UNIVERSITY OF ALBERTA

THE TECHNOLOGICAL APPROACHES IN THE DEVELOPMENT OF A WHEY-BASED YOGURT BEVERAGE

by

Heather M. Burton-Trapp

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University of Alberta
Edmonton

Canada T6G 2P5

Department of Food Science
Faculty of Agriculture and Forestry

2-06 Agriculture-Forestry Centre, Telephone (403) 492-3236
Fax (403) 492-4265

January 9, 1991

Faculty of Graduate Studies
2-8 University Hall
University of Alberta
Edmonton, Alberta

To Whom It May Concern:

This is to certify that Ms. Heather Trapp has my permission to use a table of data from my work, "Whey cheeses and beverages" in her MSc thesis.

P. Jelen, PhD, P. Eng.
Professor

PJ/gm

UNIVERSITY OF ALBERTA

RELEASE FORM

NAME OF AUTHOR: Heather M. Burton-Trapp

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(SIGNED)

PERMANENT ADDRESS:
561 Lessard Drive
Edmonton, Alberta
T6M 1A9

DATED ________, 1990.

THE UNIVERSITY OF ALBERTA

FACULTY OF GRADUATE STUDIES AND RESEARCH

The undersigned certify that they have read, and recommended to the faculty of Graduate Studies and Research for acceptance, a thesis entitled **Technological approaches in the development of a whey-based yogurt beverage** submitted by **Heather M. Burton-Trapp** in partial fulfilment of the requirements for the degree of MASTER OF SCIENCE in FOOD PROCESSING.

Dr. P. Jelen (Supervisor)

Dr. B. Ooraikul

Dr. F. Temelli

Date Nov 29, 1990

To my wonderful husband, Robert.

ABSTRACT

The use of unprocessed cottage cheese whey in cultured milk beverages is a nutritional and economically logical alternative in the current problem of whey disposal. In this research project, the technological approaches for the development of a whey-based yogurt beverage were examined in three progressive experiments.

In the first section of this work various formulations in the production of a whey-containing yogurt beverage were examined and the most suitable formulation in terms of stability and consistency was determined to be the product made by diluting set yogurt with unprocessed cottage cheese whey. This formulation was employed in the second section of this work which involved an examination of the relationship between the product's rheological characteristics and stability.

Based on the visual stability of the yogurt beverage, 0.3% pectin was found to be the minimum amount of polysaccharide stabilizer required to result in a product having little or no whey separation upon storage for 4 weeks at 4.0°C. These findings were also reflected in the results of the particle size determination and the pulse nuclear magnetic resonance test. However, the results of these two tests were determined inconclusive, with regard to the stability of the product, considering the water-containing nature of the suspended aggregates in the product. In an examination of the product's rheological parameters of elasticity and viscosity, a

negative correlation between them and the visual stability was revealed, suggesting that the results of these tests alone do not reliably reflect the stability of the product in question.

The final section of this project involved a study of the stabilization of a product manufactured with simulated and real ultrafiltration retentate of cottage cheese whey. Based on the results of this section, the most successful method of stabilization against heat-induced precipitation of the whey proteins in solution was found to be the adjustment of pH to 4.0. The use of pectin as a stabilizing agent had some positive effect at a concentration of 0.3% but this effectiveness was limited to a pH below 5.0.

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1. INTRODUCTION

1.1. BACKGROUND INFORMATION

Whey, the yellow-green aqueous portion of milk separated from the curd during cheese manufacture, constitutes approximately ninety percent of the total volume of milk used in cheese production (Jones, 1974; Kosikowski, 1979; Zall, 1979). Acid whey, having a pH between 4.4 and 4.6 (Knopp, 1988; Patocka *et al.*, 1987), is a by-product of the manufacture of unripened cheeses such as cottage cheese, cream cheese, and quarg. It is also one of the largest sources of waste from modern dairies (Marwaha and Kennedy, 1988).

Since most of the lactose of the original cheese milk remains in the whey after curd separation, cheese whey has a high biochemical oxygen demand (approximately 35,000 mg/L) which poses an environmental threat when discharged directly into natural waterways (Maddox and Archer, 1984). In light of the more restrictive anti-pollution legislation and high sewage costs, many dairies are looking for alternative solutions to their whey disposal problems.

Currently, whey is used in a wide variety of products in several different forms (liquid, concentrated, or dried). Some of these products are listed in Table 1.1. Other whey-containing products include: Animal feeds, pharmaceuticals, infant formulas, frozen desserts, beverages, and baked goods (Coton, 1985; Modler, 1982; Knopp, 1988; Kosaric and Asher, 1982).

Table 1.1. Current uses of whey solids in foods*

Use	Percent of Total Solids
Baked goods	22.3
Processed cheese	26.6
Prepared dry mixes	22.4
Processed meats	0.3
Confectionary	1.2
Margarine	1.0
Soft drinks	0.1
Soups	1.1

*Adapted from Coton, 1985; Knopp, 1988.

Whey contains nutritionally valuable components such as whey proteins, L+ lactic acid, calcium, phosphate, and vitamins including B_1, B_2, and B_6 (Prendergast, 1985). Some of the amino acids found in whey protein are listed in Table 1.2 (Mann, 1975). These components make whey a valuable, nutritious food ingredient and for this reason, the consideration of this product as a food resource rather than as waste would be beneficial to some food and dairy manufacturers.

Table 1.2. Percentage of essential amino acids in certain proteins*

Essential Amino Acids	Whey protein	Egg	Soya
Isoleucine	6.55	6.45	5.15
Leucine	14.0	8.30	7.85
Lysine	10.9	7.05	6.20
Methionine	2.35	3.40	1.35
Cystine	3.15	2.25	1.35
Phenylalanine	4.05	5.80	5.10
Tyrosine	4.80	4.05	5.10
Threonine	6.70	5.15	4.10
Tryptophane	3.20	1.50	1.25
Valine	6.85	7.15	5.30

*Adapted from Palmer, 1980.

There have been many studies on the feasibility and acceptability of using whey and whey products in beverages (Beukema, 1990; Jelen *et al.*, 1987a; Kravchenko, 1987; Tratnik and Krsev, 1988) and in cultured milk products (Broome *et al.*, 1982; Guirguis *et al.*, Jelen and Horbal, 1978; Jelen *et al.*, 1987b; Tamime *et al.*, 1989). However, little research has been carried out on the utilization of unprocessed, liquid, cottage cheese whey in food products, such as cultured milk beverages (Young *et al.*, 1980).

1.2. RESEARCH OBJECTIVES

The main research objective of this work was to examine the various technological aspects involving the production of a drinkable whey-based yogurt beverage. Specific objectives included: (1) Comparing several possible technological approaches based on the stability and apparent viscosity of products, and efficiency of manufacture; (2) Determining a method of predicting the stability of the beverage using advanced physical methods including viscosity, dynamic rheology, particle size distribution, and pulse nuclear magnetic resonance and comparing the results of these methods to the results of a visual stability examination; (3) Determining the heat stabilizing factors of a whey-based yogurt product containing a high percentage of whey proteins, with specific focus on the stability of the whey proteins as affected by the adjustment of pH and the addition of pectin.

2. LITERATURE REVIEW

2.1. WHEY

Whey is the liquid part of milk remaining following the separation of the curd produced from coagulation of milk by acid or proteolytic enzymes (Zadow, 1986). The composition of whey, as shown in Table 2.1, varies depending on the type of cheese that is produced. Whey is differentiated into one of two types depending upon its titratable acidity. Sweet whey is produced from rennet-coagulated cheeses and has a titratable acidity of approximately 0.1 - 0.2 percent; acid whey, produced from fresh

Table 2.1. Proximate composition of whey*

Constituent	Acid whey (% w/w)	Sweet whey (% w/w)
Water	93.5	93.7
Lactose	4.90	4.95
Lactic acid	0.40	0.05
Fat	< 0.1	0.50
Protein	0.75	0.80
Ash	0.80	0.50
pH	4.3 - 4.6	5.7 - 6.3

*Adapted from Coton, 1976.

cottage cheese or quarg manufacture, has a titratable acidity greater than 0.4 percent (Zadow, 1986).

2.1.1. Composition of Acid Whey

Acid whey, the by-product of unripened cheese production, has a pH of approximately 4.3 - 4.6 (Short, 1978) and contains approximately 6 percent total solids (Harper, 1984) of which 70 percent is lactose and 10 - 12 percent is the nutritive serum or whey proteins (Demott, 1974).

Whey proteins constitute approximately 20 percent of the total milk protein (Harper, 1984) and consist of ß-lactoglobulin (ß-lg), α-lactalbumin (α-la), bovine serum albumin (BSA), the

Table 2.2 Major whey proteins*

Protein	% of Total Whey Protein
ß-Lactoglobulin	55-65
α-Lactalbumin	15-25
Proteose-peptones	10-20
Immune globulins	10-15
Bovine serum albumin	5-6
Soluble caseins	1-2
Minor proteins	<0.5

*Adapted from Harper, 1984.

immunoglobulins (Ig), and proteose peptones (Evans and Gordon, 1980). The major whey proteins and their concentrations in whey are listed in Table 2.2.

The most abundant protein found in whey, comprising 55 percent of the total protein content, is ß-lactoglobulin and α-lactalbumin is the next most abundant, comprising 20 percent (Harper, 1984). The minor milk proteins found in whey contribute less towards the total protein content but from a chemical standpoint, are significant components none-the-less.

2.1.2. Characteristics of Whey and Whey Proteins

The use of cottage cheese whey in its unprocessed state may lend advantages to the dairy industry in two ways. Firstly, it serves to reduce the amount of money spent on waste processing and/or the shipping of whey to farms as fertilizer. Secondly, no further monetary investment in equipment for whey processing is needed. However, one problem encountered when using whey or whey ultrafiltration (UF) retentate in the production of a food product is its instability during heat processing.

Generally, the major whey proteins are sensitive to heat at temperatures above 70°C (Patocka *et al.*, 1987) and this sensitivity is related to many parameters including molecular structure, concentration, ionic strength, the presence of calcium or other ions, and pH (Evans and Gordon, 1980; Patocka *et al.*, 1986). The sensitivity of the whey proteins to heat is characterized by their denaturation due to the unfolding of their compact globular conformations to a random conformation upon heating to approximately 68°C or higher. As well, at certain degrees of unfolding, denaturation may be accompanied by aggregation as a result of hydrophobic interactions and intermolecular disulphide interchange resulting in the formation of a precipitate (Evans and Gordon, 1980).

Jelen and Buchheim (1984) investigated the undesirable denaturation and coagulation of whey proteins that may occur during the industrial production of some whey-containing foods including yogurt beverages. Upon examination of the whey proteins following heating in acidic conditions, Jelen and Buchheim (1984) confirmed the occurrence of heat-induced precipitation of the whey proteins evident in the formation of a visible sediment layer during storage. This defect was attributed to the thermal processing of the product in conditions such as a pH level greater than 4.0, that favours the precipitation of the whey proteins.

Numerous studies involving the heat stability of whey proteins in acidic conditions have indicated that the resistance to heat-induced coagulation increases significantly below the rather narrowly defined pH range of 3.7 - 3.9 (Patocka *et al.*, 1987; Jelen and Buchheim, 1984). Burgess and Kelly, (1979) reported that following heating, the solubility of denatured whey proteins is at its lowest point at approximately pH 4.5 but increases significantly below pH 3.0.

2.1.3. Stabilization of the Whey Proteins

In the past, there have been literature citations on the heat stability of whey proteins and methods of monitoring it (Sawyer, 1968; deWit *et al.*, 1988). Some of these reviews have included the effects of casein, calcium, and pH adjustment on the stability of whey proteins (Elfagm and Wheelock, 1978; Jelen *et al.*, 1987b; Patocka *et al.*, 1986), however, there have been few studies on methods of preventing the heat-induced denaturation and subsequent precipitation of whey proteins by means of external factors such as the addition of pectin.

2.2. PECTIN

2.2.1. The Structure and Properties of Pectin

Pectin is a complex, high molecular weight polysaccharide obtained most often from the peels of citrus fruit (Ranganna, 1977). Pectin, as shown in Figure 2.1, consists of an unbranched polysaccharide chain containing 200 - 1000 galacturonic acid units linked together by α(1-4)-glucosidic bonds (Christensen, 1982) and interrupted at intervals with α(1-2)- L-rhamno-pyranosyl residues. Other sugars attached in side chains include D-galactose, L-arabinose, D-xylose, and L-fucose (Koster *et al.*, 1989).

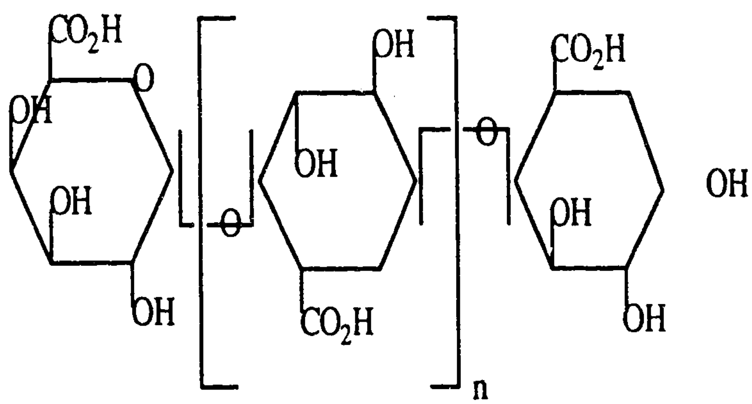

Figure 2.1. Structure of pectin D-galacturonan.
Adapted from Towle and Christensen, 1973.

Often a percentage of the galacturonic acid units are esterified by methanol and the ratio of esterified units to total galacturonic acid units in the pectin molecule is known as the degree of esterification (Towle and Christensen, 1973). High ester pectin, also called high methoxy (HM) pectin, as shown in Figure 2.2, is pectin that has greater than 50 percent of its carboxylic acid groups esterified. Low ester pectin, or low methoxy (LM) pectin, is pectin

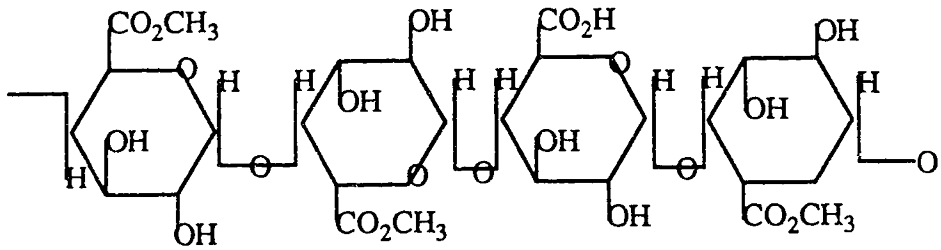

Figure 2.2. Structure of high methoxy pectin.
Adapted from Koster *et al.*, 1989.

with less than 50 percent esterification (Christensen, 1982). Due to its structural and chemical properties, HM pectin has been suggested for use as a stabilizer in certain cultured milk beverages (Towle and Christensen, 1973) and in beverages containing whey (Arolski *et al.*, 1977).

2.2.2. The Stabilizing Action of a High Methoxy Pectin

In one method of manufacturing a yogurt beverage, the casein gel of a set yogurt is broken by agitation and converted to a relatively stable suspension (Iversen, 1984). When this micellar suspension is pasteurized to increase the product's shelf life, aggregation or dehydration of the suspended casein may occur leading to sedimentation and separation of the aqueous phase upon standing (Foley and Mulcahy, 1989). To counteract this phenomenon, certain negatively charged polysaccharides, such as a high methoxy pectin, may be added which interact with the casein micelles keeping the particles in suspension (Iversen, 1984).

Increasing the viscosity of a yogurt beverage with a thickening agent such as HM pectin has been shown to prevent protein precipitation and whey syneresis during storage (Glahn, 1982). The increase in viscosity and decrease in particle size of the whey-based yogurt beverage with a HM pectin, can be explained by the binding of the stabilizer to the positively charged casein particles at a pH below their isoelectric point (Groven, 1987). This positive charge will gradually diminish as the negatively charged stabilizer binds to the particles. When this occurs, the Coulombic repulsion between the particles counteracts their tendency to adhere to one another and create larger particles which may gravitationally sediment out (Gregory, 1986). As a result, stabilization is achieved.

2.3. YOGURT

2.3.1. The Casein Micelles In Milk

During the fermentation of milk, the casein micelles play a significant role in the formation of a milk gel. The approximate composition of the casein micelles is given in Table 2.3. Casein micelles are aggregates composed, generally, of a group of strongly interacting proteins whose exact nature of bonding is not entirely known, although bonds such as hydrophobic, hydrogen, disulfide, electrostatic, and calcium bonding have been found to occur between them (Schmidt, 1982).

Table 2.3. Composition of the casein micelles in bovine milk*

Component	Content (g/100g micelles)	
Total casein:	93.3	
of which:		
α_{s1} - Casein		35.6
α_{s2} - Casein		9.9
β - Casein		33.6
κ - Casein		11.9
χ -, R-,S-, and TS - Casein		2.3
Total inorganic material:	6.6	
of which:		
Calcium		2.87
Magnesium		0.11
Sodium		0.11
Potassium		0.26
Inorganic phosphate		2.89
Citrate		0.40

*Adapted from Schmidt, 1982.

In 1976, Schmidt and Payens hypothesized the 'Subunit Theory' of casein micelles suggesting that the micelles consist of a large number of submicelles held together by colloidal calcium phosphate in an open, disordered structure. This theory has been widely supported and is based primarily upon the appearance of the casein micelles under the electron microscope. However, this theory does not account for the fact that the subunits may vary in composition or that the hydrodynamic radius of a micelle remains constant during the early stages of calcium ion dissociation, implying that the construction of the micelle is of a molecular network including colloidal calcium and phosphorus. In view of these contradictions, the 'Network Model' of the micellar structure was formed on the basis that each individual micelle is a large chemical compound composed of branched or cross-linked caseins (Dickinson and Stainsby, 1982; Schmidt, 1982). It is known that casein micelles have an open, sponge-like structure that is fairly heat stable but can be aggregated at the isoelectric point of casein, pH 4.6 (Dickinson and Stainsby, 1982; Schmidt, 1982).

2.3.2. The Manufacture of Yogurt

The fermentation of milk is one of the oldest methods of mi' . preservation (Tamime and Deeth, 1980) and has seen few dramatic changes in its long history of production. As a result, today's yogurt produced by contemporary manufacturing practices very closely resembles the original yogurt - like product that was developed in the Middle East thousands of years ago (Parnell-Clunies *et al.*, 1986). Today, yogurt is one of the most popular cultured dairy products in North America (Morley, 1979) and is produced from cow's milk using the symbiotic relationship of an added defined starter culture consisting of *Streptococcus thermophilus* and *Lactobacillus bulgaricus* (The new terminology for these traditional microorganisms is *Streptococcus salivarius* subsp. *thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus,* respectively; Heertje *et al.*, 1985).

The technical aspects of yogurt production may vary from operation to operation but generally include the following procedures: (1) Preliminary handling of the milk including screening for antibiotics and standardization of the fat and solids-non-fat contents; (2) Standardization/Fortification of the basic mix; (3) Homogenization; (4) Heat treatment; (5) Inoculation and fermentation; (6) Cooling, addition of sweeteners and flavours, and packaging (Tamime and Robinson, 1985). All processing steps in the manufacture of yogurt ultimately affect the physical characteristics of the end product, however, the most important factor affecting the final product is the heat treatment of the yogurt milk prior to inoculation of the culture and incubation (Tamime and Robinson, 1988).

2.3.3. Casein Interactions in Milk During the Heat Treatment of the Yogurt Milk and Fermentation

The application of heat to milk prior to yogurt production is a universal practice that results in a change of the physico-chemical structure of the milk proteins (Dannenberg and Kessler, 1988) and a slightly lower pH due to the decomposition of some of the lactose to organic acids and the redistribution of calcium, phosphorus, and magnesium (Tamime and Deeth, 1980). Extensive research on the effect of heat on the physical characteristics of yogurt has shown that heating to 85°C for 30 minutes produces the most desirable end product in terms of stability and viscosity of the yogurt gel (Kalab *et al.*, 1976; Mottar *et al.*, 1989).

During the heat treatment step the whey proteins undergo denaturation which significantly affects the properties of the final yogurt product. Specifically, the principal whey protein, ß-lg, plays a significant role during this heating step as it almost completely denatures and interacts with casein to increase the size of the casein micelle and its hydrophilic properties. An ultrastructural investigation of the casein micelles by Davis *et al.* (1978) revealed the formation of filamentous appendages consisting of denatured ß-lg attatched to casein micelles. This attatchment involved a disulfide linkage to κ-casein and, theoretically, would tend to inhibit micellar contact and fusion, thereby decreasing whey syneresis and increasing the firmness of the yogurt gel.

During the fermentation of yogurt milk the pH drops as starter microorganisms convert lactose to lactic acid. This acidity development destabilizes the casein micelle/denatured whey protein complex by solubilizing the calcium phosphate/citrate groups. When this occurs, the micelles aggregate and partially coalesce (Tamime and Robinson, 1985) to form a casein-whey protein gel network. This network entraps within it all the other constituents of the yogurt milk, including water, to form the yogurt gel (Iversen, 1984).

2.3.4. Yogurt Products

Prior to the 1960's yogurt did not receive as much popularity among North American consumers as it does now primarily because of its low market image and the fact that yogurt possesses a distinctly acidic, sharp flavour which may limit its acceptability. With the introduction of fruit and fruit flavours to yogurt, its popularity quickly rose causing processors to compete for ideas for new and innovative yogurt-based products (Tamime and Robinson, 1985).

Some of the more common yogurt products include the two main types of yogurt, set (sundae style and plain) and stirred, whose consistencies are based on their method of production and the physical structure of their coagulum (Tamime and Deeth, 1980). More recently, naturally or artificially flavoured fruit yogurts have been introduced to increase the marketability of this product. Other yogurt innovations include ultrahigh temperature (UHT) processed yogurt (where permitted by law), lactose hydrolyzed yogurt, concentrated yogurt, frozen yogurt, carbonated yogurt, artificially sweetened yogurt, and yogurt beverages (Mann, 1984a).

2.3.5. Yogurt Beverages

Of all the many recent introductions to the yogurt line of products, one of the most popular ones world wide has been fluid or drinkable yogurt (Mann, 1987). The results of a consumer survey taken in the USA in 1984 showed that most people prefered fruit flavoured yogurt beverages to all other types of yogurt-based products. The results of this survey further showed that a yogurt-type beverage product would be acceptable to a major segment of the consumer market and especially to those in the 19 - 40 year age bracket. There was some speculation from this investigation that the popularity of yogurt beverages would rise even further as consumers become increasingly aware of the nutritional importance of such foods (Mann, 1987).

Fluid - or drinkable - yogurt, is often stirred yogurt of low viscosity which is made when the yogurt coagulum is produced in bulk and the gel structure broken before cooling (Morley, 1979). Traditionally, fluid yogurt is manufactured by mixing equal amounts of yogurt and water (Towler, 1984); however, more recent techniques involve using a two stage homogenizer without added water (Glahn, 1980) or the addition of liquid cheese whey to yogurt (Tratnik and Krsev, 1988).

A recent tour of western European dairies and dairy markets (Trapp, personal experience) revealed a very large variety of yogurt-based beverages including some containing whey. Table 2.4 outlines a comparison of current yogurt-based beverages available in Europe and North America.

Table 2.4. A comparison of the composition of yogurt-based beverages*

Ingredients	Product		
	Yor	Interlac	Yogho-Yogho
Skim milk yogurt	46.3	44.0	90.8
Whey	43.0	45.0	- - -
Fruit juice	4.3	4.0	1.0
Sugar	5.8	7.0	7.4
Stabilizer	0.5	n.s.†	0.5

† n. s. = not specified
* Source: Jelen, 1990

The concept of a whey-based yogurt beverage has good implications from both a manufacturer's standpoint, in terms of the large profit margin possible from using an ingredient that would otherwise constitute waste, and from the consumer's standpoint in terms of nutritional value. The main disadvantage, however, of using whey in a product that would require a heat pasteurization process for microbial stability is the extreme sensitivity of the whey proteins to temperatures above 70°C (Patocka *et al.*, 1987).

2.3.6. Stability of a Yogurt Beverage

Unlike set style yogurt, the drinkable yogurt products with a low proportion of milk solids at low pH and a high proportion of sugar, are prone to sedimentation and milk protein separation which reduce its consumer appeal. Whey-based yogurt beverages are not an exception and their instability may actually be enhanced by the sensitivity of the whey proteins to heat (Jelen and Buchheim, 1984). In the past, some success in the stabilization of cultured milk beverages has been achieved with the addition of acidic polysaccharides as stabilizing agents (Towler, 1984; Glahn, 1980). Similar success has also been reported in the stabilization of whey beverages (Christensen, 1982). The use of the hydrocolloid stabilizers, propylene glycol alginate (PGA), carboxymethyl cellulose (CMC), or high methoxy (HM) pectin, has been found to minimize whey separation and sedimentation and it produces an acceptable viscosity in a yogurt beverage (Anon, 1982; Glahn, 1980; Groven, 1987; Towle and Christensen, 1973). Specifically, many researchers have reported that the stabilization provided by HM pectin in yogurt beverages is superior to that of other hydrocolloids under normal processing conditions (Foley and Mulcahy, 1989; Glahn, 1982; Towler, 1984).

2.4. RHEOLOGICAL METHODS FOR THE EVALUATION OF STABILITY OF HETEROGENEOUS LIQUID SYSTEMS

2.4.1. Basic Principles of Rheology

Rheology is the general study of deformation of matter in response to an applied force. This deformation of material as a result of an applied force provides useful information regarding that material's structure. If a material is perfectly solid it is called ideally elastic and under an applied stress (force) it will deform at a rate directly proportional to that force. When the force is removed, an ideally solid material will completely resume its original shape (McLeod, 1963). If a material is perfectly liquid it is called ideally viscous, and possesses no rigid structure. Under an applied stress a perfectly viscous material will move to a new position, without modifying its original structure, until the external force is removed. The rate at which this type of material deforms (shear rate Y) depends on the rate of the applied force (stress rate τ) per unit area (Prentice, 1984; Rha, 1975).

Perfectly elastic and perfectly viscous materials, whose relationship between shear rate and shear stress is shown in Figure 2.3, are called Newtonian fluids. The viscosity of most foods, that have a non-Newtonian structure, is not constant and varies with the rate of shear or stress. The ratio of the stress to the rate of strain at any given point is called the apparent viscosity. This relationship is outlined in Figure 2.3.

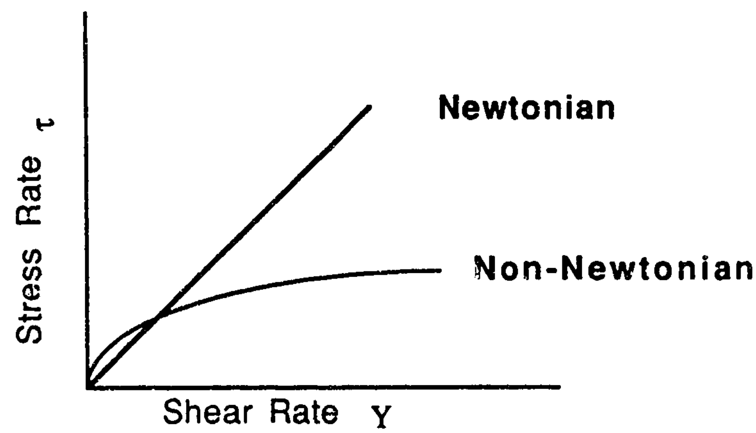

Figure 2.3. Flow behavior of Newtonian and non-Newtonian fluids.
Adapted from Rha, 1975.

Materials that exhibit an elastic type of structure in appearance but flow as liquids when under stress are said to be viscoelastic materials. Most foods, including yogurt beverages, are structurally viscoelastic and undergo both flow and deformation when subject to an applied stress.

When the plot of shear stress to shear rate of a fluid produces a non-linear curve through the origin, that fluid is said to be non - Newtonian. Non - Newtonian viscoelastic fluids are those fluids that exhibit time dependent elastic behavior (Geankoplis, 1983). More specifically, when the strain of elastic deformation of a viscoelastic element is dependent upon time, the material is considered to be a non - Newtonian viscoelastic fluid (Szczesniak, 1983). One rheological parameter often used to assess the structural nature of a non - Newtonian viscoelastic material is the viscosity measurement.

2.4.2. Viscosity

The viscosity of a fluid is related to its flow properties when an external stress is applied. Under an applied force, a fluid will flow at a rate that will be decreased by the internal forces of friction within the fluid. This decrease in flow rate or resistance to flow is measured by the fluid's viscosity (Johnson *et al.*, 1975).

Two factors affecting the apparent viscosity of a solution include the temperature of the solution and the nature of the particles suspended within it. A yogurt beverage consists of many suspended particles that make up its structural network. If these aggregates were to be broken down, the suspended liquid entrapped within the network would be released resulting in a decrease in the apparent viscosity of the yogurt beverage. For this reason, the stability of a material under stress also affects its structural performance.

Analytical methods which may be useful in determining the viscosity and stability of a heterogeneous viscoelastic material, such as a whey-based yogurt beverage, include the high frequency - high amplitude oscillation, strain sweep, and particle size distribution tests.

2.4.3. High Frequency - High Amplitude Oscillation

The high frequency - high amplitude oscillation test often forms the basis for the dynamic viscoelastic measurements of a material (Barfod *et al.*, 1990). Subjecting a viscoelastic material to a high frequency - high amplitude treatment over a specified time interval provides information regarding the factors that cause the structural molecules of the substance to adhere. When a stress is applied to a material sinusoidally with time, the behavior of the material is directly proportional to its rheological characteristics. If the material is perfectly elastic and has no viscous characteristics, the stress will be in phase (0°) with the strain. If the material is an ideally viscous Newtonian fluid, the stress will be in phase with the strain rate and have a phase angle of 90° to the strain. Almost all foods, including whey-based yogurt beverages, are viscoelastic materials that exhibit a phase angle between 0° and 90° (Szczesniak, 1983).

2.4.4. Strain Sweep

The purpose of the strain sweep test is to obtain information of the flow behavior of a viscoelastic fluid under minimal stress. This is accomplished by holding the frequency of stress (movement) constant while varying the amplitude of stress applied to the sample. From this information, the elastic modulus (G') and the viscous modulus (G") are given and define the total stiffness or apparent viscosity (G*) of the test sample (Barfod *et al.*, 1990).

Figure 2.4 details the normal flow behavior of a viscoelastic element during the strain sweep test. Non - Newtonian fluids, such as yogurt beverages, exhibit a decrease in apparent viscosity as the rate of strain is increased. This is usually attributed to a shear induced alignment of the molecules in the direction of flow to produce a decreased resistance to flow (Johnson *et al.*, 1975). This stage can be recognized as the destructive stage where, under increased shear or strain rate, the structure of the sample is broken down (Barfod *et al.*, 1990). When the alignment of molecules is complete, a second linear Newtonian region is reached where the viscosity is again independent of shear (Johnson *et al.*,1975) and no further decrease in viscosity is observed.

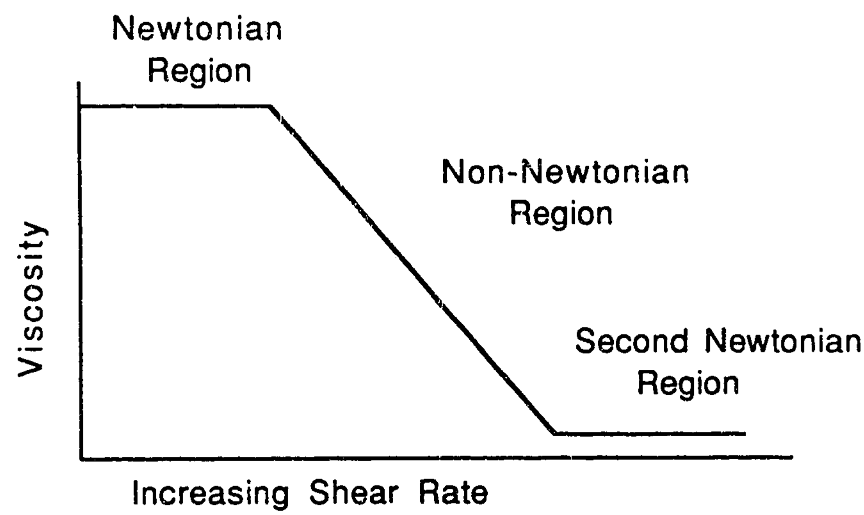

Figure 2.4. A Strain Sweep Curve.
Adapted from Johnson *et al.*, 1975; Barfod *et al.*, 1990.

The flow behavior of a material exhibited in the second Newtonian region of the strain sweep test is comparable to the flow behavior, or curve, obtained from a rotational viscometric test carried out under very low shear rate (Barfod *et al.*, 1990).

2.5. PARTICLE SIZE DISTRIBUTION FOR THE EVALUATION OF HETEROGENEOUS LIQUID SYSTEMS

The size of a particle is that dimension which best characterizes its state of subdivision (Stockham, 1977). For a spherical, homogeneous particle, the diameter is that dimension and can unambiguously be used as a measure of its size (Allen, 1968). The size of particles suspended in an emulsion or a suspension ultimately affects the stability, rheology, and quality of the final system (Irani and Callis, 1963) in that an increased tendency of the particles to collide and adhere to one another (as large sized particles do) decreases the apparent viscosity of the emulsion as the particles settle out (Allen, 1968).

The Fraunhofer diffraction, or low angle forward light scattering principle, is the most widely used method of particle size determination using light scattering. The Fraunhofer theory is based on the fact that when a beam of light strikes an assembly of particles, some of it is transmitted, some absorbed, and some scattered (Allen, 1968). The scattered radiation includes the diffracted, refracted, and reflected parts of the original beam and the absorbed radiation is retransmitted at a longer wavelength outside the range of the receiver (Allen, 1968). The mean size of the particles may be determined by measuring the intensity of the scattered light at fixed angles to the direction of the incident beam (Allen, 1968).

In practice, particles in suspension are illuminated by a collimated, monochromatic beam of light formed from a helium-neon laser. This light hits the particles and illuminates them forming diffraction patterns. These patterns are optically filtered and electronically summed to produce a multichannel histogram of the particle size distribution (Frock, 1987).

The particle size determination is based on the ratio of a particle's diameter to the wavelength of the monochromatic beam. For a particle whose radius is sufficiently large, compared to the wavelength, the intensity distribution of the Fraunhofer diffraction is given by the Airy formula:

$$I(w) = EK^2a^4 \left[\frac{J_1(Kaw)}{Kaw}\right]^2$$

where I(w) is the light intensity as a function of angle with respect to the optical axis; E is the intensity of the incident beam; $K=2\pi/\lambda$ (λ is wavelength); a is the particle radius; $w = \sin\theta$, with θ being the angle relative to the direction of the incident beam; and J_1 is the first order Bessel function of the first kind. This formula presumes that a particle having a diameter larger than the incident wavelength scatters the light in a direction similar to that which it was travelling prior to the interaction (Muly and Frock, 1982).

2.6. NUCLEAR MAGNETIC RESONANCE FOR THE EVALUATION OF HETEROGENEOUS LIQUID SYSTEMS

The water content of a heterogeneous system, such as food, influences the food's stability and controls the growth of microorganisms (Hardman, 1986). If bound water in a food can be defined as the water that remains in liquid state at sub-freezing temperatures, then nuclear magnetic resonance (NMR) may be used to measure this liquid water at low temperatures (Hardman, 1986). When polymers such as acidic polysaccharides are added to yogurt beverages for increased stability, they interact with some of the water present in the beverage, binding and retaining it, resulting in a decrease in syneresis and a more consistent product (Guirguis et al., 1984; Lillford, 1988).

The water binding effect of polysaccharide stabilizers in a whey-based yogurt beverage can be examined as a function of the transverse relaxation rate of protons by pulse nuclear magnetic resonance (pNMR). This relaxation rate or time may be affected by the concentration of stabilizer (pectin) used. The results of the relaxation rate of protons in the pNMR test may relate to the ability of the pectin-yogurt gel network to physically retain water.

2.6.1. Theory of NMR

Used to analyze the interactions of nuclei with their environments, NMR is a form of low energy spectroscopy commonly employed in the study of food systems (Streitwieser and Heathcock, 1981). For the pNMR examination of water in food the study of ^{1}H is the most desirable because its analysis is comparatively inexpensive and results are adequately accurate (Umbach, 1989).

A food sample consists of not one, but many nuclei whose environments influence the environments of the others based on the theory of quantum mechanics. This theory relates nuclear behavior to the distinct energy levels in which the nuclei can exist. The actual number of energy levels possible is equal to $2I + 1$ so for ^{1}H with $I = 1/2$, two energy levels are present and are termed parallel and antiparallel (where I is the spin quantum number of the nucleus).

The most stable and preferred energy state of any nucleus is the lowest energy state. This is parallel to the magnetic field. Energy of specific frequency and in a precise amount is applied to a sample. Upon absorption of this energy, the protons of the free water present in the sample are excited and move from a lower to higher energy level. In this instance, the system is said to be in resonance (Streitwieser and Heathcock, 1981). However, the nuclei are unable to remain at the higher energy level so they release the absorbed energy and return to the lower level. This is called relaxation of the water molecules and its rate is governed by the strength of the magnetic interactions between the water nuclei and the molecular constituents of the other components, such as pectin, present in the sample (Currie, 1984).

2.6.2. Pulse NMR

The behavior of the nuclear magnetization in a rotating 3-axis coordinate system best describes the fundamental elements of the pulse NMR experiments (pNMR) performed in this study. Figure 2.5 illustrates the events that occur in an NMR pulse.

A pulsed frequency of energy in the form of a spectrometric magnetic field is applied along the Z axis as the XY axes rotate around it. As the water protons, or nuclei, absorb the energy they become excited and align themselves randomly about the lowest energy state. When this occurs, a residual magnetization, or net magnetization (M_o) becomes evident along the Z axis (Dybowski and Lichter, 1987).

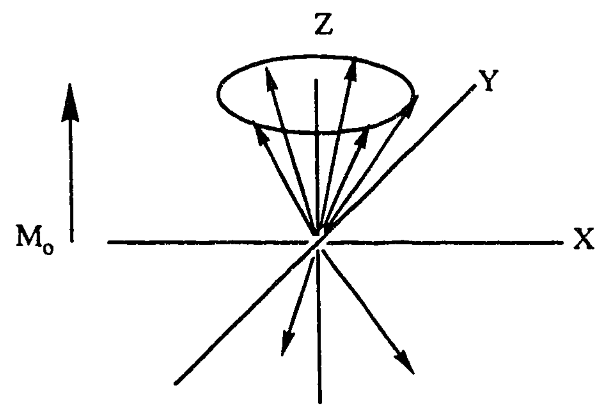

Figure 2.5. The net magnetization (M_0) of nuclei in the positive Z direction.
Adapted from Dybowski and Lichter, 1987.

As the excited protons lose their energy, relaxation occurs. In pulse NMR experiments, the net magnetization in the XY plane relaxes by the spin-spin relaxation (T_2) of protons whose rate is affected by the molecular interactions between the environments of the individual nuclei (Currie, 1984). T_2 relaxation is measured using a spin echo technique consisting of the application of, first, a 90° pulse followed by a series of 180° pulses (Hahn, 1950). These pulses used in most NMR experiments, 90° ($\pi/2$) and 180° (π), are important in that the time of pulse be regulated to ensure the exact 90 or 180 degree shift of net magnetization so as to minimize interference that would otherwise alter accurate readings (Umbach, 1989).

2.7. INVESTIGATION OF A WHEY-BASED YOGURT BEVERAGE

Within the approaches to the development of a whey-based yogurt beverage, many possible facets of investigation may be suggested for the important parameter of stability. A determination of the relationship between this parameter and various physical characteristics would be feasible if the results of these tests could accurately predict the stability of a cultured milk beverage or other heterogeneous systems including yogurt beverages, whey-based yogurt beverages, or whey UF retentate-based beverages.

An analysis of these physical characteristics could be carried out using the following methods: (1) A visual sedimentation test for stability; (2) The strain sweep and high frequency - high amplitude oscillation tests for characterizing the rheological properties (Barfod *et al.*,1990); (3) Particle size distribution for determining the size of particles suspended in a system (Muly and Frock, 1982); (4) Pulse nuclear magnetic resonance test for measuring the amount of bound water (Streitwieser and Heathcock, 1981).

When whey is used for no other purpose than as animal feed or agricultural fertilizer, the lost potential in terms of economics and nutrition is enormous. For this reason, the utilization of natural and modified whey as the chief ingredient in beverages has been, and continues to be, extensively researched.

2.7.1. The Feasibility of the Development of a Whey-Based Yogurt Beverage

Modifications of whey for use in whey-containing beverages have included fermentation, clarification using heat denaturation and ultrafiltration, demineralization, lactose hydrolysis, and deodorization (Kravchenko, 1987). The final whey beverages produced using these methods as a starting point can be enriched with fruit and fruit concentrates, vegetable concentrates or organic acids. In addition, these products could be further sweetened using sugar and sugar syrups (Kravchenko, 1987) or artificial sweeteners (Beukema, 1990).

Some examples of the types of cheese whey beverages produced using these methods include: (1) Those based on natural, unprocessed whey; (2) Those based on whey permeate or other refining process including clarification or deproteinization; (3) Those based on hydrolyzed whey permeate; (4) Those based on the ultrafiltered retentate of whey; (5) Those based on whey protein concentrates. Yogurt type beverages may be included in any category (except perhaps methods 2 and 3).

Within the past two decades, yogurt has emerged as one of the most popular cultured dairy products on the retail market (Lindsay *et al.*, 1981). The reason for this popularity is not only the unique flavour of yogurt but its image as a calorie wise, nutritionally valuable food that can be consumed for breakfast, as a dessert, or as a snack.

Ironically, the latest selling feature of this product has not necessarily been due to the diversity of yogurt-based products but rather to the desire of consumers to achieve greater health and longevity. This new health and nutrition outlook of consumers is based on the concept of incorporating desirable microflora in the intestines; a notion that was initiated and advocated by Eli Metchnikoff, a scientist who died without ever knowing the world wide impact of his "yogurt" theory of the early 1900's (Bibel, 1988). This "new" idea of promoting foods, such as yogurt, that contain high levels of viable microorganisms was launched on the hypothesis that bacteria such as *Lactobacillus bulgaricus*, *L. acidophilus*, and *Bifidobacterium bifidum* are beneficial colonizers of the intestines which may have many therapeutic qualities including decreasing the risk of cancer or coronary heart disease, and improving longevity (Mann, 1984b).

Whey-based yogurt beverages should also be included under this healthful image label and, in fact, should have the added selling feature of having a high protein and low fat content; characteristics that would be of interest to consumer groups such as athletes and expectant mothers (Mann, 1987). In light of the ever growing concern of waste and pollution, the development of a product that would serve both to promote positive waste management and to provide consumers with a nutritious, healthful food product suggests that there is great opportunity for the development of a whey-based yogurt beverage.

3. MATERIALS AND METHODS

In this research project, the investigation of a whey-based yogurt beverage was divided into three areas including: (1) Comparison of technological approaches in the development of a whey-based yogurt beverage; (2) The examination of the relationship between the rheological and stability characteristics of a whey-based yogurt beverage; (3) A study of the use of pH adjustment and acidic polysaccharide addition as methods of stabilizing whey proteins against heat-induced precipitation in simulated and real UF retentate model systems.

3.1. MATERIALS

3.1.1. Whey

Fresh cottage cheese whey having a pH of approximately 4.4 was supplied weekly by Palm Dairies Ltd. of Edmonton, Alberta and stored at 4.0°C. This whey was used, without further modification, in the production of the whey-based yogurt beverage made by diluting set yogurt with fluid, unprocessed cottage cheese whey, in the production of the beverage produced by liquid fermentation (section 3.2), and for the production of the samples based on genuine whey UF retentate.

In the study of the comparison of the rheological characteristics and stability of a whey-based yogurt beverage (section 3.3), fresh, fluid cottage cheese whey was unavailable, therefore, an acid whey model was constructed using Flavolin, a whole whey powder supplied by Grindsted Products A/S. Since the rehydrated whey had a pH of approximately 6.3, the pH was lowered to a level closer to that of fresh unprocessed acid whey (4.3) by dairy fermentation with a yogurt starter culture.

Concentrated whey protein solutions were used in this study in both real and simulated form. The ultrafiltration (UF) retentate of cottage cheese whey, having a total solids content of 13.1% and a protein content of approximately 3.0%, was produced on a DDS Lab-Module Ultrafiltration/Reverse Osmosis system (model no. 20-36 LAB, Nakskov, Denmark). This concentrated whey protein solution was used in the production of the whey-based yogurt beverages containing UF retentate (section 3.4).

Simulated UF retentate (SUFR) was used in section 3.5 and was produced by blending water with Flavolin, a dried whey powder consisting of 96% total solids and 35% protein and supplied by Grindsted Products A/S. This SUFR solution was used in the examination of the heat stabilization of whey proteins with pectin using the gravitational separation and apparent viscosity tests. Another SUFR solution was used in the analysis of the particle size distribution. This SUFR was produced using Lactoprodan 80, a dried

WPC containing 94.5% total solids and 78±4% protein, and supplied by Grindsted Products A/S.

When required, the whey-based yogurt beverages and some model systems were adjusted to a pH level of approximately 4.1 with 80% lactic acid. All ingredient concentrations listed in sections 3.2, 3.3, and 3.4 are given as a percentage of the total weight of the final product.

3.1.2. Stabilizer

Two stabilizers, Pectin 1400 and Mexpectin RS450, shown in Table 3.1, were used in the course of this research. Both stabilizers, high methoxy (HM) pectins produced by acid extraction from lime peels, were supplied by Grindsted Products A/S.

Table 3.1. Profile of the stabilizers used

Stabilizer	Pectin 1400	Mexpectin RS450
Product Number	044082	044969
Lot Number	115138	90925
DM*	70.7%	70-75%

* Degree of Methoxylation

Pectin 1400, a non commercial HM pectin, was used for the development of the whey-based yogurt beverages (sections 3.2 and 3.4), in the stability study of the UF retentate of cottage cheese whey (section 3.5) and in the confirmatory study (section 3.6). This stabilizer is only available as an experimental stabilizer and is not distributed for use in the food industry. Mexpectin RS450, is the commercially available counterpart of the pectin 1400 stabilizer. Results of studies on the use of HM pectin for the stabilization of cultured milk beverages by Towle and Christensen (1973), Smalbrink and van Hooydonk (1983), and Gregory (1986) plus the availability of the product, prompted the use of Mexpectin RS450 as the experimental stabilizer for the comparison of the rheological and stability characteristics of the whey-based yogurt beverage described in section 3.3.

3.1.3. Yogurt Culture

The starter culture used in the production of all the yogurt solutions was a mixture of *Streptococcus salivarius* subsp. *thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus*. This mixture was added in the form of a fresh commercial plain yogurt.

3.2. MANUFACTURE OF WHEY-BASED YOGURT BEVERAGES FOR TECHNOLOGICAL COMPARISONS

In these experiments, a yogurt milk base containing 13.4% total solids was prepared by reconstituting commercial skim milk powder (SMP) in water. An outline of the technological approaches used in this section is given in Table 3.2.

Table 3.2 Technological approaches in the development of a whey-based yogurt beverage

Method	Method of Production
3.2.1	Manufacture of a beverage by diluting set yogurt with unprocessed cottage cheese whey
3.2.2	Liquid fermentation of a yogurt milk base produced from unprocessed cottage cheese whey
3.2.3	Liquid fermentation of unpreheated yogurt milk base - no whey added
3.4.1	Dilution of the set yogurt with the UF retentate of cottage cheese whey
3.4.2	Liquid fermentation of a yogurt milk base produced from the UF retentate of cottage cheese whey
3.4.3	Use of the unfermented UF retentate of cottage cheese whey as the base for the production of a cultured milk beverage

The complete formulations of all the beverages described in this section are listed in Table 3.3. The solution of SMP in water was

Table 3.3. Formulation of yogurt beverages outlined in section 3.2

Method (Section)	Product	Ingredient	% (w/w)
3.2.1	yogurt:	water	86.6
		skim milk powder	13.4
		culture	3.0
	beverage:	yogurt mixture	43.0
		cottage cheese whey	51.1
		sucrose	5.6
		pectin	0.3
3.2.2	yogurt:	whey	92.0
		skim milk powder	5.0
		culture	3.0
	beverage:	yogurt mixture	94.1
		sucrose	5.6
		pectin	0.3
3.2.3	yogurt:	water	
		skim milk powder	as in 3.2.1
		culture	
	beverage:	yogurt mixture	
		sucrose	as in 3.2.2
		pectin	

allowed to rehydrate for four hours at 4.0°C, then it was heated for thirty minutes in an 80°C hot water bath and cooled to 37°C. The fermentation process was initiated by inoculation with a 3% yogurt culture in the form of a plain commercial yogurt. Following a twelve hour incubation period at 37°C, a pH reading was taken and, when required, adjustment to pH 4.1 was made using 80% lactic acid.

3.2.1. Dilution With Unprocessed Cottage Cheese Whey

In this approach, a beverage containing 5.6% sucrose in the form of Table sugar and 0.3% stabilizer was produced from a solution of set yogurt diluted with cottage cheese whey. This sample was homogenized on a Westinghouse WK16 dairy-type piston homogenizer (15M8BA Manton-Gaulin Mfg. Co., Everett, Mass.) at 345 bar then batch pasteurized at 78°C for five minutes in a hot water bath.

3.2.2. Using Cottage Cheese Whey in the Yogurt Milk Base

The method used in this experiment was similar to that described in section 3.2.1 with the fundamental difference being the ingredients in the yogurt milk base. Yogurt milk containing 13.4% total solids was prepared by reconstituting skim milk powder in fresh, unprocessed, cottage cheese whey. The solution was allowed to rehydrate for four hours at 4.0°C then it was heated to 65°C in a hot water bath and cooled to 37°C. After inoculation and incubation, beverages were produced by combining sucrose and stabilizer and adding this mixture to 94.1 percent of the yogurt mixture. The beverages were homogenized and pasteurized at 78°C then stored prior to analysis as in section 3.2.1.

3.2.3. Using an Unheated Yogurt Milk Base in the Yogurt Beverage

The beverage in this section was produced from yogurt as in the previous section but without the initial preheating of the yogurt milk. This beverage composition was similar to that in section 3.2.1 except whey was not added as the dilutant.

3.3. Production of the Yogurt Beverage Used For Rheological and Stability Comparisons.

The beverage produced by diluting set yogurt with unprocessed whey was chosen for further in-depth analysis of its physical properties and stability. The beverage formulation method is given in Table 3.4.

Table 3.4 Formulation of whey-based yogurt beverages outlined in section 3.3

Product	Ingredient	% (w/w)
yogurt:	water	86.6
	skim milk powder	13.4
	culture	3.0
whey:	water	92.9
	whole whey powder	7.1
	culture	3.0
whey/sucrose solution:	whey	35.0
	sucrose	65.0
sucrose/pectin solution:	whey/sucrose soln.	90.0
	pectin	10.0
beverage*:	yogurt	42.8-43.3
	whey	49.5-51.1
	sucrose	2.7-5.6
	sucrose/pectin soln.	0.0-5.0

*Beverages contained: 0-0.5% pectin and 5.6% sucrose (some was added as powder depending upon the addition of pectin).

In the production of the samples in this experiment, skim milk powder was first diluted with water to produce a solution containing 13.4% total solids. Culture in the form of a fresh commercial yogurt was added at a concentration of 3% by total weight and the mixture was allowed to incubate at 37°C for twelve hours. A simulated whey solution was produced by blending whole whey powder with water and allowing this mixture to hydrate for four hours at 4.0°C. Following hydration the simulated whey solution was inoculated with 3.0% culture and incubated at 37°C for twelve hours in order to achieve a pH level similar to that of fresh acid whey.

Following incubation and the pH adjustment, where required, the simulated whey solution was blended with sucrose (granulated Table sugar) to produce a solution containing 35% whey solution and 65% sugar. This mixture was stirred vigorously to dissolve the sugar, then blended with powdered Mexpectin RS450 to produce a solution containing 90% whey/sugar solution and 10% pectin. This final whey/sugar/pectin solution was used in the final beverage at concentrations of 0.0 - 5.0% to produce final beverages containing concentrations of 0.0, 0.1, 0.3, 0.4, or 0.5% pectin. In order to include the correct amounts of sugar, whey, and yogurt, these substances were added separately according to the specifications to achieve beverages containing final concentrations of 5.6% sucrose, 51.1% whey, and 42.8 - 43.3% yogurt (depending on the concentration of pectin).

These final beverage samples were then divided into two subsamples, A and B, and homogenized at 200 bar on a Rannie

Homogenizer (Lab Type 12.50A, Albertslund, Denmark). The set of subsamples designated A was homogenized and batch pasteurized at 78°C for five minutes (after a come-up time of 5-10 min) in a hot water bath. Subsamples B were homogenized and pasteurized on the same equipment at 90°C. All samples were hot filled into 250 mL bottles and placed in a 4.0°C still air refrigerator for 24 hours prior to further analysis.

3.4 METHODS USED IN THE PRODUCTION OF BEVERAGES BASED ON THE UF RETENTATE OF WHEY

The methods described in this section and summarized in Table 3.5, were similar to those used in the previous sections with the fundamental difference being the incorporation of genuine or simulated UF retentate into the beverages.

3.4.1. Diluting Set Yogurt with UF Retentate

The beverage outlined in section 3.4.1 was produced as that in section 3.2.1 with the difference being the use of UF retentate rather than unprocessed whey as the dilutant of set yogurt.

Table 3.5. Formulation of yogurt beverages outlined in section 3.4

Method	Product	Ingredient	% (w/w)
3.4.1	yogurt:	water	86.6
		skim milk powder	13.4
		culture	3.0
	beverage:	yogurt mixture	43.0
		whey UF retentate	51.1
		sucrose	5.6
		pectin	0.3
3.4.2	yogurt:	whey UF retentate	92.0
		skim milk powder	5.0
		culture	3.0
	beverage:	yogurt mixture	94.1
		sucrose	5.6
		pectin	0.3
3.4.3	yogurt base:	whey UF retentate	95.0
		skim milk powder	5.0
	beverage:	as in 3.4.2	

3.4.2. Using a Liquid Fermentation of UF Retentate

The beverage described in this section was produced as in section 3.2.2 with the difference being the use of the UF retentate of cottage cheese whey rather than unprocessed cottage cheese whey.

3.4.3. Using UF Retentate Without Fermentation

The beverage produced by this approach was made using the method outlined in section 3.4.2. However, in this experiment, the yogurt milk base produced with UF retentate was neither inoculated nor incubated, but was used directly in the production of beverages with final concentrations of 94.1% yogurt mixture, 5.6% sucrose, and 0.3% pectin.

3.5. THE STABILIZATION OF SIMULATED AND GENUINE UF RETENTATE-BASED PRODUCTS

3.5.1. Methods Used in the Production of SUFR-Based Model Systems

The product used in this study was made using the method outlined in Table 3.6. Dried whey protein concentrate was added to fresh tap water to produce simulated whey UF retentate solutions containing 3.0% protein. All solutions were mixed vigorously to disperse the whey powder then centrifuged in an International Equipment Centrifuge (S2K, International Equipment Co., Needham Heights, Mass., U.S.A.) to separate any insoluble particles.

Mexpectin RS450 was added at concentrations of 0.0, 0.3, or 0.5 percent to the aqueous whey protein solutions that had been adjusted to one of pH levels 4.0, 5.0, or 6.5. The mixtures were homogenized in a Westinghouse WK16 dairy-type piston homogenizer, as described in section 3.2.1, then batch pasteurized in a hot water bath at 78°C for five minutes.

Table 3.6. Formulation of yogurt beverages outlined in section 3.5.1

Product	Ingredient	% (w/w)
simulated whey UF retentate:		
	water	90.0
	dried WPC	10.0
pectin solution:		
	water	96.0
	pectin	4.0
beverage:		
	water	14.3/6.8/1.8
	whey retentate	85.7
	pectin soln.	0.0/7.5/12.5

3.5.2. Methods Used in the Production of Genuine UF Retentate-Based Beverages

The formula base of the yogurt samples in this experiment was produced by adjusting the pH of fresh UF retentate from approximately 4.4 to 5.5 with 1M NaOH. This allowed the pH level to fall during fermentation. The solution was heated to 37°C in a hot water bath, then inoculated with a 3% culture of plain, commercial yogurt, and incubated at 37°C for twelve hours. The fermented solution was divided in half and each half was again divided into three equal subsamples which were then adjusted to 0.0, 0.3, or 0.5% powdered pectin and 5.6% sucrose (granulated Table sugar) by total weight. The beverages were homogenized at room temperature and divided in half. One half of the samples were pasteurized for five minutes at 78°C, the other, for five minutes at 90°C in a hot water bath.

3.6. DETERMINATION OF VISUAL STABILITY OF THE EXPERIMENTAL BEVERAGES

A visual examination of whey separation was used as the principal stability test for the whey-based yogurt samples. This examination was carried out by filling samples to the 100 mL line in 100 mL graduated cylinders and allowing them to separate by gravity for four weeks at 4.0°C. After periods of four, fourteen, and 28 days the samples were visually inspected, and the amount of clear whey separated at the top was recorded as evidence of the heat instability of the beverages.

3.7. METHODS FOR OBJECTIVE ANALYSIS OF THE EXPERIMENTAL BEVERAGES

3.7.1. Viscosity Measurements

The apparent viscosity of the various yogurt beverages was determined using a Haake Rotoviscometer (Model RV3, Berlin, Germany) and calculated as follows:

$$\text{Apparent Viscosity} \quad (\eta) = \frac{G\,S}{n} \text{ mPa.sec}$$

Where G was the instrument factor 29.39; S, the scale reading; and n, the rpm setting. All readings were taken at 20°C maintained by a tempering vessel attatched to a Thermomix 1480 circulating water bath.

3.7.2. Rheology: Strain Sweep Test

The viscosity and dynamic rheology data for the beverage samples produced in section 3.3 were obtained with the strain sweep test. This was completed on a Bohlin VOR Rheometer (Ole Romers Vag 12, S-22370, Lund Sweden) with the equipment settings listed in Table 3.7.

The strain sweep test was carried out in duplicate on one day old samples. Due to the nature of the equipment used in the study, all samples were tested within the second linear area of the strain sweep curve.

Table 3.7. Parameter settings for the Bohlin Rheometer: Strain sweep test

Parameter	Setting
mode	Strain Sweep
temperature	20°C
frequency	1.0 Hz
amplitude	5%-100%
logarithmical amplitude sweep	5-100% (in 50 steps)
measuring system	C25
torsion bar	0.34 g/cm

Adapted from Anon, 1985.

3.7.3. Rheology: High Frequency - High Amplitude Oscillation Test

This test provided further information on the dynamic rheology of the beverage samples and, like the strain sweep test, was carried out on the Bohlin Rheometer on one day old samples. The equipment settings for this investigation are listed in Table 3.8.

Table 3.8. Parameter settings for the Bohlin Rheometer: High frequency - high amplitude oscillation test

Parameter	Setting
mode	Oscillation
temperature	20°C
frequency	5.0 Hz
amplitude	100%
measuring system	C25
torsion bar	1.77 g/cm

Adapted from Anon, 1985

3.7.4. Particle Size Analysis

The particle size distribution of the whey-based yogurt samples produced as described in sections 3.3 and 3.5 was analysed using a Malvern 2600 Easy Sizer (WR141AQ Malvern, Spring Lane, Worcestershire, England). One mL samples were diluted in a 9 mL citric acid/sodium phosphate buffer solution at pH 4.0. The samples were stirred by hand then added by drop to a sample cuvette containing a magnetic stirring bar and citric acid/sodium phosphate buffer solution until the obscuration of particles in the beam of monochromatic light was approximately 0.17 to 0.20 (Malvern 2600 Easy Sizer Manual).

3.7.5. Pulse Nuclear Magnetic Resonance

In this experiment the transverse relaxation time, or T_2, of the excited nuclei was examined. T_2 relaxation time was measured by Pulse NMR analysis carried out on an IBM PC/20 series NMR analyser (Minispec, Analytische Messtechnik GmbH, Bruker, West Germany). Whey/yogurt samples were equilibrated in NMR tubes for five minutes in a 40°C water bath prior to analysis. The settings used in the analysis of the whey-based yogurt beverage samples are outlined in Table 3.9. Each NMR measurement took approximately eight seconds and the resulting signals were transferred on line to a PS-2 IBM computer.

Table 3.9. Parameter settings for the Bruker Minispec NMR Analyser: Pulse NMR

Program	Parameter Settings
Pulse Sequence	3
Calculation Constant	87
Relaxation Delay (sec.)	2
Attenuation (mv)	37
Band Width of Signal	LOW
MODE	(DIO) Diode Detection

3.8. STATISTICAL TREATMENT OF DATA

Each set of experimental measurements was carried out at least in duplicate. The results of these tests were analysed statistically to determine if any significant differences existed between treatments including heat treatment, pectin concentration, or pH adjustment. The three statistical methods used in these examinations included the Duncan Multiple Range test, the Least Significant Difference test and the Correlation - Regression analysis test (Steele and Torrie, 1980). All the statistical examinations were carried out with the APL program of the Main Terminal System of the University of Alberta.

4. RESULTS AND DISCUSSION

4.1. A COMPARISON OF VARIOUS TECHNOLOGICAL APPROACHES IN THE DEVELOPMENT OF A WHEY-BASED YOGURT BEVERAGE

In the first section of this study, various technological approaches were used in the formulation of cultured whey-based yogurt beverages using fresh cottage cheese whey. These technological approaches included incorporating unprocessed cottage cheese whey and the ultrafiltered (UF) retentate of cottage cheese whey into cultured milk beverages.

While the aim of this research project was not one of product development, the stability and viscosities of the formulated beverages were compared to that of a commercially marketed product in order to maintain the standards of a yogurt beverage that had been successful on the retail market.

4.1.1. Beverages Based on Unprocessed Cottage Cheese Whey

The technological approaches examined in this experiment included blending unprocessed cottage cheese whey with set yogurt, fermenting an unheated yogurt mixture to a final liquid form, and incorporating unprocessed cottage cheese whey into a yogurt milk base. The viscosity and stability measurements of the yogurt beverages produced with fresh cottage cheese whey are compared to the commercial yogurt beverage in Table 4.1.

Table 4.1. Viscosity and stability measurements of the yogurt beverages produced by various technological approaches

Method of Production	pH	Viscosity (mPa.s)	Stability (mL Sep.*/100mL)
set yog. diluted with unprocessed whey	4.1	23.5	0
liquid fermentation: using unprocessed whey in the yogurt milk	4.2	37.8	84
liquid fermentation: no preheating of the yogurt milk	4.1	118.1	0
commercial yogurt beverage	3.8	64.1	0

*Separated whey

The viscosity of the commercial yogurt beverage was higher than that of the experimental whey-based yogurt beverage produced by diluting set yogurt with liquid whey. This experimental beverage, made from whey - diluted yogurt and subjected to a final homogenization step, was thin enough to be drawn through a drinking straw, yet still maintained some body (a quality which may be proven acceptable through consumer research).

Previous industrially produced yogurt beverages were manufactured by diluting set yogurt, using water, rather than whey, as the dilutant (Tamime and Deeth, 1980). However, more recent techniques used in the manufacture of drinking yogurt involve the homogenization of yogurt without additional fluid (Morley, 1979). One yogurt beverage produced in this study was produced without additional fluid and the yogurt milk base was not preheated prior to its inoculation and incubation. Theoretically, the formation of the yogurt gel during fermentation should have occurred less readily in this product because the κ-casein would not have been initiated to react with the ß-lactoglubulin in the preheating step (Zittle *et al.*, 1962; Kalab *et al.*, 1983). Instead, the casein would be expected to precipitate at its isoelectric point as the pH dropped. However, possibly because the yogurt milk was made with medium heat skim milk powder, a semi-firm gel was produced during incubation and the resulting homogenized yogurt beverage was quite stable but almost too viscous to pour.

Cottage cheese whey appeared to be more stable when it was subjected to a mild heat pasteurization after being blended into the final beverage than when it was heated in the yogurt milk, prior to culture inoculation. The beverage produced from the yogurt milk made by combining skim milk powder and fluid whey appeared to be quite viscous but readily separated upon standing despite the fact that the yogurt milk, made of unprocessed whey and skim milk powder was heated only to 65°C (to avoid the possible denaturation of the whey proteins). The formation of a small amount of precipitate during heating seemed to be unavoidable and the final fermented yogurt milk did not gel and had a coagulum at the bottom that closely resembled a soft, white precipitate. The final beverage was unhomogeneous, unstable, and generally not acceptable in appearance.

4.1.2 Beverages Based on the UF Retentate of Cottage Cheese Whey

The second part of this study involved using the genuine UF retentate of cottage cheese whey in the formulation of a cultured whey-based yogurt beverage. Table 4.2 outlines the average viscosity and stability measurements of the yogurt beverages produced with the UF retentate of cottage cheese whey. The viscosity of the beverage produced by blending set yogurt with the UF retentate was significantly lower than that of the commercial yogurt beverage as shown in Table 4.1. This lower viscosity was probably due to the dilution of the yogurt gel with fluid retentate and the homogenization of the final product. This product also appeared to be stable, showing no evidence of separation after twenty-eight days of storage at 4.0°C.

Table 4.2. Viscosity and stability measurements of yogurt beverages produced with the genuine UF retentate of cottage cheese whey

Method of Production	pH	Viscosity (mPa.s)	Stability (mLSep.*/100mL) (28 days)
set yogurt diluted with UF retentate	4.1	18.8	0
liquid fermentation: using UF retentate in the yogurt milk base	4.3	21.7	81
unfermented UF retentate used as the beverage base	4.9	22.5	28

*Separated whey

As in the previous section which involved using cottage cheese whey as an ingredient in the yogurt milk base, the stability of the retentate-based beverages decreased greatly when the whey UF retentate was used in the yogurt milk and heated prior to the addition of the other beverage ingredients. The gel that formed during the incubation of this yogurt mixture more closely resembled a soft curd-like precipitate than a firm gel. The beverage produced from this type of yogurt was very unstable and showed evidence of separation almost immediately, indicating that, even at the reduced heating temperature, the whey proteins were still destabilized.

The beverage produced from yogurt made from the UF retentate of whey was no more viscous in appearance than the beverages made with regular set yogurt, however, the curdy precipitate and unhomogeneity of this product resulted in a greater viscosity reading on the Rotoviscometer. This phenomenon resulted in an inconsistency between the results of the viscosity and stability tests as they neither coincided with nor reflected one another. This finding strongly suggests that the measurement of viscosity alone may not be sufficient for predicting the stability of the whey-based yogurt beverages.

The yogurt beverage made with the unfermented, unheated retentate of cottage cheese whey was significantly more stable than the UF retentate beverage that underwent an initial preheating step of 65°C. Although sensory characteristics were not specific experimental parameters in this project, it was noted that this beverage lacked the pleasant acidic flavour associated with fermentation, as reflected by the pH values shown in Table 4.2.

Based on the results of the viscosity and stability measurements of the beverage samples produced by the various technological approaches, the beverage formulation chosen for further study involved the dilution of set yogurt with unprocessed cottage cheese whey. This product formulation was selected based on the stability of the final product and the fact that the use of unprocessed cottage cheese whey resulted in a more feasible end product in terms of time and economics (as compared to the retentate-based product).

Further examination of the whey-based yogurt beverage selected from this experiment included studying the rheological properties, particle size distribution, and pulse nuclear magnetic resonance measurements of the product in relation to its visual stability over four weeks. These tests were completed to determine if these sophisticated analytical methods could be used as reliable alternatives for predicting the shelf stability of such a product.

4.2. AN EXAMINATION OF THE RELATIONSHIP BETWEEN THE RHEOLOGICAL CHARACTERISTICS AND STABILITY OF A WHEY-BASED YOGURT BEVERAGE

One quality parameter associated with the acceptability of a product is its shelf stability. The second objective of this research project was to determine if the measurement of physical properties by advanced methods such as strain sweep and high frequency - high amplitude oscillation for viscosity and dynamic rheology; particle size determination; or pulse nuclear magnetic resonance could reflect or predict the stability of a whey-based yogurt beverage more accurately than a simple gravitational separation test. The advantage of using these methods, over a long term shelf test would be the rapid assessment of stability immediately following production rather than after the long period of time necessary for a reliable shelf life evaluation.

Centrifugal sedimentation may be another method used for evaluating the shelf stability of a product such as a whey-based yogurt beverage. However, in some instances, this method may not provide an entirely accurate profile of a solution's physical stability.

The centrifugal sedimentation test alone may not be a reliable indication of stability depending upon the time, force, and speed of centrifugation. For example, under a very high centrifugal force, casein micelles may be physically separated from solution in a sample containing milk. In a case such as this, a measure of the amount of centrifugal sedimentation may not accurately reflect the stability of the sample. Because of the difficulty in equating the conditions of a centrifugal sedimentation test to the conditions of a gravitational sedimentation test, this method was not employed as an experimental parameter in this study.

In this study, the physical characteristics of the formulated whey-based yogurt beverage chosen from section 4.1 were analysed. The effects of pasteurization temperature and stabilizer concentration on the stability and physical properties of the beverage were examined with advanced technological equipment and compared to the results of the visual sedimentation test used as an indication of stability.

The experimental portion of this section was carried out over a limited period of time at the Grindsted Products A/S research facility in Brabrand, Denmark. The analytical procedures used were chosen based on availability and the resources of the Grindsted company.

4.2.1. Visual Stability

The results of the visual stability test shown in Table 4.3 indicate that the average amount of separated whey in the whey-based yogurt beverages decreased with an increasing concentration of pectin. However, from the point of view of a food processor, the minimum acceptable level of stability provided by the minimum amount of added stabilizer is often significant. Based on this fact and the results of the visual stability test, 0.3 percent appears to be the minimum concentration of pectin required to reduce whey separation resulting in an acceptable end product. The results of this test are referred to in each of the following sections as they relate to or reflect the findings of the advanced physical analyses.

Table 4.3, summarizing the average amounts of separated whey recorded from the beverage samples after four weeks of storage at

4.0°C, indicates that there was no appreciable difference in the amount of separated whey between the samples pasteurized at the two temperatures. The results of the stability test shows, however, that at both pasteurization temperatures, there was a statistically significant decrease in the amount of separated whey between the 0.1 and 0.3 percent pectin levels. Samples containing less than 0.3 percent pectin appeared to be unstable, exhibiting gross separation upon standing as early as four hours following processing.

Table 4.3. Visual stability of the whey-based yogurt beverages pasteurized at 78°C and 90°C

% Pectin:	0.0	0.1	0.3	0.4	0.5
		mL Sep*/100mL			
Past. Temp:					
78°C	50	45	3	1	0
			- - - - -	- - - - -	- - - - -
	- - - - -	- - - - -			
90°C	53	48	3.5	2	0
			- - - - -	- - - - -	- - - - -
	- - - - -	- - - - -			

Underlined data sharing a common line are not significantly different (p>0.05).
*Separated whey

These observations were in agreement with the data reported by Towler (1984) who found that increasing the level of stabilizer to a point where the viscosity was not at a minimum, decreased sedimentation. These findings also coincided with the recommendations of Barfod *et al.* (1990) that not less than 0.4 percent stabilizer be added to a cultured milk beverage to reduce the casein aggregate size to achieve product stability.

Cultured milk beverages which appear to be stable at pectin concentrations of 0.3 percent and greater, theoretically achieve their stability by the binding of the negatively charged polysaccharide stabilizer to the positively charged casein particles at the pH of the yogurt beverage (4.1) which is below the isoelectric point of casein (4.6) (Gregory, 1986). In these beverage samples, where both casein and whey proteins were present, the action of the stabilizer may be similar, however, the exact mechanism of stabilization is not known.

4.2.2. Viscosity: Strain Sweep

Figure 4.1 illustrates a comparison of the viscosities of the whey-based yogurt beverages pasteurized at the two different temperatures. This Figure indicates that at all levels of added pectin, there was no significant difference in viscosity between the samples pasteurized at the two different temperatures, the only

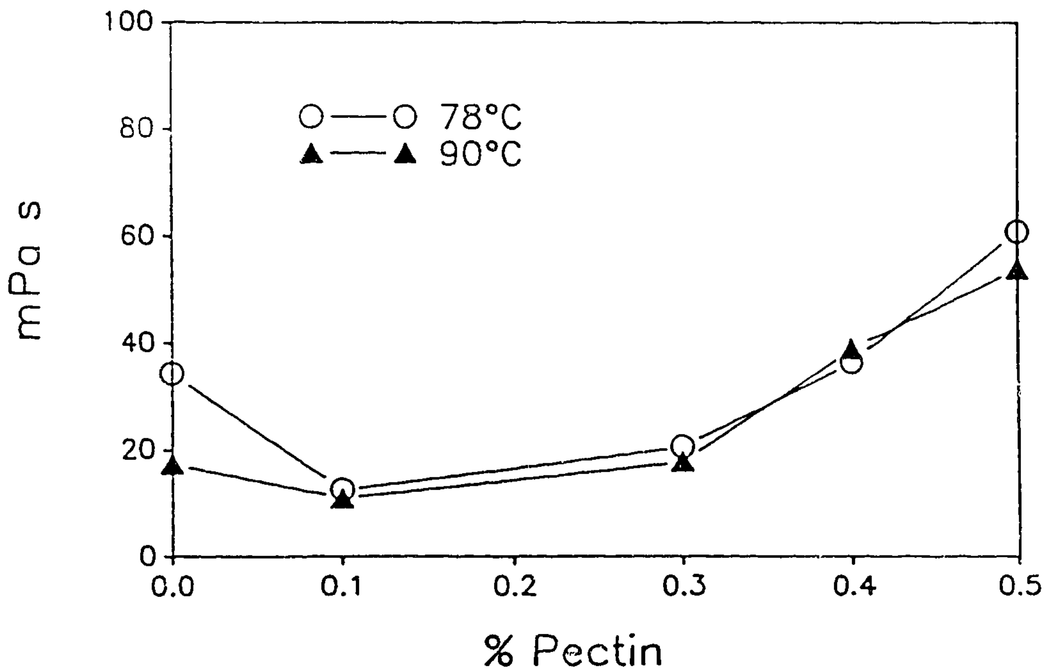

Figure 4.1. Viscosity of whey-based yogurt beverages pasteurized at 78°C and 90°C (Strain Sweep).

exception being the samples containing no pectin. This indicates that, at all concentrations of added pectin, the samples pasteurized at either temperature were equally stable. This finding was supported by the results of the visual stability test which showed that there was no significant difference in the amount of separated whey between the samples pasteurized at different temperatures.

It was observed that, as the concentration of pectin was increased from 0.0 to 0.1 percent, the viscosity dropped slightly in both sets of samples. As the concentration was increased to 0.3 percent, the viscosity rose slightly. At stabilizer concentrations greater than 0.3 percent, the viscosity rose sharply and significantly ($p \leq 0.05$). In this study and in those completed by Glahn (1982), Towler (1984), and Gregory (1986) a rise in viscosity at stabilizer concentrations of 0.3 percent and greater was observed. This indicated a possible saturation of the surface of the protein aggregates with stabilizer suggesting that 0.3 - 0.4 percent was the minimum concentration of pectin required for beverage stability with the increase in viscosity at higher concentrations being attributed to the presence of unbound pectin.

4.2.3. Viscosity: High Frequency - High Amplitude Oscillation

The results of the high frequency - high amplitude oscillation test did not provide any new information regarding the relationship between the viscosity of the whey-based yogurt beverages and product stability. The viscosity pattern obtained from the oscillation test shown in Figure 4.2 was similar in trend to that shown by the strain sweep test in that there was no significant difference in viscosity between the samples heated at the two temperatures of pasteurization.

As the level of stabilizer was increased, the viscosity increased gradually until the 0.3 percent level when additional stabilizer caused the viscosity to increase sharply and significantly ($p \leq 0.05$). Like the strain sweep results, this suggested that the

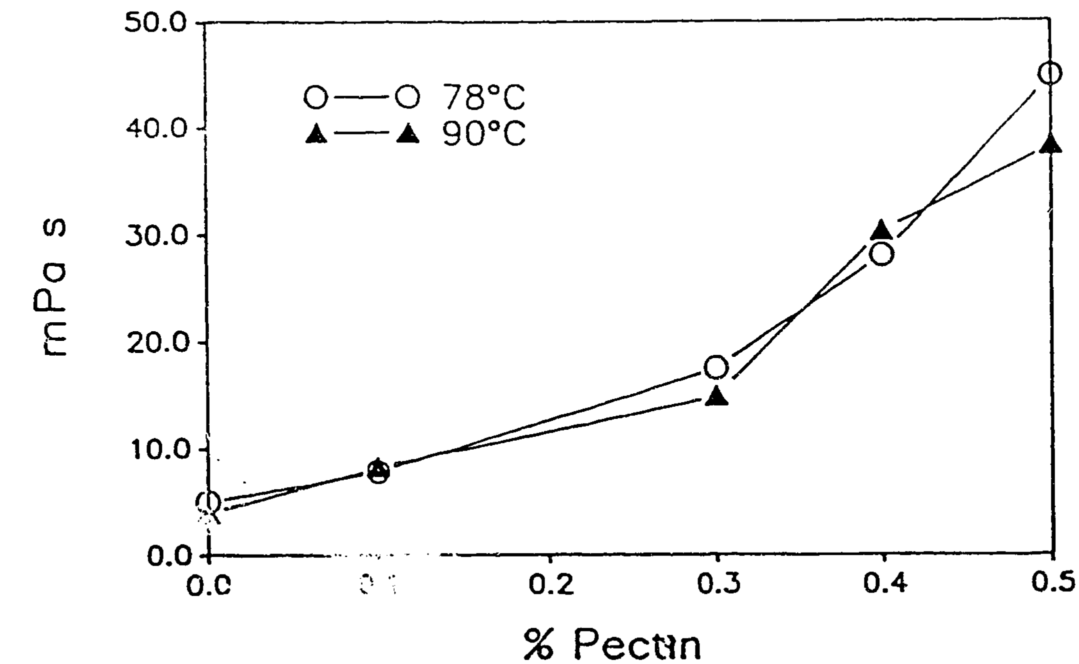

Figure 4.2. Viscosity of whey-based yogurt beverages pasteurized at 78°C and 90°C (Oscillation Test).

protein aggregates may have been saturated with pectin after the 0.3 percent concentration and the sharp viscosity increase, a reflection of the viscosity of the free pectin in the sample. However, this finding, as in the strain sweep results, was not supported by the findings of the visual stability test which showed no significant difference between the samples containing 0.3 and 0.5 percent pectin. Consequently, the values of the high frequency - high amplitude oscillation test did not positively correlate to the values obtained from the visual stability test at either temperature of pasteurization.

4.2.4. Dynamic Rheology: Strain Sweep

In this study, the structural parameters of the whey-based yogurt beverages were studied as they were influenced by the concentration of stabilizer, based on the elastic (G') and viscous (G") moduli of the rheological profile.

Figures 4.3 and 4.4 show that at all levels of added pectin, the structure of the beverages was more viscous than elastic. In each sample, the viscous and elastic moduli appeared to decrease with the addition of 0.1 percent stabilizer. However, an increase in the pectin concentration to 0.3 percent, resulted in a much more rapid increase in the viscous than the elastic parameter.

At approximately 0.3 percent pectin there appeared to be a significant increase in the viscous parameter of the beverage structure suggesting that, at this concentration, the protein aggregates were saturated with pectin and stability had been achieved. The further increase in sample viscosity at the 0.5 percent level may have been a reflection of the viscosity of the free, hydrated pectin in the system.

Based on the results of the measurements of the viscous and elastic parameters, it appeared that these methods of analysis did not provide a good indication of stability as the results of the measurements of either test did not positively correlate to the results of the visual stability test.

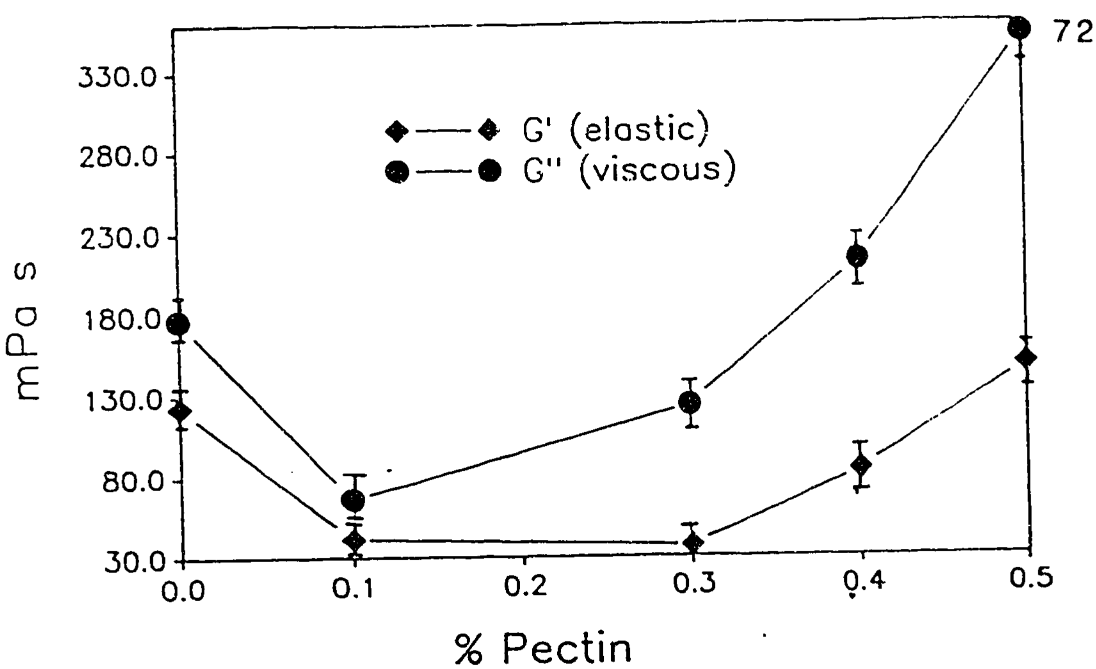

Figure 4.3. Measurement of the viscous and elastic parameters of a whey-based yogurt beverage pasteurized at 78°C (Strain Sweep. Bars indicate 95% confidence limits).

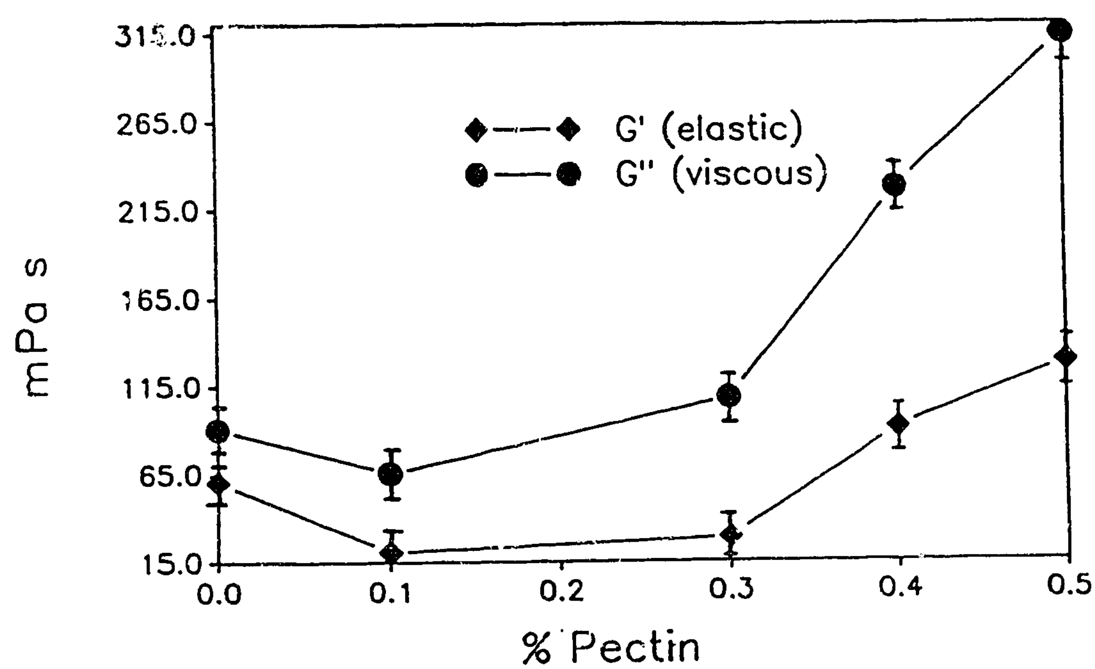

Figure 4.4. Measurement of the viscous and elastic parameters of a whey-based yogurt beverage pasteurized at 90°C (Strain Sweep. Bars indicate 95% confidence limits).

The results of this experiment showed that there was a significant difference in the values of both G' and G" between the samples pasteurized at the two different temperatures. This finding was in agreement with the results of Foley and Mulcahy (1989) who reported that an increase in pasteurization temperature from 70°C to 80°C or 90°C decreased the final viscosity of drinking yogurt. The basis of this report suggested that the higher temperature of pasteurization may have weakened the structure of the yogurt gel during processing or may have further denatured the whey proteins. These findings reflect the measurements of the elastic and viscous parameters but not the results of the viscosity test of the whey-based yogurt beverages which indicated that there was no significant difference ($p>0.05$) between the samples pasteurized at the two different temperatures.

4.2.5. Dynamic Rheology: High Frequency - High Amplitude Oscillation

The high frequency - high amplitude oscillation test used to further analyse the physical structure of the whey-based yogurt beverages indicated some discrepancy between the results of this test and the results of the strain sweep and visual stability tests.

The results provided by the oscillation test shown in Figures 4.5 and 4.6 regarding the dynamic rheology of the beverage samples showed that the relative contributions of the elastic (G') and viscous (G") moduli changed as the concentration of stabilizer was increased. At low pectin concentrations and at both temperatures of pasteurization, the elastic modulus was more dominant, however, this changed at approximately the 0.3 percent level at which point both properties contributed equally. At high concentrations of pectin (0.4 and 0.5 percent), the viscous module became more dominant reflecting the possible presence of free pectin in the system.

These results suggest that at approximately 0.3 percent pectin, the structural make up of the yogurt beverages were equally

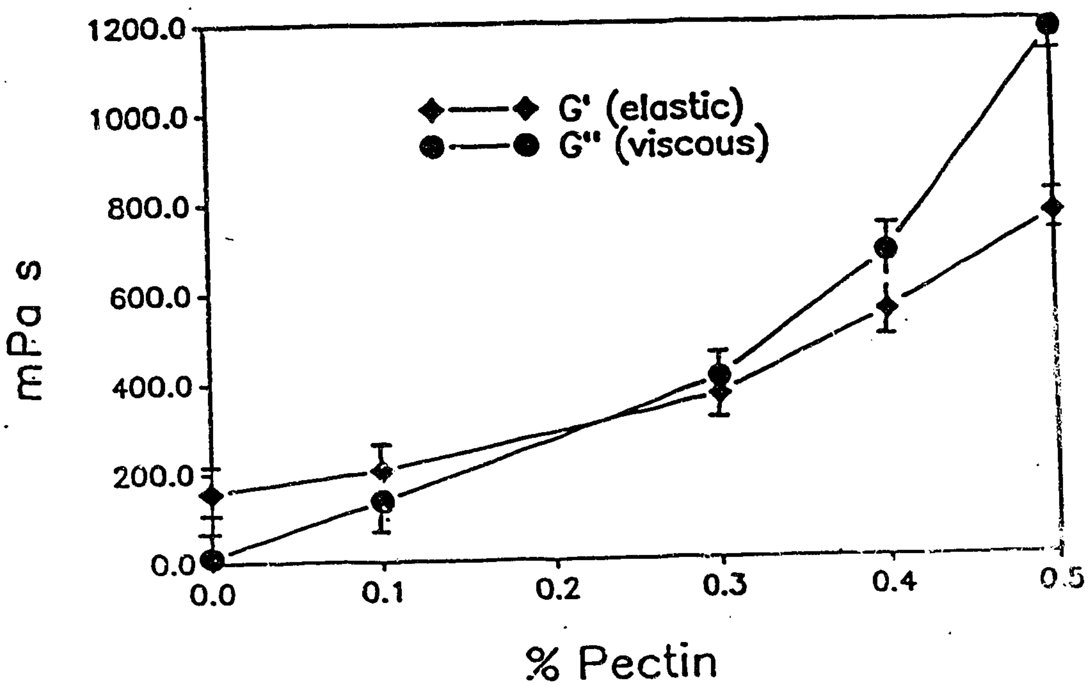

Figure 4.5. Measurement of the viscous and elastic parameters of a whey-based yogurt beverage pasteurized at 78°C (Oscillation Test. Bars indicate 95% con. lim.).

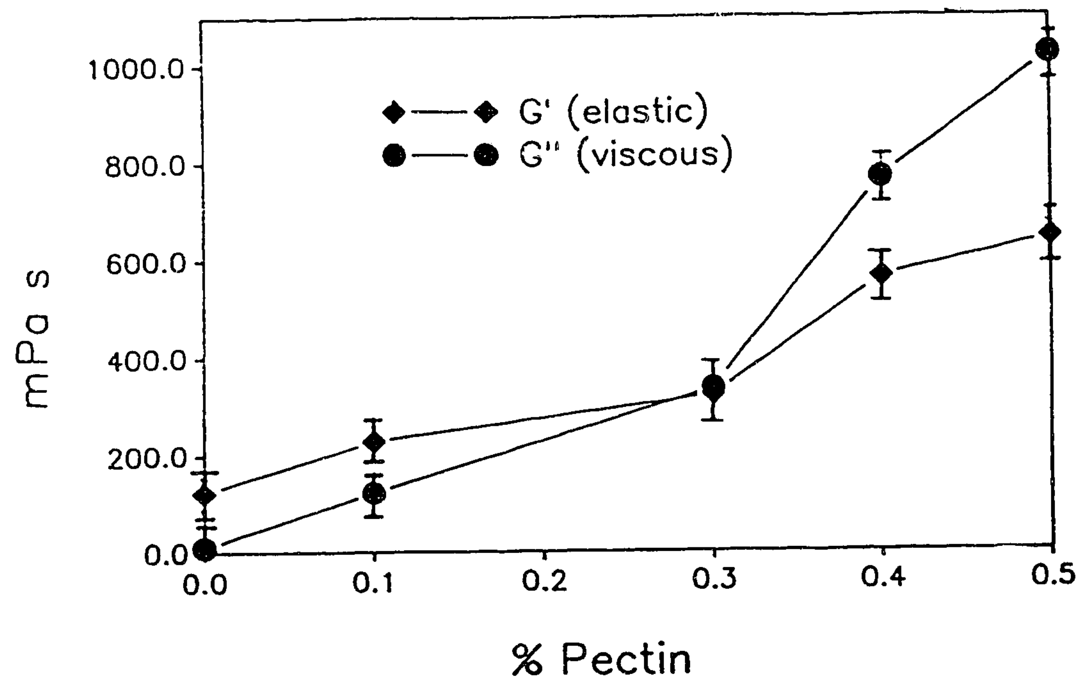

Figure 4.6. Measurement of the viscous and elastic parameters of a whey-based yogurt beverage pasteurized at 90°C (Oscillation Test. Bars indicate 95% con. lim.).

affected by the elastic and viscous moduli. Based on the results of Barfod *et al.* (1990) this phenomenon signifies a possible point of stabilization in a cultured milk beverage. However, because this trend was not reflected in the strain sweep test and the results of the oscillation test did not positively correlate with the results of the visual stability test; this analytical method and the strain sweep test may not suitably reflect the stability of the whey-based yogurt beverage.

4.2.6. Particle Size Distribution

The determination of the average size of the particles suspended in a heterogeneous system may provide a general overview of its stability (Irani and Callis, 1963). In a cultured milk product, such as a yogurt beverage, casein micelles may agglomerate and form larger particles which tend to sediment and separate upon standing (Gregory, 1986). Because smaller particles tend to remain in solution longer, a solution consisting of small particles should be more stable than one consisting of large particles. However, the velocity of sedimentation also depends upon the viscosity of the continuous phase of the suspension and the density of the particles, as some large sized particles may be completely stable and not exhibit sedimentation if their densities are similar to the density of the aqueous phase of the solution.

The theory of particle separation is based on Stokes' Law which infers that the velocity with which a particle separates from a heterogeneous system is directly proportional to the square of its radius and the difference in densities between the particle and the continuous phase. This velocity is inversely proportional to the viscosity of the continuous phase of the system. This relationship is given by the equation (Towler, 1986):

$$fr = 6\pi\eta rv$$

where fr = the frictional force inhibiting the separation of the particle

η = the viscosity of the continuous phase
r = the radius of the particle
v = the velocity of separation of the particle from the continuous phase

Theoretically, the determination of the average particle size of a whey-based yogurt beverage, should provide some indication of the product's stability (Barfod *et al.*, 1990).

The results of the particle size analysis of the whey-based yogurt beverage, shown in Figure 4.7, indicate that with an increase in the concentration of pectin, the size of the suspended particles decreased and levelled off at approximately 0.3 - 0.4 percent added pectin regardless of the temperature of pasteurization.

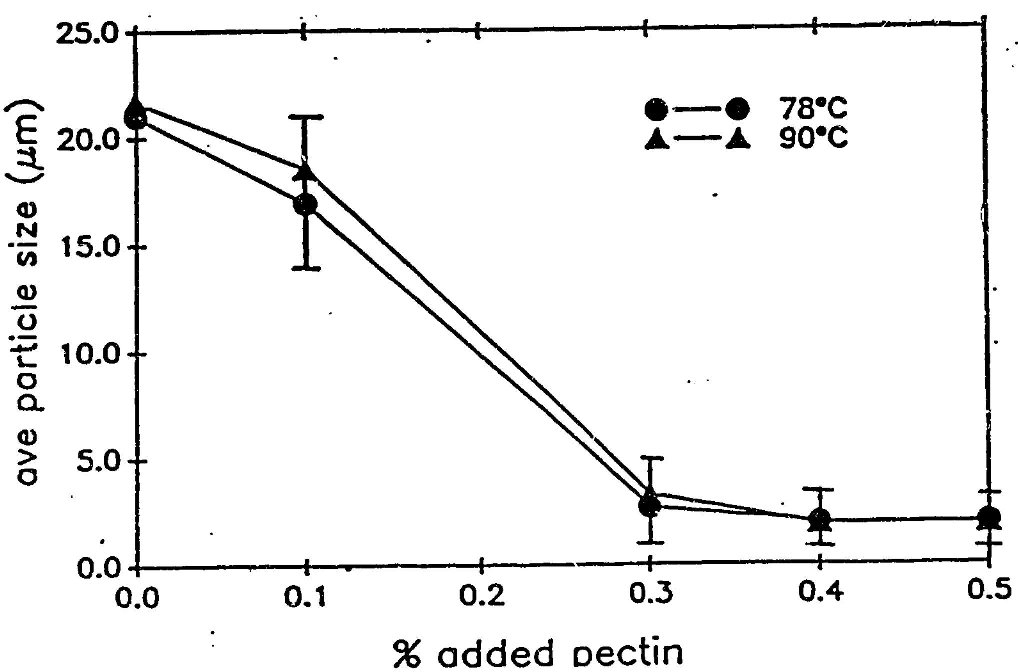

Figure 4.7. Particle size distribution of whey-based yogurt beverages pasteurized at 78°C and 90°C. (Bars indicate 95% con. lim.)

From these results, it appeared that, at a pasteurization temperature of 78°C or 90°C, approximately 0.3 percent pectin was sufficient to produce particles of minimum size with no significant decrease in size at a higher concentration. These findings positively correlated with the results of the visual stability test which showed that significant stabilization occurred at the 0.3 percent level for both 78°C and 90°C pasteurization with no significant increase in stability at the 0.4 or 0.5 percent levels. Figure 4.7 shows that, at the lower pectin levels (0.0 - 0.1 percent), the more severely heated samples appeared to have slightly larger particles although these differences were not statistically significant.

Similar findings regarding the stability of cultured milk beverages were reported by Glahn in 1982 who hypothesized that the relationship between the minimum stabilizer concentration and the particle size (the total surface area of the particle) indicated that the total surface of the particle must have been covered with stabilizer. More specifically, the casein particles must have been saturated with stabilizer and could no longer attract one another to form the larger particles that tend to settle out more readily. Since this trend was similar in beverages heated at both pasteurization temperatures, the results of this study indicated a new finding regarding the interaction of pectin and milk proteins in that the solutions examined in this study were enriched with whey proteins.

The results of tests on these samples containing a greater concentration of whey protein, and on those studied previously which consisted mainly of casein (Towler, 1984) are similar which suggests that the presence of whey proteins had little effect on the action of the polysaccharide stabilizer on the milk proteins present.

4.2.7. Pulse Nuclear Magnetic Resonance

The pulse nuclear magnetic resonance test is a one that is often used in food analyses to determine the level of bound water in a product (Streitwieser and Heathcock, 1981). In a product such as a whey-based yogurt beverage, bound water may be defined as the water that exists in liquid form at sub-freezing temperatures (Hardman, 1986) and a measure of this water may be an indication of the product's stability as unbound water may contribute to syneresis in a product such as a cultured milk beverage (Barfod *et al.*, 1990).

Figure 4.8 illustrates the apparent immobilizing effect of the pectin stabilizer on some of the free water in the whey-based yogurt beverages with respect to the T_2 relaxation time of water protons in the system. This Figure illustrates that, while the T_2 relaxation time continues to decrease after the addition of 0.3 percent pectin this decrease is not significant ($p>0.05$).

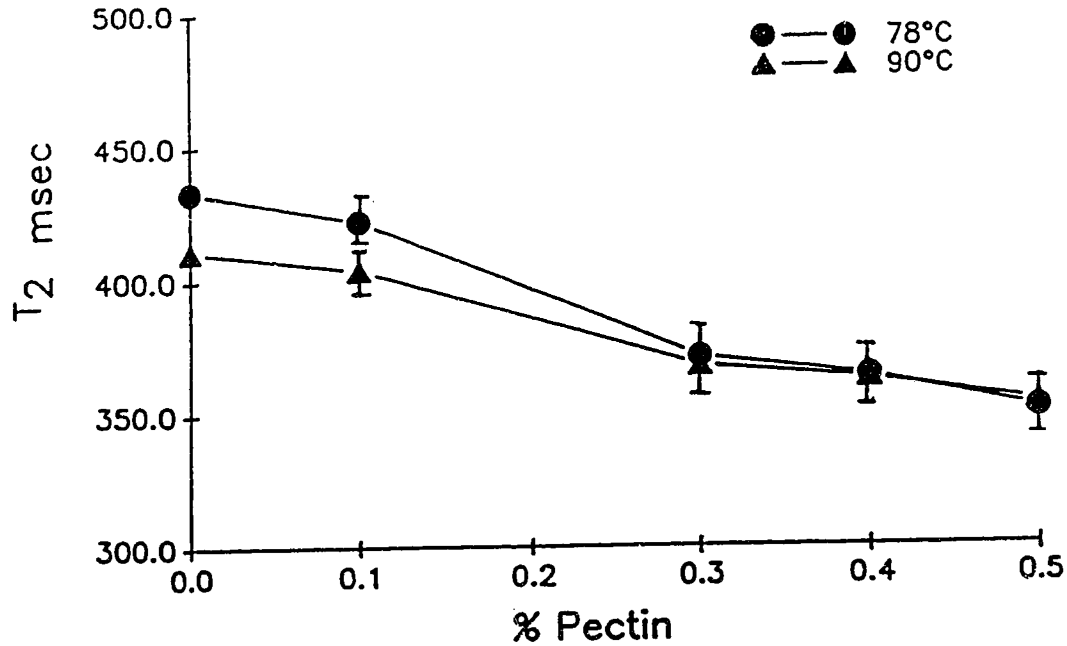

Figure 4.8. T_2 relaxation times of whey-based yogurt beverages pasteurized at 78°C and 90°C. (Bars indicate 95% con. lim.)

The findings of the pNMR test appear to indicate some significant point at the 0.3 percent level of added pectin as the inflection, at this concentration, positively correlates at $p \leq 0.05$ with the results of the visual stability test. These results show that, at 0.3 percent pectin, the product appears to have achieved stability. However, based on the pNMR test alone, these findings would not be entirely conclusive considering the fact that this product contains approximately 85 - 86 percent water and only a small fraction of this could possibly be bound by such an insignificant amount (0.0 - 0.5 percent) of stabilizer.

4.2.8. A Summary of the Comparisons of the Rheological Characteristics and Stability of a Whey-Based Yogurt Beverage

Based on the examination of the relationships between the rheological characteristics, particle size distribution, and T_2 relaxation time and the stability of the whey-based yogurt beverage it appeared that only the results of the particle size distribution gave some questionably accurate indication of the product's shelf stability. However, despite the fact that the results of this test positively correlated with the results of the visual stability test, this method of analysis also had its shortcomings in terms of accuracy as some food systems may actually be shelf stable regardless of the size distribution of their particles. If the suspended particles in a food system are large water-containing aggregates whose densities are near that of the continuous phase, separation of these particles will not readily occur and the system will remain stable upon standing.

The results of the pulse NMR test also positively correlated with the results of the visual stability test of the whey-based yogurt beverage, at 0.3 percent stabilizer, however, the use of T_2 relaxation time as an indication of the stabilizing effect of added pectin was not considered to be reliable due to the large ratio of water to pectin in the samples.

The inconsistencies in the results of the rheological tests of the beverages analysed by the strain sweep and high frequency - high amplitude oscillation tests suggest that the viscosity and dynamic rheology measurements did not provide an accurate indication of the stability of the beverage examined in this section. In addition, the fact that neither the results of the strain sweep test nor the high frequency - high amplitude oscillation test positively correlated to the visual stability test, further supports this conclusion.

The most significant finding of section 4.2 was the fact that 0.3 percent HM pectin stabilized a cultured milk beverage containing a high whey protein to casein ratio.

4.3. THE STABILIZATION OF SIMULATED AND GENUINE WHEY UF RETENTATE-BASED PRODUCTS

In the previous experiments in this research project, an examination of the heat-induced destabilization of whey-based yogurt beverages, containing both casein and whey proteins, was carried out using various technological methods. In this study, the feasibility and acceptability of using the UF retentate of cottage cheese whey as the basis for a cultured milk beverage was examined and included a study of the stabilization of whey proteins in a model solution with HM pectin.

The stability of cultured milk beverages has been examined previously and researchers including Gregory (1986) and Glahn (1982) have proposed a mechanism by which pectin may stabilize the casein micelles in cultured milk products. However, the mechanism of stabilization of a cultured milk beverage containing a high whey protein to casein ratio or a cultured product containing only whey proteins and no casein is much less definite.

In the previous sections of this project the satisfactory stabilization of a cultured milk beverage containing approximately 51 percent whey was achieved, although because of its complexity, the exact mechanism of this stabilization was not studied in detail. In this final section, an investigation of possible approaches to the stabilization, against heat-induced precipitation, of a cultured whey-based beverage containing only whey proteins was carried out.

To determine if the production of a shelf-stable product containing only whey proteins was feasible, an initial study was carried out with a simulated whey UF retentate (SUFR) solution, examining the effect of pH adjustment and the addition of pectin on the heat stability of the solution. An analysis of the visual stability, viscosity, and particle size distribution were used to determine the stability of the simulated whey UF retentate prototype as it was stabilized with increasing amounts of pectin.

4.3.1. Visual Stability of a SUFR Model System

Previous research has demonstrated that HM pectin may be used to stabilize whey proteins againstheat-induced precipitation and that cultured milk systems containing pectin are most stable in the pH range of 3.0 - 4.0 (Gregory, 1986; Towler, 1984). At lower and higher pH values pectin exhibits ß-elimination (hydrolysis) and as a consequence, may be ineffective as a stabilizer of whey proteins at pH levels other than in the narrowly defined region (Barfod, personal communication). The results of this study showed that the heat stability of the whey protein system was affected mainly by the presence of pectin in the pH range of 4.0 - 5.0.

Table 4.4. Average stability and viscosity values for simulated UF retentate solutions adjusted to different levels of pH

pH:		4.0			5.0			6.5	
% Pectin:	0.0	0.3	0.5	0.0	0.3	0.5	0.0	0.3	0.5
Stability: (mL Sep*/100mL) (28 days)	28f	0d	0d	20e	1.5d	.5d	70c	81b	86a
Viscosity (24hr): (mPa.s)	2.9c	7.8d	14.7a	3.0c	7.6d	16.9a	3.4c	4.[illegible]	13.1a

Data sharing a common letter in horizontal rows are not significantly different (p>0.05).
*Separated whey

The effect of the adjustment of pH and added stabilizer on the stability of the SUFR solutions is shown in Table 4.4. At pH 4 the samples containing 0.3 and 0.5 percent pectin appeared to be stable, showing no evidence of whey separation after four weeks of storage at 4.0°C. However, the sample containing 0.0 percent pectin at this pH level, was significantly less stable showing 28 mL of separated whey after 28 days of storage. Likewise, at pH 5, the SUFR solutions were significantly more stable when 0.3 or 0.5 percent pectin was added than when no pectin was added. The solutions adjusted to pH 6.5 were not stabilized by any concentration of added pectin and a substantial amount of whey separation occurred in all samples upon standing.

The addition of pectin to the whey protein model systems appeared to have a positive effect on the visual stability of the solutions, however, this effectiveness was significantly limited at a pH level higher than 5.0. At pH levels 4.0 and 5.0, an addition of the pectin stabilizer significantly decreased the amount of whey separation that occurred in the samples. At pH 6.5 the presence of stabilizer did not reduce the amount of whey separation and, in fact, appeared to enhance it. These findings are in agreement with previous literature reviews which reported that pectin was inactive as a stabilizer at higher pH levels because it becomes negatively charged and unable to interact with the negatively charged milk protein complexes (Anon, 1982; Gregory, 1986).

4.3.2. Viscosity of a SUFR Model System

Table 4.4 indicates that at all levels of pH, the viscosity of the SUFR solutions increased as the concentration of stabilizer was increased with the greatest viscosity apparent in the samples containing 0.5 percent pectin.

From these results, it also appeared that at 0.0 percent pectin, the viscosity of the solutions increased slightly but not significantly with increasing pH. At the 0.3 percent level of added pectin, the viscosity decreased greatly between pH levels 5 and 6.5. This decrease may have been a result of the ineffectiveness of pectin as a stabilizer at this pH; the result of pectin hydrolysis and the lack of binding of the stabilizer to the protein aggregates. At a pectin concentration of 0.5 percent, the viscosity of the beverage samples seemed to be unaffected by an increase in pH, indicating that this concentration of pectin was greater than what was required to stabilize the whey protein aggregates. Free pectin may have been present in the solutions at all levels of pH. As a result, no significant difference in the viscosities of these samples at the different levels of pH was noted.

The viscosity trends of the solutions adjusted to pH 6.5 reflected the trend of the stabilities (shown in Table 4.4) in that the higher the viscosity of a sample, the lower is its stability. However, this contradicts both the results of the stability/viscosity relationship indicated at pH levels 4 and 5, the results of section 4.2 of this project which indicated that product stability increases with an increase in viscosity, and the findings of previous researchers involving the stabilization of cultured milk beverages (Towler, 1984; Glahn, 1982). These contradictions further confirm one of the previous indications from this study, that viscosity measurements alone may not provide a good indication of the stability of the whey-based cultured milk beverages.

The above discrepancies imply that the stabilization of whey proteins in a solution against heat-induced separation could be much more complex than the stabilization of caseins. The addition of pectin to the whey protein model solutions at concentrations of 0.3 percent and greater appeared to stabilize the solutions, however, the stabilization by pectin appeared to be ineffective at pH 6.5.

4.3.3. Particle Size Distribution of a SUFR Model System

The results of the particle size analysis shown in Figure 4.9 closely resemble the results found in the visual stability test in the whey protein model systems at pH levels 4 and 6.5.

A statistical analysis of the particle size distribution of the samples adjusted to pH 5, indicates that there was no significant difference between the samples containing 0.0 and 0.3 percent pectin. This suggests that the relative stabilities of these samples should also be similar. However, the results of the visual stability test, shown in Table 4.4, indicate that the samples containing 0.0 percent pectin were significantly less stable than those containing 0.3 percent at this pH. However, there was a significant difference in the viscosity measurements of these samples (Table 4.4), which

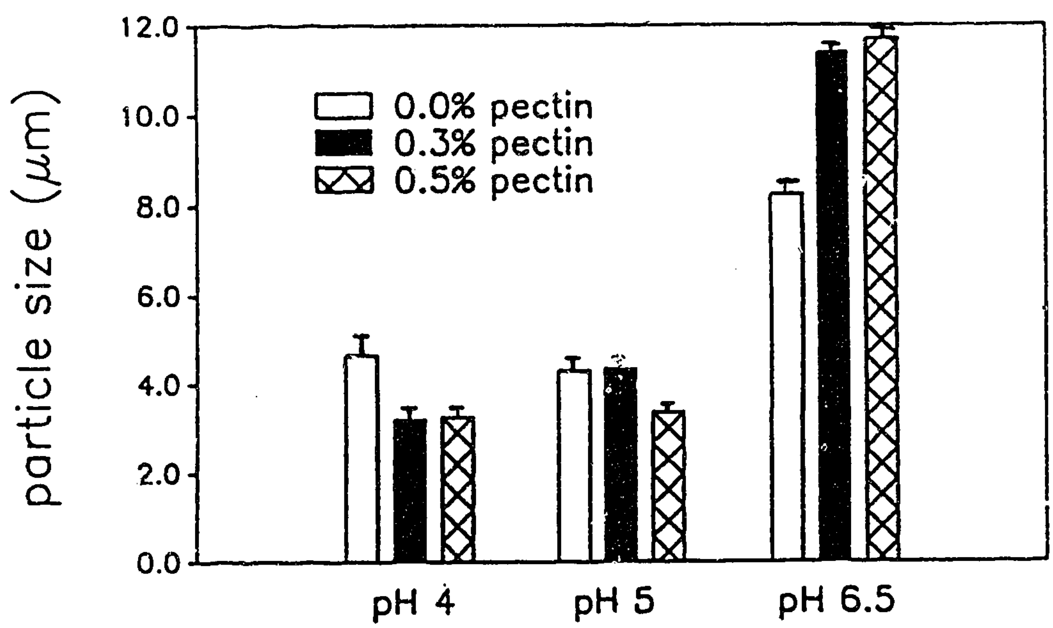

Figure 4.9. Particle size distribution of simulated UF retentate solutions adjusted to various levels of pH and pectin. (Bars indicate 95% confidence limits)

may have accounted for the greater stability in the sample containing 0.3 percent pectin. The results of the particle size determination showed that there was a significant difference ($p \leq 0.05$) between the particle size of the model systems containing 0.3 and 0.5 percent pectin. However, the results of the visual stability test on the same samples showed that there was no significant difference between the two pectin levels, at pH 5.0.

These phenomena may be an illustration of what was suggested previously regarding the stability of large, water-containing aggregates. This indicates that more research is needed to develop a method suitable for predicting a relationship between the particle size test and the results of a visual stability test in whey protein solutions. Perhaps both particle size and viscosity may be important analytical methods useful for assessing the shelf stability of whey protein-containing cultured milk products.

Based on the results of this study the optimum method of stabilization of whey proteins in a model solution containing only whey proteins and no casein appeared to be the adjustment of pH to 4.0 and the addition of no less than 0.3 percent pectin. The best method of examining the stability of a product produced in this manner appeared to be the standard visual stability test rather than an analysis of the size distribution of the suspended particles.

4.3.4. The Stabilization of a Genuine Whey UF Retentate-Based Beverage

In this experiment, whey protein-containing beverage prototypes were produced using the UF retentate of genuine cottage cheese whey (consisting of 13.1% total solids and 3.0% protein) and stabilized with various amounts of added pectin. Table 4.5 summarizes the results of the viscosity and stability tests carried out on these samples (all adjusted to pH 4.0) which contained 0.0, 0.3, and 0.5 percent added pectin.

Table 4.5. Average stability and viscosity values for genuine UF retentate-based beverages

Pasteurization Temp.:	78°C			90°C		
% Pectin:	0.0	0.3	0.5	0.0	0.3	0.5
Stability: (mL Sep*/100mL) (28 days)	8c	0a	0a	22b	0a	0a
Viscosity: (mPa.s)	6.05d	17.4c	44.3b	12.1c	43.9b	63.4a

Data sharing a common letter in horizontal rows are not significantly different (p>0.05)
*Separated whey

4.3.5. Visual Stability of a UF Retentate-Based Beverage

These preparations, consisting of whey proteins, pectin, sugar, and yogurt culture, appeared to be stable against the heat-induced separation of the whey proteins during pasteurization. Evidence of whey separation occurred only in those samples containing no added pectin. In the samples produced without pectin, there was evidence of whey separation at both temperatures of pasteurization, with a significantly higher ($p \leq 0.05$) amount of separation being noted in the samples subjected to the more severe heat treatment of 90°C; a finding that is in accordance with the known instability of heated whey proteins above pH 3.9 (Jelen *et al.*, 1987b).

With an increased concentration of pectin from 0.0 to 0.3 percent, no amount of whey separation was evident in the samples regardless of the temperature of pasteurization. When the concentration of pectin was increased from 0.3 to 0.5 percent, no change in visual stability was noted in the samples pasteurized at either temperature as none of these samples exhibited gravitational separation after 28 days of storage at 4.0°C.

4.3.6. Viscosity of a UF Retentate-Based Beverage

The findings of the visual stability test were reflected somewhat in the results of the viscosity test as the samples containing 0.3 and 0.5 percent pectin had significantly higher viscosity values than the samples containing 0.0 percent pectin at either pasteurization temperature. However, unlike the results of the visual stability test, there was a significant difference ($p \leq 0.05$) between the viscosities of the samples that were produced with 0.3 and 0.5 percent pectin as well. The samples pasteurized at the higher temperature showed significantly higher viscosity values at all levels of added pectin.

The findings in the study of stabilization of a beverage based on genuine whey UF retentate indicate that at a pectin concentration of 0.3 percent the whey proteins in this project were stabilized against heat-induced separation by the presence of added pectin. This indicates that the whey proteins may be affected by the mechanism of stabilization of the pectin in solution.

Based on the results of the visual stability test, it appears that, again like the casein-containing beverages, 0.3 percent added pectin was sufficient to decrease whey separation in the model solutions. The mechanism of action of the stabilizer is hypothesized to be either the adequate stabilization of the whey proteins against heat-induced whey separation or the increase in viscosity of the model solutions by the pectin causing the heat-denatured whey proteins to remain in suspension.

The higher viscosity of the samples containing 0.5 percent pectin may simply have been a reflection of the viscosity of the free pectin in the system, assuming that the pectin was actually bound to the whey proteins.

5. SUMMARY OF RESEARCH FINDINGS, CONCLUSIONS, AND RECOMMENDATIONS FOR FURTHER RESEARCH

5.1. SUMMARY OF RESEARCH FINDINGS AND CONCLUSIONS

5.1.1. Technological Approaches in the Development of a Whey-Based Yogurt Beverage.

In this project, the stability and viscosity parameters of yogurt beverages produced by various technological approaches were examined and compared to the stability and viscosity characteristics of a commercial yogurt beverage. The manufacture of two of these products included the use of unprocessed cottage cheese whey, three included the use of UF whey retentate of cottage cheese, and one, containing no whey, was regarded as a control.

In the production of the whey-based yogurt beverages, the use of either unprocessed cottage cheese whey or the UF retentate of cottage cheese whey was considered desirable, in terms of time and cost efficiency, if they were used as an ingredient in the milk base for the production of a yogurt-like beverage product. However, it appeared that due to the heat sensitivity of the whey proteins, a more stable beverage was produced when the whey was used as an ingredient in the final product. The beverages produced with whey or UF whey retentate as a yogurt dilutant were found to be significantly more stable than those products made with whey or UF whey retentate added to the yogurt milk base prior to the high heat treatment; the latter of these products being generally not acceptable in terms of texture and appearance. The beverages produced with whey or the UF retentate of whey as a dilutant for the set yogurt were more stable even after a severe (90°C) heat treatment, possibly because the whey protein mixture was heated in the presence of the casein-containing yogurt gel. For this reason, these products should be able to undergo a mild pasteurization step without destabilization of the whey proteins.

Shah and Jelen (1987) examined the use of the UF retentate of cottage cheese whey in the production of yogurt. They reported some success in the production of a consistent and stable yogurt gel with the use of 11 - 20 percent UF retentate as an ingredient in the yogurt milk base. One experiment in this project involved the fermentation of a mixture consisting of 92 percent UF retentate, five percent skim milk powder, and three percent yogurt culture with the resulting yogurt-like product having very poor consistency and stability. This finding suggests that using UF retentate as an ingredient in the yogurt milk base for a cultured product may be acceptable but at a concentration of less than 92 percent.

It is possible to conclude from the results of this study that of the six technological approaches examined in the development of a whey-based yogurt beverage, the most suitable beverage formulation, in terms of stability and consistency and which would presumably have the greatest possibility of success on the consumer market, was the approach involving the dilution of set yogurt with unprocessed cottage cheese whey.

5.1.2. An Examination of the Relationship Between the Rheological Characteristics and Stability of a Whey-Based Yogurt Beverage

In these experiments, the physical characteristics of whey-based yogurt beverages containing various concentrations of HM pectin and pasteurized at two different temperatures (78°C and 90°C), were analysed by advanced physical methods. These results were then compared to the results of a gravitational stability test. The results of these analyses indicated that, of the advanced methods, including rheological examinations by strain sweep and high frequency - high amplitude oscillation, particle size distribution, and pulse nuclear magnetic resonance testing, the most acccurate in terms of reflecting the results of the visual stability test was the particle size determination. However, the reliability of this technique may be uncertain with solutions consisting of large, well hydrated particles.

The temperature of pasteurization did not appear to significantly affect the size of the suspended particles or the level of whey separation in the samples and it did not significantly affect the viscosity of the samples at any level of added pectin. However, the viscous and elastic parameters of the structural characteristics of the samples were significantly affected by the temperature of pasteurization as the values obtained from the milder heat treated samples were significantly higher at all levels of added pectin.

These findings suggest that some weakening of the yogurt gel may have occurred in this product (Dannenberg and Kessler, 1988; Foley and Mulcahy, 1989) but this was not reflected in a change in particle size distribution nor in the stability of the beverage samples tested in this experiment. The inconsistencies between the results of the viscosity and rheological parameters and those of the visual stability test suggest that these methods of analyses do not reliably reflect or predict the stability of the whey-based yogurt beverages.

Based on the incidence of the various "kinks" in the experimental test curves (including the viscosity, dynamic rheology, and T_2 relaxation time curves), product stability appeared to be achieved at a concentration of at least 0.3 percent added HM pectin; a concentration recommended by previous researchers including Glahn (1980), Towler (1984), and Barfod *et al.* (1990) for casein-based products. Furthermore, considering the expense involved with a higher heat treatment, 78°C or lower is recommended as the temperature of pasteurization for this product.

5.1.3. Study of the Stabilization of Simulated and Genuine Whey UF Retentate-Based Products

Stabilizing whey proteins in a solution intended for the production of a cultured beverage is difficult as they are extremely sensitive to heat at and above temperatures normally used in the pasteurization of dairy products (Evans and Gordon, 1980). Based on the results of this study, the optimum method of stabilization against the heat-induced separation of the whey proteins in solution was shown to be the adjustment of pH to 4.0. However, based on the results of previous research (Patocka *et al.*, 1986; Jelen *et al.*, 1987b), the suggested transitory range below which the whey proteins exhibit maximum stability against heat-induced precipitation is 3.7 - 3.9. For the scope of this research, including an attempt to simulate a consumer accepted product with desirable flavour attributes, the addition of at least 0.3 percent pectin and an adjustment of pH to 4.0 was found to sufficiently reduce the occurrence of whey separation upon heating.

The use of the acidic hydrocolloid, HM pectin, as a stabilizing agent appeared to have some effect on the whey proteins in solution, however, its effectiveness was severely limited to pH levels at or below 5.0; a finding which confirms previous reports that pectin is less effective as a stabilizer of cultured milk products at pH levels above 4.5 (Anon, 1982). For this reason, the effect of all experimental levels of added pectin on the stability of the whey protein solutions adjusted to pH 6.5 was significantly less than that of the solutions adjusted to pH levels 4.0 and 5.0.

Unlike the results of one experiment whereby the use of the particle size analysis was found to provide a fairly accurate indication of the stability of the whey-based, casein-containing yogurt beverages, the use of this analytical method for the prediction of the stability of the heated whey protein solutions did not correlate well with the observed gravitational stability results. For this reason, the use of particle size distribution of a SUFR-based model system for the purpose of predicting the long life stability of the product cannot be recommended, however, the use of this method of analysis for the use of predicting the stability of casein-containing cultured milk beverages, in conjunction with a visual stability test, may be satisfactory.

In one section of this project, which examined whey-based yogurt beverages containing casein, no significant difference ($p \geq 0.05$) was evident between the viscosity values of the samples pasteurized at the two different temperatures. Unlike these results, the findings in the examination of a genuine whey UF retentate - based beverage indicated that this type of product, containing only whey proteins, exhibited significantly higher viscosity values at all levels of added pectin when they were pasteurized at the higher temperature.

The higher viscosities in the pectin-containing samples pasteurized at 90°C suggest that, unlike the structure of the casein-containing yogurt gel which can be weakened by a more severe heat treatment (Foley and Mulcahy, 1989) perhaps the structure of the whey protein - pectin aggregates, was not weakened but actually enhanced by the more severe heat treatment.

5.2. RECOMMENDATIONS FOR FURTHER RESEARCH

The results of the study involving the use of unprocessed, fluid cottage cheese whey or the UF retentate of cottage cheese whey as an ingredient in a yogurt-type beverage indicated that production of this type of product was possible, but it appeared that these ingredients should be blended with the set yogurt in the beverage formulation to achieve maximum stability during processing.

In the production of a set yogurt intended for use in the manufacture of a yogurt beverage, the use of cottage cheese whey or the UF retentate of cottage cheese whey as an ingredient in the yogurt milk base may be economically feasible but not in the concentrations used in this study. Further research is recommended based on the results of this research and on that of Shah and Jelen

(1987) and Jelen *et al.* (1987b) to determine the optimum amount of cottage cheese whey or UF retentate which, when used in the production of yogurt, would result in the production of a cultured milk beverage that has good gel consistency and little whey syneresis.

Further research is also recommended based on using sweet whey or a combination of sweet and acid wheys in the production of cultured milk beverages. Previous research by Park and Lund (1984) has shown that sweet whey is stable upon heating at 90°C for thirty minutes in the pH range of 5.0 - 7.0. Work completed by Jelen *et al.* (1987b) showed that the use of sweet whey in the production of yogurt resulted in a heat stable product that had good gel consistency.

The results of the second section of this research project indicated that an analysis of the particle size distribution of a whey-based yogurt beverage, containing both casein and whey proteins, provided a rapid and fairly accurate assessment of its stability. Based on the results of the third section of this project, it was shown that this method of analysis was not suitable in terms of accuracy for determining the stability of a solution containing purely whey proteins as the results of this analysis were not reflected by the results of the visual stability test. For this reason, further research is recommended to develop a rapid and reliable method for the prediction of stability of a whey-based cultured milk product.

The determination of the size of the particles of a heterogeneous system appears to offer an advantageous alternative (in terms of time and shelf space) to the tedious gravitational sedimentation test, therefore, further research is recommended on the study of the exact correlation between the stability of a heated solution containing whey proteins and its particle size distribution. In order to accurately assess the stability of a sample and refine the particle size technique for use in the dairy industry, further research should include an examination of the relationship between the viscosity of the sample and the size of its suspended particles in order to determine the relative densities of the particles and the continuous phase of the system.

The results of this project indicate that an acceptable cultured beverage produced from the UF retentate of unprocessed cottage cheese whey could be developed. It is recommended that the base of this beverage be cultured to lower the initial pH of the product to 4.0 or below where the whey proteins have greater stability upon heating. In addition, it is also recommended that 0.3 - 0.5 percent HM pectin be added for increased stability as well. For economic reasons and for reasons of better stability of the whey proteins, it is recommended that this product be pasteurized at a temperature lower than 90°C.

An investigation of further possible factors in the stabilization of the whey proteins against heat-induced precipitation is recommended. In light of the consumers' new awareness of the health benefits of milk and specifically, calcium containing foods (Rusoff, 1987; Avioli, 1987), a product of this nature would have the benefit of being heat stable and nutritious. Other facets of investigation involving the increased heat stability of whey proteins could include the addition of fat (Trapp and Jelen, 1990) or other protein.

From an industrial and marketing standpoint, further research is recommended based on the interests and preferences of consumers. Specific areas of research should include the effect of ultrahigh temperature (UHT) pasteurization on the stability of such a product, the addition of fruit and flavourings for increased consumer acceptance, and the use of artificial sweetening agents such as aspartame and acesulfame-K. Specific areas of examination should include the effect of these factors on the stability, flavour compatibility of the added sweeteners with the beverage (including aftertaste effects), and the consumer acceptance of the final cultured whey-based beverage. With the market approval of a whey-based yogurt beverage, the emergence of this dynamic, new product could bring renewed life to the currently insipid dairy industry in North America.

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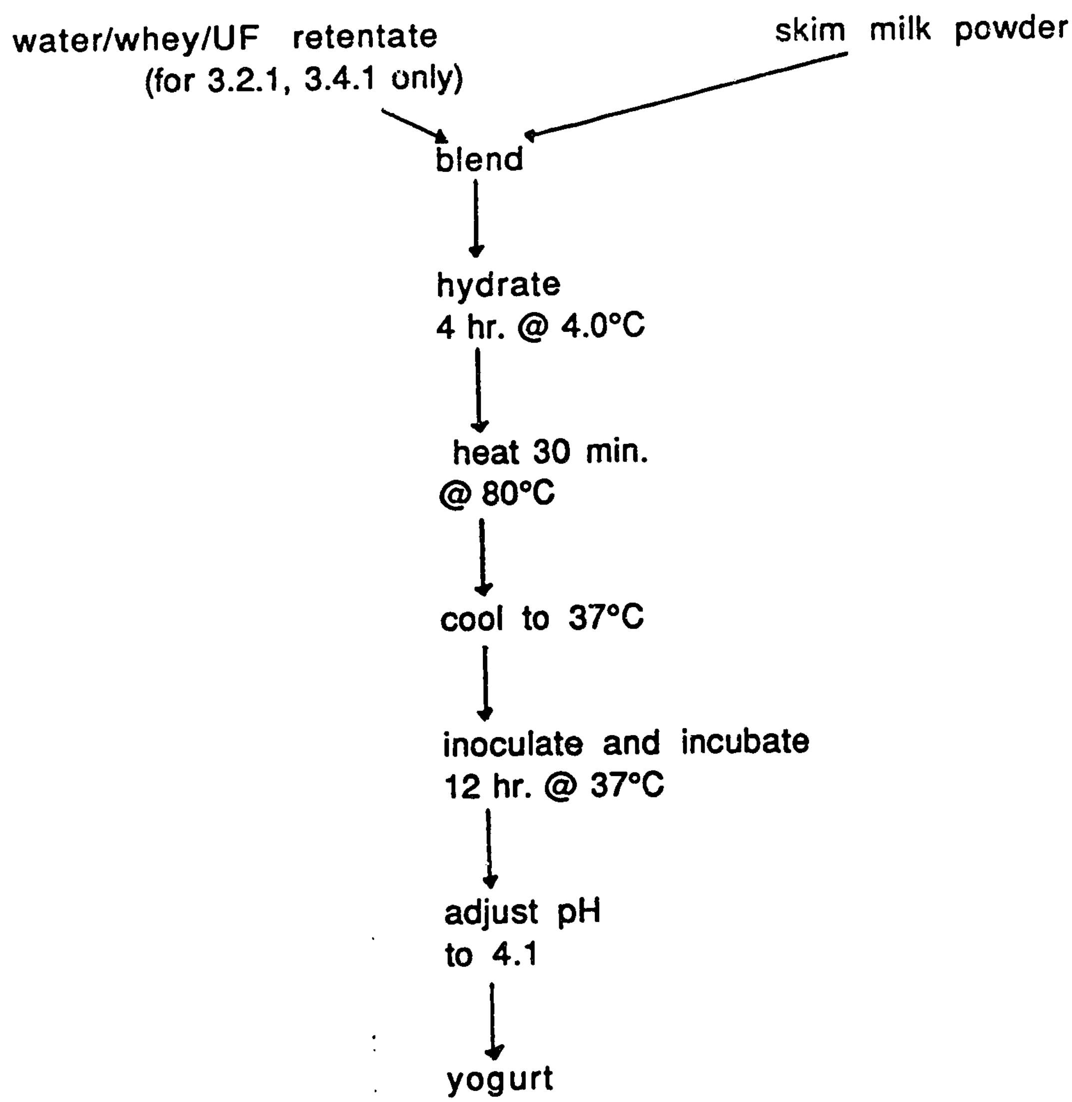
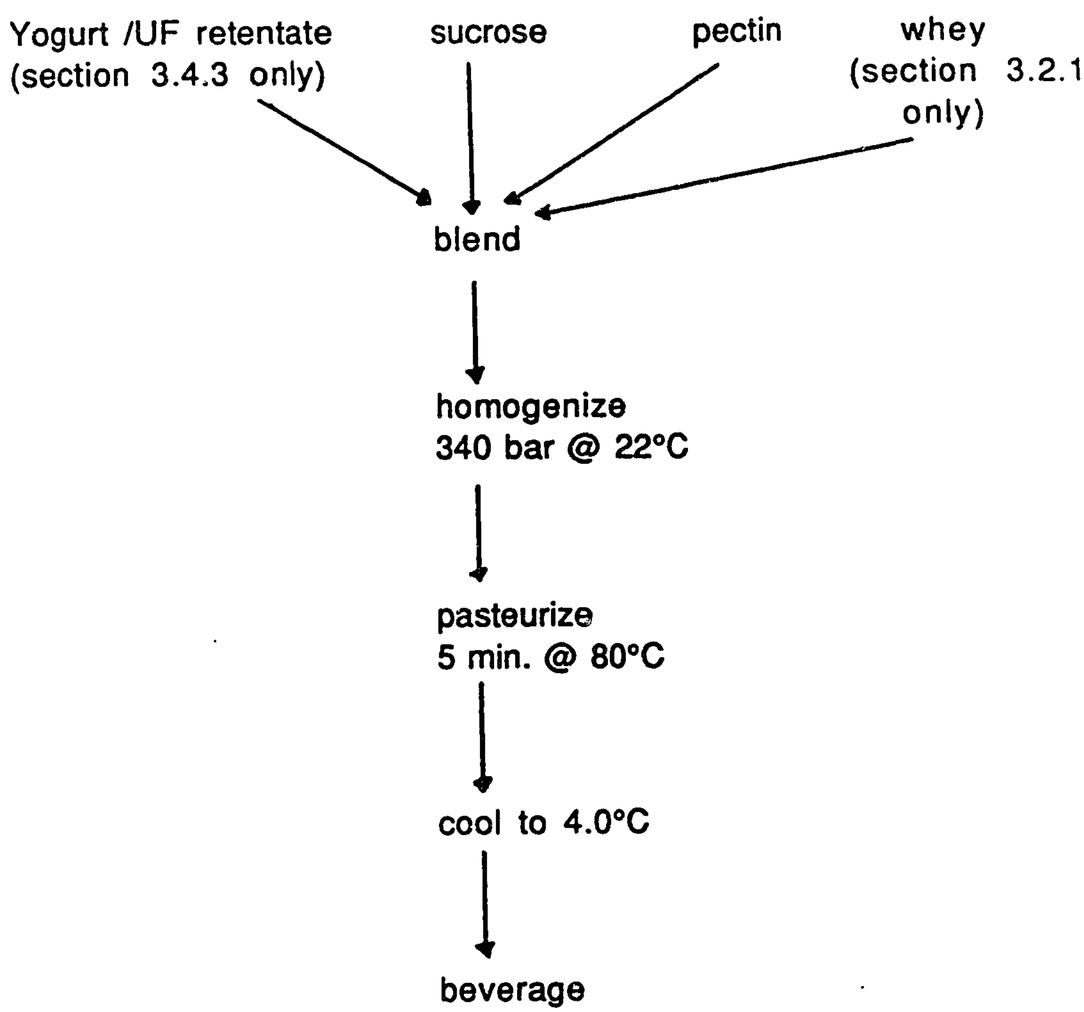
Appendix 1. Flow chart of production method for yogurt outlined in sections 3.2.1, 3.2.2, 3.4.1, and 3.4.2.

YOGURT PRODUCTION:



Appendix 2. Flow chart of production method for yogurt beverages outlined in sections 3.2.1, 3.2.2, 3.4.1, 3.4.2, and 3.4.3.

BEVERAGE PRODUCTION:



Appendix 3. Flow chart of production methods for beverages outlined in section 3.3.

YOGURT PRODUCTION:

water + skim milk powder

↓

blend

↓

hydrate
4 hr. @ 4.0°C

↓

heat 5 min.
@ 90°C

↓

homogenize
200 bar @ 90°C

↓

cool to 37°C

↓

inoculate and incubate
12 hr. @ 37°C

↓

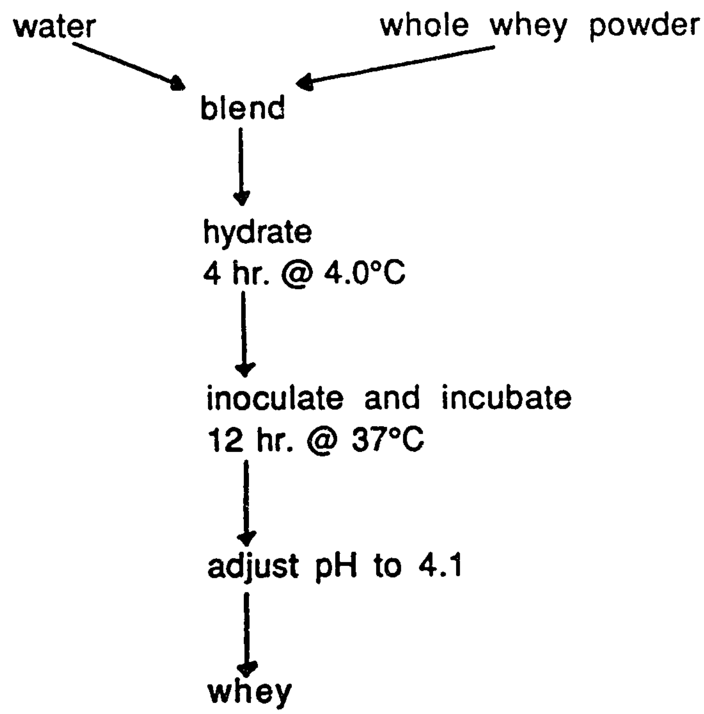
adjust pH to 4.1

↓

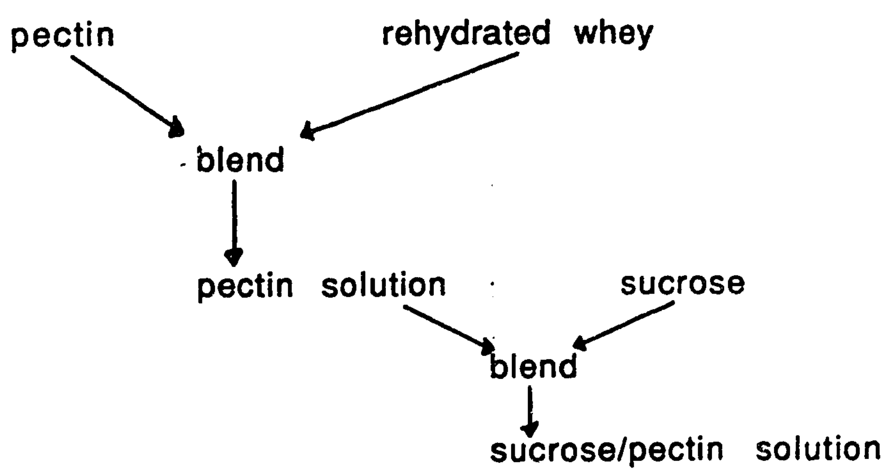
yogurt

Appendix 3. continued: Flow chart for production methods of beverages outlined in section 3.3.

WHEY PRODUCTION:

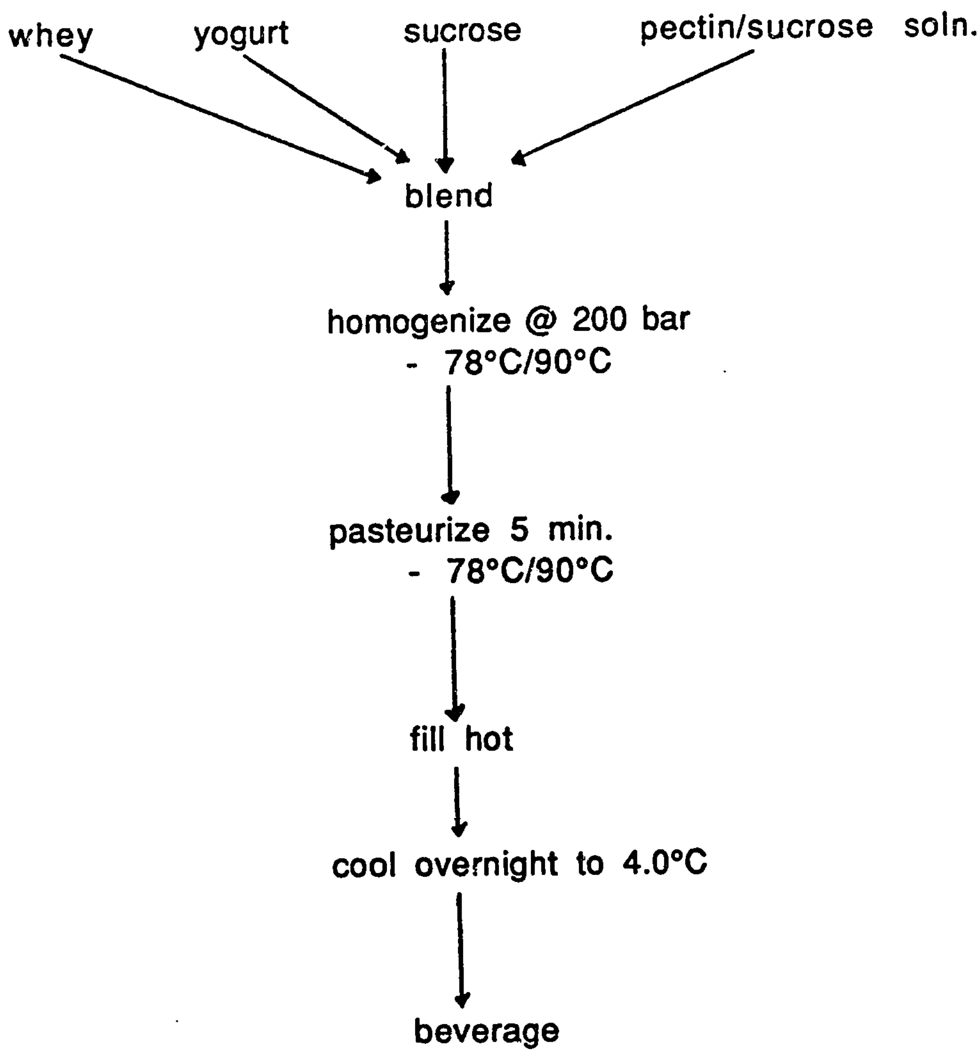


SUCROSE/PECTIN SOLUTION PRODUCTION:



Appendix 3. continued: Flow chart of production methods for beverages outlined in section 3.3.

BEVERAGE PRODUCTION:



Appendix 4. Flow chart of production methods for the SUFR model systems outlined in section 3.5.1

SIMULATED UF RETENTATE PRODUCTION:

water → blend ← dried whey protein concentrate

blend
↓
hydrate
4 hr. @ 4.0°C
↓
adjust pH
4.0/5.0/6.5
↓
SUFR

PECTIN SOLUTION PRODUCTION:

pectin → blend ← water

blend
↓
pectin solution
↓

BEVERAGE PRODUCTION:

pectin solution → blend ← SUFR

blend
↓
homogenize
200 bar @ 22°C
↓
pasteurize @ 78°C
↓
fill hot
↓
cool overnight to 4.0°C
↓
beverage