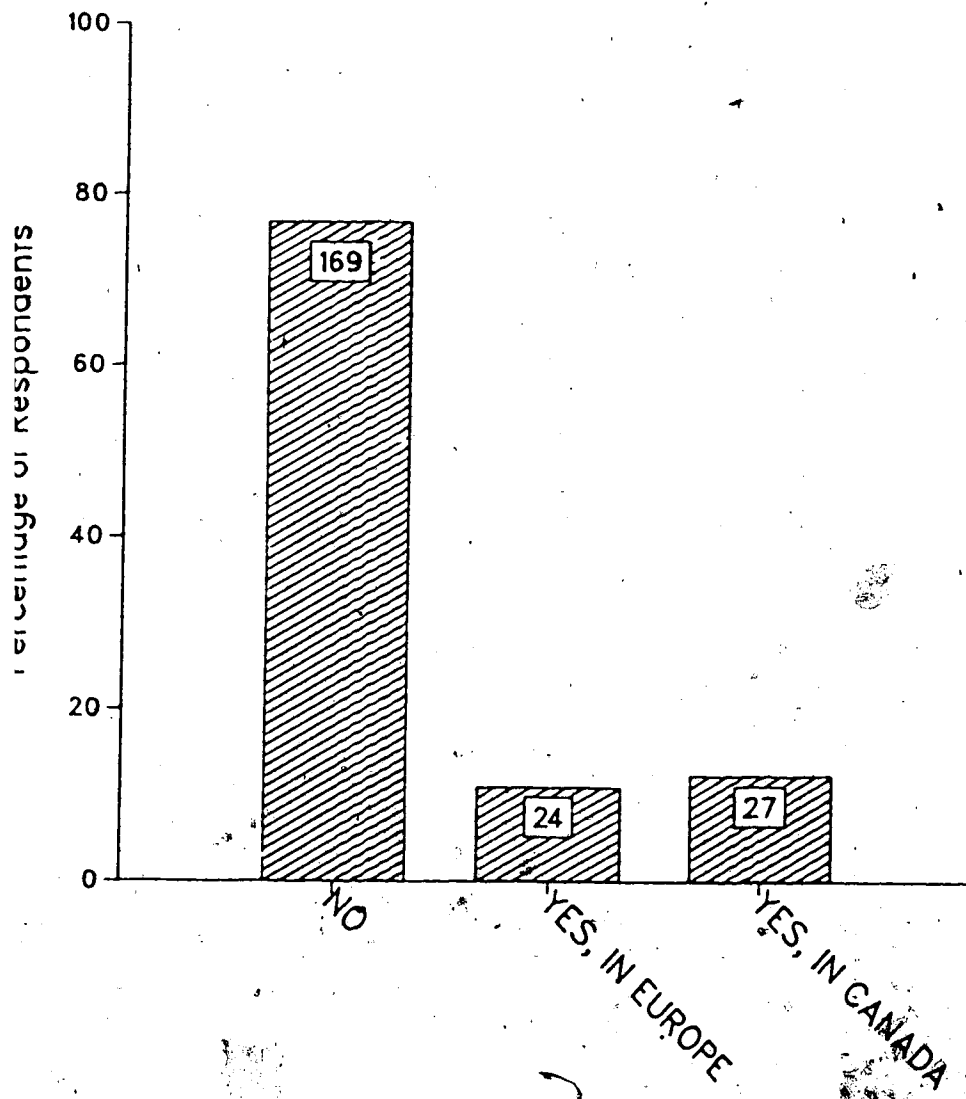


Figure 4.4 Respondents' previous exposure to quartz. The numbers at the top of bars indicate totals of respondents in each category.

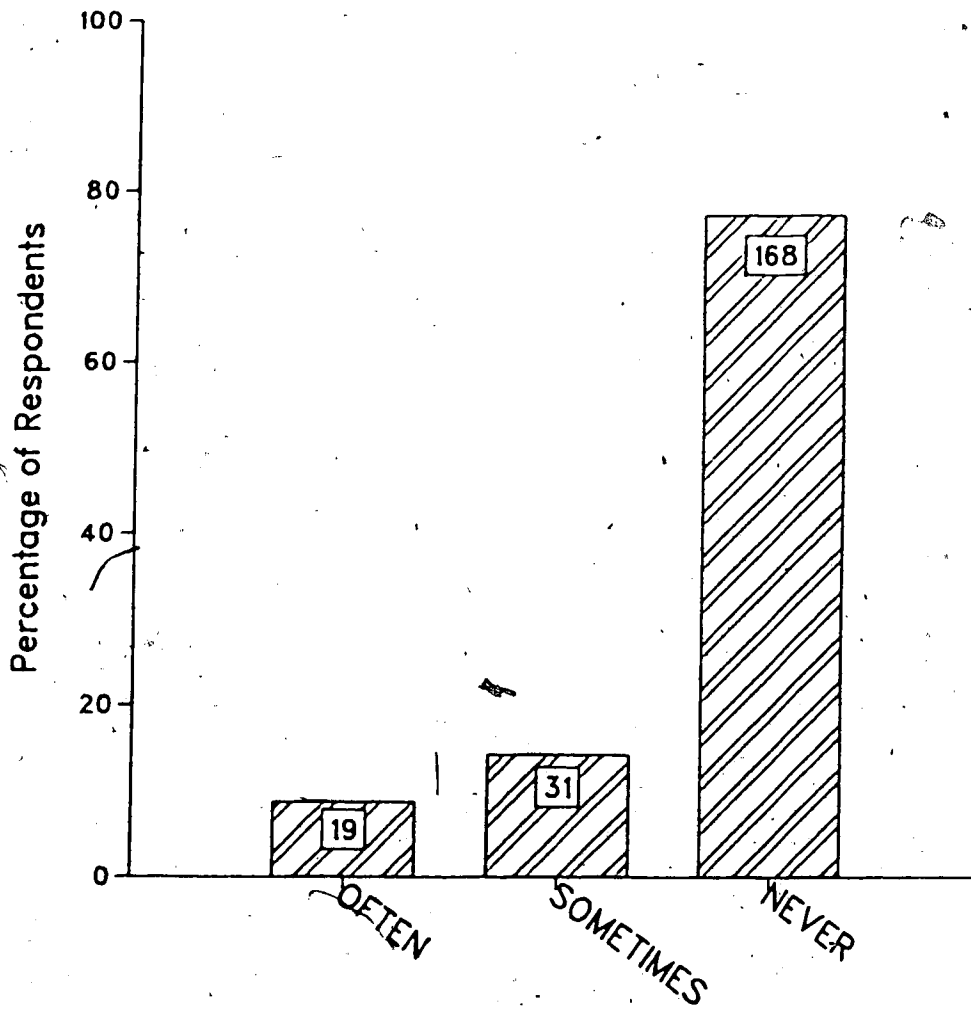


Figure 4.5 Frequency of respondents' previous consumption of quarg. The numbers at the top of bars indicate totals of respondents in each category.

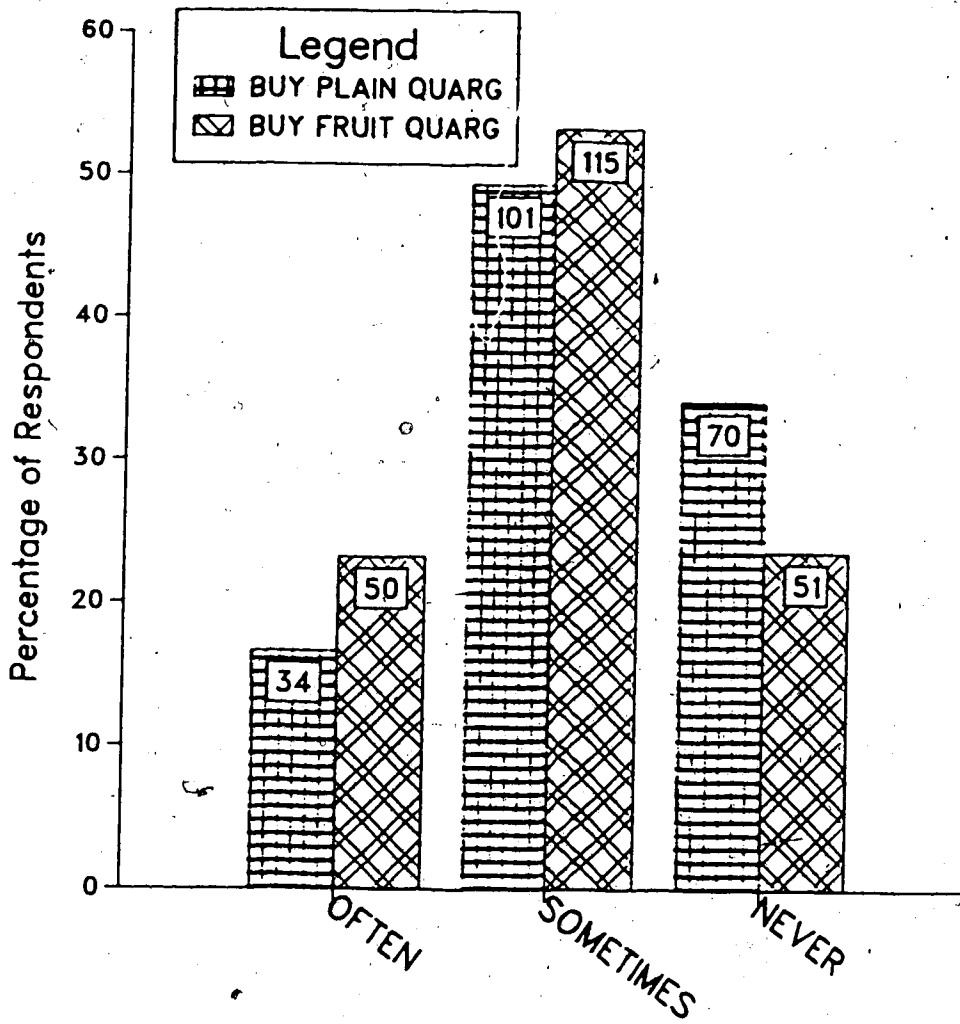


Figure 4.6 Respondents' willingness to buy plain and fruit quarg. The numbers at the top of bars indicate totals of respondents in each category.

Table 4.29 Consumption pattern of quarg by respondents.

Consumption Time	% of Respondents
Breakfast	12.2
Lunch	29.9
Supper	18.1
Other	24.9
More than one meal	14.9

that the plain quarg was slightly acidic in taste, whereas the fruit quarg appeared to have artificial flavour. They also indicated that it will take some time to get used to quarg flavour. One of the attractive characteristics of quarg appreciated by respondents was the "absence of salt". No respondents found quarg to be bitter tasting.

2. Texture: In general, no texture defect was pointed out. Most of the respondents liked the smooth texture of quarg.
3. Overall Quality: Most of the respondents liked the overall quality of both plain and fruit quarg. A few of the respondents described quarg as "thick, yogurt" and did not like the overall appearance.
4. Mode of Use: Most people consumed quarg with crackers, on toast, in sandwiches, in salads and on buns. Some also ate quarg mixed with seasonings. Some respondents also used quarg as a substitute for sour cream on baked potatoes, while others used it in cheese cake and also as a dessert.

It is evident from the results of the survey that the potential for fruit quarg being accepted as a new dairy product is greater than plain quarg. Majority of the respondents have indicated a high degree of acceptability of both plain and fruit quarg, even though most of them were not exposed to the product previously. Respondents' comment on quarg being a "no-salt" product can be used as a marketing strategy to increase the sale of this soft,

unripened cheese.

5. CONCLUSIONS AND RECOMMENDATIONS

Quarg is a relatively new dairy product on the Canadian market. Its consumption in Alberta is very low compared to many European countries. Bitterness in the commercially available quarg was a flavour defect that limited the shelf-life of this product to two weeks and consequently the product could not be marketed outside Alberta. This study was aimed at investigating the possible remedies for the bitterness development in quarg during storage and to ascertain the market potential of this dairy product in Alberta.

5.1 Summary of Results

Our investigation with five different levels of rennet indicated that the quantity of rennet used has a strong influence on the development of bitterness during storage. The intensity of the bitterness development increased when a higher level of rennet was used. It was found that bitterness development could be prevented by decreasing the rennet level from the amount used by industry, i.e., from 3876 units to 388 units per 1000 kg of milk. The quarg thus produced could be stored for more than 4 weeks at refrigeration temperature without development of detectable bitterness. However, to prove this statement conclusively, further study would be desirable on the retention of rennet in quarg. The counts on psychrotrophs, yeasts and molds were low (Tables 4.8-4.12) even after 4 weeks of storage. These

low levels of predominating microorganisms indicate that their involvement in bitterness development is unlikely, since non-bitter cheeses were produced with counts higher than those observed in this study.

The study on the effect of starter culture on bitterness development did not indicate a major role of starter organisms in bitterness development in quarg. The average bitterness scores (Table 4.21) on quarg produced with four different starters indicate that these cultures did not produce a high degree of bitterness after 4 weeks of storage. In the case of single strain cultures of *Streptococcus cremoris* the bitterness could not be detected even after 4 weeks of storage. This study with starter cultures confirmed that the amount of rennet used in quarg making plays a dominant role in the development of bitterness in quarg. The counts on predominating organisms such as psychrotrophs, yeasts and molds were low and similar to those observed with different levels of rennet over a period of 4 weeks.

The results of the laboratory study were translated to production of quarg in a commercial environment. Quarg was produced with 388 units rennet per 1000 kg milk and 1% starter culture (Flora Danica) in a local cheese plant according to the standard procedure (Figure 3.3). Following production, quarg samples were stored for a period of 4 weeks at 7°C. This study again indicated that good quality commercial quarg, with an extended shelf-life of at least 4

weeks, can be produced if the rennet level is decreased from 3876 units to 388 units per 1000 kg of milk. Bitterness could not be detected in this commercial quarg after 4 weeks of storage and the yield was similar to that produced with 3876 units rennet per 1000 kg milk.

The study of the consumer acceptance of quarg was very encouraging. Though quarg is a relatively new dairy product in North America, the respondents have reacted positively to plain and fruit quarg. This study indicated that both plain and fruit quarg could have a good market potential in Alberta. Further study should be carried out to assess the market potential of quarg in other areas of North America. Respondents have also shown significant preference for fruit quarg over that of plain quarg. From this study, it was apparent that more research is needed to develop new varieties of quarg with different fruit and other flavours.

5.2 Future Research Needs

Based on the results obtained in this study, it is reasonable to conclude that the amount of rennet used in quarg making is responsible for development of bitterness in quarg. A natural extension of this work would be to investigate in detail the retention of rennet by quarg after manufacture. Further work is necessary to clarify this question and also to conclusively resolve any possible involvement of microorganisms.

Detailed study is required on isolation and characterization of bitter peptides and the mechanism of their formation during storage.

In terms of future processing research, it would be desirable to use ultrafiltration or high heat treatment of milk to increase the yield in quarg making and to make quarg from skim milk. Further research is also needed in the area of product development. New varieties of quarg with different fruits and other flavours (vegetables, sea food etc.) should be developed. Vigorous effort is also needed to develop a market for these varieties.

The consumer study has indicated good market potential for plain and fruit quarg in Alberta. With further research and development, quarg can become as popular in Alberta and other areas of North America as it is in Europe.

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APPENDIX 1 : TRIANGLE TEST FORM

TRIANGLE TEST

DEFFERENCE ANALYSIS

DATE ----- TASTER -----

PRODUCT -----

Instruction: Here are the samples for evaluation. Two of these samples are duplicates. Separate the odd sample for difference only. Please taste the samples in the order indicated

	(1)	(2)
	Sample	Sample
1.	-----	-----
2.	-----	-----
3.	-----	-----

(3) Indicate the degree of difference between the duplicate samples and the odd sample.

Slight----- Much-----
Moderate----- Extreme-----

(4) Describe the type of difference between odd and duplicate sample -----

APPENDIX 2 : SENSORY EVALUATION OF QUARG

SCORING FORM FOR SENSORY EVALUATION OF QUARG

Name ----- Date -----

Sample No.	Not Bitter	Trace of Bitterness	Slightly Bitter	Very Bitter	Extremely Bitter
------------	------------	---------------------	-----------------	-------------	------------------

1

2

3

4

5

Comments -----

APPENDIX 3. : NEW DAIRY PRODUCT SURVEY

As part of a research project in the Food Science Department, University of Alberta, we are trying to determine consumer likes, dislikes, and overall acceptability, of "Quarg" (unripened, soft cheese produced commercially in Alberta, and very popular in Europe).

Please use the following six-point scale to indicate each house-hold member's individual opinion about the three indicated quality aspects for both plain and fruit quarg:

- | | |
|-----------------------|----------------------|
| A. like very much | B. like moderately |
| C. like slightly | D. dislike slightly |
| E. dislike moderately | F. dislike very much |

Sample	Appearance	Flavour	Overall Acceptability	Number of member
Plain Quarg				
Fruit Quarg				

COMMENTS : _____

Have you ever eaten this type of dairy product before? _____
 If yes, where did you try it? _____
 And how often did you eat it? _____
 Would you buy either of these products, if they were available at a reasonable price?

Plain Quarg	---Often	---Sometimes	---Never
Fruit Quarg	---Often	---Sometimes	---Never

What time of the day did you taste the sample products.

---Breakfast ---Lunch ---Supper ---Other

Please return the questionnaire in the stamped, self-addressed envelope provided no later than March 15. Your cooperation is highly valued and greatly appreciated.

For further information regarding this product or survey, please call Dr. Pavel Jelen, Professor, University of Alberta, 432-2480.

APPENDIX 4 : SENSORY EVALUATION DATA OF QUARG

Table A-4.1 Sensory evaluation data of fresh quarg produced with 1% starter culture (Flora Danica) and different levels of rennet.

Judges	Rennet level (units/1000 kg milk)				
	0	388	775	1550	3876
1	1.0	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0	2.0
2	1.0	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0	2.0
3	1.0	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0	1.0
4	1.0	1.0	1.0	1.0	1.0
	1.0	1.0	2.0	1.0	1.0
5	1.0	2.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0	1.0
6	1.0	1.0	2.0	1.0	1.0
	1.0	1.0	1.0	1.0	2.0
Mean	1.00	1.08	1.17	1.00	1.25

Table A-4.2 Sensory evaluation data of quarg after 1 week of storage. The quarg was produced with 1% starter culture (Flora Danica) and different levels of rennet.

Judges	Rennet level (units/1000 kg milk)				
	0	388	775	1550	3876
1	1.0	2.0	1.0	1.0	4.0
	1.0	1.0	2.0	1.0	2.0
2	1.0	1.0	1.0	2.0	1.0
	1.0	1.0	1.0	1.0	1.0
3	1.0	1.0	1.0	2.0	4.0
	1.0	1.0	1.0	3.0	1.0
4	1.0	1.0	1.0	2.0	4.0
	1.0	1.0	2.0	2.0	3.0
5	1.0	1.0	1.0	1.0	2.0
	1.0	1.0	1.0	2.0	1.0
6	1.0	2.0	2.0	2.0	3.0
	1.0	1.0	1.0	1.0	3.0
Mean	1.00	1.17	1.25	1.67	2.42

Table A-4.3 Sensory evaluation data of quarg after 2 weeks of storage. The quarg was produced with 1% starter culture (Flora Danica) and different levels of rennet.

Judges	Rennet level (units/1000 kg milk)				
	0	388	775	1550	3876
1	1.0	2.0	2.0	1.0	4.0
	1.0	1.0	1.0	2.0	3.0
2	1.0	1.0	1.0	1.0	2.0
	1.0	1.0	1.0	1.0	3.0
3	2.0	1.0	2.0	2.0	1.0
	1.0	1.0	1.0	3.0	4.0
4	1.0	1.0	3.0	2.0	3.0
	1.0	1.0	1.0	2.0	3.0
5	1.0	1.0	1.0	1.0	2.0
	1.0	1.0	1.0	1.0	3.0
6	1.0	2.0	2.0	2.0	3.0
	1.0	1.0	2.0	2.0	3.0
Mean	1.08	1.17	1.50	1.67	2.83

Table A-4.4 Sensory evaluation data of quarg after 3 weeks of storage. The quarg was produced with 1% starter culture (Flora Danica) and different levels of rennet.

Judges	Rennet level (units/1000 kg milk)				
	0	388	775	1550	3876
1	1.0	1.0	2.0	1.0	5.0
	1.0	1.0	1.0	2.0	4.0
2	1.0	1.0	1.0	3.0	4.0
	1.0	1.0	2.0	1.0	4.0
3	2.0	2.0	1.0	3.0	6.0
	1.0	1.0	1.0	3.0	4.0
4	1.0	1.0	1.0	2.0	5.0
	1.0	1.0	1.0	2.0	4.0
5	1.0	1.0	2.0	1.0	6.0
	1.0	1.0	1.0	3.0	3.0
6	1.0	2.0	1.0	3.0	5.0
	1.0	1.0	2.0	2.0	4.0
Mean	1.08	1.17	1.33	2.17	4.50

Table A-4.5 Sensory evaluation data of quarg after 4 weeks of storage. The quarg was produced with 1% starter culture (Flora Danica) and different levels of rennet.

Judges	Rennet level (units/1000 kg milk)				
	0	388	775	1550	3876
1	1.0	2.0	1.0	3.0	5.0
	2.0	1.0	3.0	3.0	5.0
2	1.0	1.0	2.0	2.0	5.0
	2.0	2.0	2.0	2.0	4.0
3	1.0	2.0	1.0	3.0	6.0
	1.0	2.0	2.0	4.0	5.0
4	1.0	2.0	2.0	3.0	5.0
	2.0	2.0	2.0	3.0	4.0
5	1.0	3.0	2.0	5.0	6.0
	1.0	1.0	2.0	4.0	4.0
6	1.0	1.0	3.0	2.0	6.0
	1.0	1.0	2.0	4.0	4.0
Mean	1.25	1.67	2.0	3.17	4.92

Table A-4.6 Sensory evaluation data of fresh quarg. The quarg was produced with 388 units rennet per 1000 kg milk and four different starter cultures.

Judges	Starter Cultures			
	Flora Danica		<i>S. cremoris</i>	
	with <i>S. diacetylactis</i>	without	Single Strains - # 134	Single Strains - # 584
1	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0
2	1.0	1.0	1.0	1.0
	1.0	2.0	1.0	1.0
3	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0
4	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0
5	2.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0
6	1.0	2.0	1.0	1.0
	1.0	2.0	1.0	1.0
Mean	1.08	1.25	1.0	1.0

Table A-4.7 Sensory evaluation data of quarg after 1 week of storage. The quarg was produced with 388 units rennet per 1000 kg milk and four different starter cultures.

Judges.	Starter Cultures			
	Flora Danica		<i>S. cremoris</i>	
	with	without	Single Strains	
	<i>S. diacetylactis</i>		# 134	# 584
1	2.0	1.0	1.0	1.0
	1.0	2.0	1.0	1.0
2	1.0	1.0	1.0	1.0
	1.0	2.0	1.0	1.0
3	1.0	1.0	1.0	1.0
	1.0	2.0	1.0	1.0
4	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0
5	1.0	1.0	1.0	1.0
	1.0	1.0	2.0	1.0
6	2.0	2.0	2.0	2.0
	1.0	1.0	1.0	1.0
Mean	1.17	1.33	1.17	1.08

Table A-4.8 Sensory evaluation data of quarg after 2 weeks of storage. The quarg was produced with 388 units rennet per 1000 kg milk and four different starter cultures.

Judges	Starter Cultures			
	Flora Danica		<i>S. cremoris</i>	
	with	without	Single Strains	
	<i>S. diacetylactis</i>		# 134	# 584
1	2.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0
2	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0
3	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0
4	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	1.0
5	1.0	1.0	1.0	1.0
	1.0	1.0	1.0	2.0
6	2.0	1.0	1.0	1.1
	1.0	1.0	1.0	1.1
Mean	1.17	1.00	1.00	1.08

Table A-4.9 Sensory evaluation data of quarg after 3 weeks of storage. The quarg was produced with 388 units rennet per 1000 kg milk and four different starter cultures.

Judges	Starter Cultures			
	Flora Danica		<i>S. cremoris</i>	
	with	without	Single Strains	
	<i>S. diacetylactis</i>		# 134	# 584
1	2.0	2.0	1.0	1.0
	1.0	1.0	1.0	1.0
2	1.0	2.0	1.0	1.0
	2.0	1.0	1.0	1.0
3	1.0	2.0	1.0	1.0
	1.0	1.0	1.0	1.0
4	1.0	2.0	1.0	1.0
	1.0	2.0	1.0	1.0
5	2.0	1.0	1.0	1.0
	1.0	1.0	1.0	2.0
6	1.0	3.0	1.0	1.0
	2.0	2.0	1.0	1.0
Mean	1.33	1.67	1.00	1.08

Table A-4.10 Sensory evaluation data of quarg after 4 weeks of storage. The quarg was produced with 388 units rennet per 1000 kg milk and four different starter cultures.

Judges	Starter Cultures			
	Flora Danica		<i>S. cremoris</i>	
	with	without	Single Strains	
	<i>S. diacetilactis</i>		# 134	# 584
1	1.0	2.0	1.0	1.0
	3.0	2.0	1.0	1.0
2	2.0	2.0	1.0	1.0
	2.0	2.0	1.0	1.0
3	1.0	1.0	1.0	1.0
	2.0	2.0	1.0	1.0
4	2.0	3.0	1.0	1.0
	2.0	2.0	1.0	1.0
5	2.0	1.0	1.0	1.0
	2.0	1.0	1.0	2.0
6	3.0	2.0	1.0	1.0
	2.0	3.0	2.0	1.0
Mean	2.00	1.92	1.08	1.08