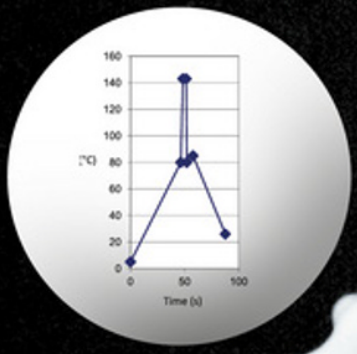
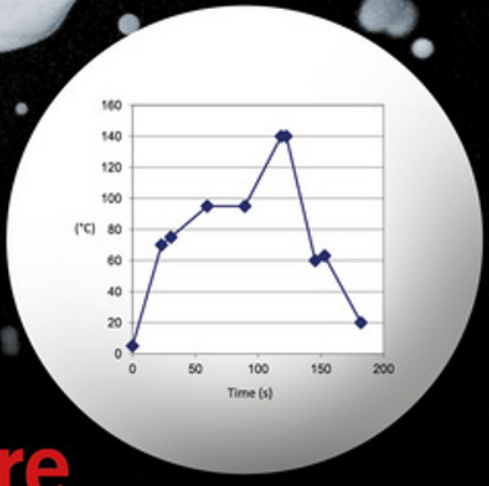
HILTON C. DEETH AND
MICHAEL J. LEWIS





High Temperature Processing of MILK AND MILK PRODUCTS

WILEY Blackwell

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About the Authors

Hilton C. Deeth

Hilton Deeth grew up on a dairy farm in Australia which engendered in him a love of all things dairy from an early age. After completing a science degree and a PhD in organic chemistry at the University of Queensland, he worked as a research food scientist in the Queensland Department of Primary Industries (QDPI) for 23 years. His areas of research included quality aspects of milk, butter and cheese, specialising in lipase and lipolysis, and flavour problems associated with milk fat. He also initiated and led a seafood research group which became the major seafood group in Australia. At the time of leaving QDPI to take up an academic position at the University of Queensland in 1995, he was Manager of Food Research and Development, responsible for a range of projects on dairy, seafood, meat, and fruits and vegetables.

At the University of Queensland, he taught dairy science, seafood science, emerging food technologies and food product development. He also supervised research projects on various dairy and seafood topics and was advisor for more than 30 PhD and research Masters students, as well as several coursework Masters students. His main dairy research interests, and the topics of his students' theses, were UHT processing and products, quality aspects of milk, yogurt and milk powders, and new processing technologies.

In 1996, he established a specialist Centre for UHT Processing and Products for the Australian dairy industry at the University of Queensland and directed the Centre until it was merged with four other specialist dairy centres in Australia to form Dairy Innovation Australia Ltd (DIAL) in 2008. The research initiated in the UHT Centre was continued in a DIAL-funded Food Science Research Program at the University of Queensland which he managed until his retirement in 2011. He has published 150 research papers and reviews, and 28 book chapters. A large proportion of these are on topics directly related to this book and hence the book contains numerous references to them.

Since retiring as Emeritus Professor of Food Science, he has remained involved in dairy science and technology as a consultant assisting dairy companies with product and process development, and trouble shooting, and also providing technical training in UHT processing and other dairy topics for companies in Australia and other countries. This book also draws on the knowledge and experience gained in these industry involvements.

Michael J. Lewis

Mike Lewis worked for over 38 years at the University of Reading as a Lecturer and Senior Lecturer in the School of Chemistry, Food and Pharmacy before retiring in Sept 2011. He was educated at the University of Birmingham in the Department of Chemical Engineering where he gained a BSc, MSc (Biol Eng) and PhD. Over the last 40 years, he has acquired considerable expertise in many topics related to food science and technology, including physical properties of foods, food processing operations, milk and milk processing, heat treatment, evaporation, drying and membrane technology. He has an extensive publication record in these areas, with over 80 refereed papers and over 20 book chapters and three books. In addition, he was actively involved in maintaining the University pilot plant and generating considerable income from industry and research funders. In the context of this book he has worked with a UHT pilot plant since 1976, when Reading University acquired an APV Junior UHT plant. Since then it has been used for teaching, product development and research and has earned the University in excess of £350,000 from outside work. He has helped many companies with product and process development and staff training activities in different countries.

He supervised over 30 PhD students and over 150 BSc and MSc project students. His research activities have focused on minerals in milk and their interactions with proteins, especially with regard to calcium and also magnesium and their role in casein micelle stability. Stability aspects that have been studied include ethanol stability, heat coagulation, involving heat coagulation times, stability to in-container sterilization, UHT sterilization, involving fouling of heat exchangers and deposit formation and fouling of UF membranes. His most recent work involves developing procedures for measuring pH and ionic calcium at high temperatures, to better understand their role in heat stability of milk and the effects of chelating agents on these parameters related to calcium fortification, calcium removal and stabilizer addition. Much of his research has been conducted in close partnership with the food and dairy food industries and he has recently completed four sets of workshops for Dairy Innovation Australia Ltd (DIAL) on UHT processing and several for major multi-national producers of milk products. He remains research active and is a regular reviewer for a number of the major food and dairy journals. He loves teaching and sharing his knowledge and wishes to continue doing this.

Preface

This book has arisen from a productive period of collaboration between the authors. They first met when Hilton visited the University of Reading in 1995, on a fact-finding mission for setting up a UHT Centre in Australia. In 2003, he returned to the UK to spend a short sabbatical period at Reading University and shortly afterwards Mike spent time at the UHT Centre at the University of Queensland. This led to funding by Dairy Innovation Australia Ltd (DIAL) for a PhD student, who studied at both Universities and helped to cement a fruitful partnership and long lasting friendship.

In 2000, Mike produced a book in collaboration with Neil Heppell on continuous thermal processing (Lewis & Heppell, 2000). This arose from a suggestion by the publisher for a revision of Harold Burton's book on UHT processing of milk (Burton, 1988), which was published in 1988 and which was then out of print. Harold dedicated that book to all those who worked on UHT processing and aseptic filling at the National Institute for Research in Dairying (NIRD) between 1948 and 1985 and particularly those in the Process Engineering Group, of which he was Head for much of the time. During that period Harold's group carried out much of the fundamental work on understanding the safety and quality of UHT milk and his name was known worldwide. This was the major publication in this area in that era and should still be consulted by anybody involved with UHT processing. Harold retired in 1985. Mike's relationship with Harold extended for over 20 years and is described in the preface to the Lewis and Heppell book.

The Lewis and Heppell book concentrated on continuous processing and aimed to expand the range of food products that were featured beyond milk products. This aim was simple to state but more difficult in practice to achieve, as the majority of publications dealt with milk and milk-based products. Today, the commercial reality is that the range of heat-treated, particularly sterilised, products available to the consumer is much wider, although important technical information on matters such as formulations and processing conditions is less readily available in the public domain. This book also incorporated pasteurisation and heat treatments designed to further extend the shelf-life of pasteurised products and also acidic products such as fruit juices. In fact, pasteurised products are more widespread than sterilised products in many countries.

In this volume we have aimed to produce a book that gives a clear explanation of the principles involved in high-temperature heat treatment processes. The main emphasis throughout is on product safety and quality. To fully understand these issues involves integrating a number of important scientific disciplines covering the physical aspects of foods, the transfer of energy and the effects of heat on the chemical, biochemical and

sensory characteristics and the problems inherent in dealing with biological raw materials. Thus there is a section which describes the basic physical properties of the products that are to be heat treated. We have aimed to provide a good balance between the engineering aspects and the chemical, biochemical, microbiological and sensory issues which have to be considered to produce foods which are both safe and of a high quality. One of the innovations is a better understanding of factors affecting heat stability and the role of pH and ionic calcium and the interesting relationship between them, along with suggestions for measuring these parameters at sterilisation conditions. Another is the use of temperature-time profiles for assessing the microbiological and chemical effects of a given process.

The book covers microbiological issues, other thermal processes such as pasteurisation, extended shelf-life (ESL) and in-container sterilisation, UHT processing conditions and characterisation of processes, engineering aspects, heat stability, fouling and cleaning, changes during storage, quality assurance procedures, alternative technologies, shelf-stable products other than sterilised cow's milk, products that can be manufactured from UHT milk and analytical procedures. We have devoted a chapter specifically to products other than white cow's milk to reflect the increasing importance of these products.

Some products that are UHT processed are considerably more viscous than milk or cream are, and some of these contain discrete particles, deliberately added and not present as sediment. Thus it covers situations where streamline flow conditions are likely to prevail, as well as the thornier problem of heat-treating products containing particles, ideally ensuring uniform heating of the solid and liquid phases. One observation is that there are still relatively few UHT products in this category, although this may change as Chinese consumers like drinks containing particulates. The Lewis and Heppell book still remains worth consulting in this area.

There is a great deal of interest and research activity in alternative technologies and processes for pasteurising and sterilising foods and these have been addressed in this book. However, these technologies have to compete against heat treatment, which is a very effective, convenient and energy-efficient method of processing foods. In fact, the application of heat in HTST pasteurisation and UHT sterilisation are two well established processes. Nevertheless, alternative technologies are finding applications, mainly in niche areas and these aspects are discussed. In most cases, they add a considerable processing cost to the product.

The layout of this volume should help the reader who wishes to explore specific topics in depth. We have taken care to ensure that the book is well cross-referenced and indexed, which will help the reader who wishes to browse. Perhaps a novelty is the chapter on analytical procedures which can be used to further understand some of the issues involved in UHT processing and products. A recent excellent publication on analytical procedures for milk and milk products contains only three indexed references to UHT milk. We hope this chapter goes some way to redressing this imbalance. One interesting challenge regarding analytical methods is to identify applications for some of the powerful instrumental techniques which are now available. Some of these, such as proteomics and molecular-based microbiological techniques, are now well established and represent quantum leaps in milk analysis.

In the final chapter (Chapter 12) we have outlined several aspects on which we believe there is currently insufficient information and require further research. These have

been identified through our research and consultancy activities and confirmed during the preparation of this book. In that chapter we have also collated several key references to books, book chapters and review articles which can be consulted for further information on specific topics.

We hope that this book will be stimulating to undergraduate and postgraduate students of food science and technology, as well as industry biotechnologists, food technologists and engineers who are involved or interested in heating and cooling milk and other products of a biological nature.

We believe that it will provide a useful reference source for the food industry and provide a focus for gaining a better understanding of the factors influencing safety and quality of heat-treated products. We are confident that a major strength of the book is the combination of theoretical knowledge derived from the considerable research output in the subject area with our practical experience of heat processing. We have tried to make our explanations as clear as possible, especially when interpreting results from those articles where it was unclear what was really intended.

There have been many other constraints and competing pressures in meeting the publisher's deadlines. However, as we have both recently retired, issues such as teaching, quality audits, research assessment exercises, enjoyable teaching activities, research priorities and University administration are no longer excuses.

Finally, returning to Harold Burton, in brewing technology, there is a process known as "Burtonising" the water, to ensure that water used for beer production has a composition similar to that found in Burton-on-Trent, one of the great brewing centres in the UK. It could well be argued, considering his enormous contribution to the subject area, that the term Burtonising the milk, should be the synonymous with UHT treatment. In support of this, it is especially noteworthy that Harold Burton's 1988 book on UHT processing has recently been reprinted by Springer, in its original form.

List of Abbreviations

A.	*Anoxybacillus*
AQL	Acceptable quality limit
ATP	Adenosine triphosphate
B.	*Bacillus*
BCA	Bicinchoninic acid
C.	*Cronobacter, Coxiella*
CAR	Carboxen
CCP	Colloidal calcium phosphate
Cl.	*Clostridium*
CMC	Carboxymethyl cellulose
cP	Centipoise
cSt	Centistoke (Unit of kinematic viscosity)
DEFT	Direct epifluorescent technique
DSHP	Disodium hydrogen phosphate
DVB	Divinylbenzene
E401	Sodium alginate
E407	Carrageenan
E410	Locust bean gum
E412	Guar gum
E451	Sodium & potassium triphosphates
E466	Cellulose gum, carboxymethyl cellulose
E471	Mono & diglycerides of fatty acids
EDTA	Ethylenediaminetetraacetic acid
EGTA	Ethylene glycol tetraacetic acid
ELISA	Enzyme-linked immunosorbent assay
ESL	Extended shelf-life
FCM	Flow cytometry
FDNB	1-Fluoro-2,4-dinitrobenzene
FFA	Free fatty acids
FID	Flame ionization detection
FITC	Fluorescein isothiocyanate
FOS	Fructo-oligosaccharide
FPD	Freezing point depression
FTIR	Fourier transform infrared
G.	*Geobacillus*

GC	Gas chromatography
GOS	Galacto-oligosaccharide
HCA	Hierarchical cluster analysis
HCT	Heat coagulation time
HMF	Hydroxymethylfurfural
HPCD	High pressure carbon dioxide
HPH	High pressure homogenisation
HPLC	High performance liquid chromatography
HPP	High pressure processing
HTST	High-Temperature, Short-Time
ICP	Inductively coupled plasma
IDF	International Dairy Federation
L.	*Listeria*
LA	Lactic acid
LAL	Lysinoalanine
LMTD	Logarithmic mean temperature difference
LPS	Lactoperoxidase system
LRM	Lactose-reduced milk
LTI	Low-temperature inactivation
MCC	Microcrystalline cellulose
MF	Microfiltration
MPC	Milk protein concentrate
MSNF	Milk solids non-fat
MWCO	Molecular weight cut-off
NCN	Non-casein nitrogen
NMR	Nuclear magnetic resonance
NPN	Non-protein nitrogen
OHTC	Overall heat transfer coefficient
OPA	Ortho-phthalaldehyde
PAGE	Polyacrylamide gel electrophoresis
PATP	Pressure-assisted thermal processing
PATS	Pressure-assisted thermal sterilisation
PCA	Principal component analysis
PDMS	Polydimethylsiloxane
PEF	Pulsed electric field
PFPD	Pulsed flame photometric detector
PHE	Plate heat exchanger
PPC	Post-processing contamination or post-pasteurisation contamination
Ps.	*Pseudomonas*
RDA	Recommended daily allowance
RDI	Recommended daily intake
RP	Reversed phase
RSMP	Reconstituted skim milk powder
SCC	Somatic cell count
SDHP	Sodium dihydrogen phosphate
SDS	Sodium dodecylsulfate
SFR	Sterility failure rate

SGE	Starch gel electrophoresis
SHMP	Sodium hexametaphosphate
SMP	Skim milk powder
SPME	Solid phase microextraction
St.	*Staphylococcus*
Str.	*Streptococcus*
TA	Titratable acidity
TBA	Thiobarbituric acid
TCA	Trichloroacetic acid
TNBS	Trinitrobenzenesulfonic acid
TSC	Trisodium citrate
TVC	Total viable count
UHT	Ultra-high temperature
UTP	Uniform trans-membrane pressure
WPC	Whey protein concentrate

1

History and Scope of the Book

1.1 Setting the Scene

Bovine milk is the main source of milk in the world today. Table 1.1 illustrates some production data for the leading bovine milk producing countries in the world. The first column shows total milk production, whereas the second shows milk production expressed as per head of population. Thus countries like New Zealand and Ireland (see footnote) produce large quantities per capita, whereas countries such as China, although positioned in the top five milk producers in the world, are most probably not producing sufficient milk for their increasing populations who are developing a taste for milk and milk products. USA and Brazil are also large producers of bovine milk. Much of the milk in Brazil is consumed as liquid milk with a fair proportion being UHT processed.

It is very exciting time to be writing a book on high-temperature processing, particularly ultra-high temperature (UHT) processing. UHT is a continuous process and as such is applicable to any product that can be pumped through a heat exchanger and then aseptically packaged, although the vast majority of products are either milk or milk-based. UHT milk and milk products are now global commodities and are being transported large distances to all parts of the world. In a number of traditional milk-drinking countries, for example, UK, Greece and Australia, pasteurised milk is still the milk of preference and the cooked flavour that is associated with UHT and sterilised milk is given as a major reason for maintaining this status quo (Perkins & Deeth, 2011). In contrast, in some other countries much more UHT milk is consumed than pasteurised milk. For example, in France, Belgium and Portugal, more than 90% of all liquid milk purchased is UHT-treated, whereas in the UK, Norway, Sweden, Australia and New Zealand, it is less than 10%. Similar variations are also found in other parts of the world, with less than 5% of UHT milk being consumed in India and USA but over 60% in Vietnam and China. In other words, availability and also preferences for pasteurised or sterilised milk vary from country to country. Some examples for Europe and other parts of the world are given in Table 1.2.

Recently, there has been a substantial increase in UHT capacity in all parts of the world. In part, this is to supply the increased demand for UHT milk from China. It is also predicted that there will be an increased demand from Africa and other parts of South East Asia. Since UHT milk does not require refrigeration and has a long shelf-life, it provides a very convenient way of providing good quality milk to large populations in remote areas, without the need for the expensive cold chain infrastructure. UHT milk is

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Table 1.1 Leading producers of bovine milk in 2012, with populations and production per head of population.

	Milk production, 2012 (billion L)	Population (billion)	Per capita consumption (L/person)
United States of America	90.9	0.318	286
India	54.0	1.244	43.4
China	37.8	1.364	27.7
Brazil	32.3	0.204	158
Russian Federation	31.6	0.146	216
Germany	30.5	0.081	377
France	24.0	0.066	364
New Zealand	20.0	0.0046	4350
Turkey	16.0	0.078	205
United Kingdom	13.9	0.065	214
World	**620.3**	**7.25**	**85.6**

from: http://dairy.ahdb.org.uk/market-information/supply-production/milk-production/world-milk-production/#.VzxQVHn2aUk and world population figures

Table 1.2 Percentage of drinking milk which is UHT processed in various European countries and worldwide.

Europe	
Greece	0.9
Norway	5.3
UK	8.4
Austria	20.3
Germany	66.1
France	95.5
Spain	95.7
Belgium	96.7
Worldwide	
US	2
India	3
Australia	11
Japan	11
Malaysia	28
China	32
Thailand	46
Vietnam	62

Information from Wikipedia and Datamonitor (China has the largest forecast growth increase in UHT milk consumption over the period 2012 to 2020. India also has a high projected growth rate but is starting from a much lower base level).

now transported to China and other parts of South East Asia from countries such as Australia, New Zealand and even longer distances from USA and several countries in Europe. Both large multinational conglomerates and much smaller companies are engaged in these activities.

The demand for UHT milk is increasing worldwide. It has been estimated that the compound annual growth rate for UHT milk in the world between 2013 and 2019 will be 12.5%, with the global market reaching USD 137.6 billion in 2019 (Persistance Market Research, 2014). In locations where fresh milk is not available, UHT milk can be produced from milk powder. Also milk demand is increasing in locations where there has previously been no strong culture of drinking milk; there is a continuing investment in UHT capability in various parts of the world to meet this demand.

Demand for UHT milk is not the only factor that is changing in relation to the market for milk and milk products. The variety of milk-based beverages is constantly expanding. In the early days of UHT processing, only white milk and some cream products were processed. The variety in milk drinks has since mushroomed and now includes flavoured milk and products containing additives offering health benefits, derived either from naturally occurring components in the milk or non-milk components, such as plant extracts, fruit juices and other substances such as melatonin and dietary fibre (see Tables 1.3 and 1.4). There are also many products of non-dairy origin; these are covered in more detail in Chapter 9.

Whatever type of UHT product is being produced, a key consideration is to ensure that the formulation has good heat stability. The first consideration is a knowledge of the chemical composition of raw milk which is complex and subject to day-to-day and seasonal variation, as illustrated by data on a bulk milk supply collected in the UK over 15 months (Chen *et al.*, 2014) (see Table 1.5). Secondly, it is crucial to understand how different additives, for example, fruit essences, flavours, mineral salts, stabilizers and emulsifiers will influence heat stability in order to ensure that fouling of the UHT plant and sediment formation in the treated product are minimized. This has been one of the authors' main areas of research and an aim of this book is to share our experiences dealing with these topics. Similar issues arise with some non-bovine milk products, such as goat's and camel's milk, which have poorer heat stability than cow's milk and need to be stabilized to be suitable for UHT processing. Historically, pH was considered to be a very important determinant of heat stability of milk, but now the role of both pH and ionic calcium and their interrelationship is better understood, as is how they change when milk is heated to 140 °C and then cooled; these issues are discussed in Chapter 6.

The first and overriding objective is to make UHT products safe to drink by ensuring that they are adequately sterilised and that they will not cause outbreaks of food poisoning. The most heat resistant pathogen is *Clostridium botulinum.* It is noteworthy that raw milk is not considered to be a source of this pathogen and incidents of botulinum have not been attributed to liquid milk and only very rarely to milk products. However, raw milk may contain some bacterial spores that are more heat resistant than *Cl. botulinum* and ensuring that these are inactivated during the UHT process will ensure the UHT milk is free of *Cl. botulinum*, even if the bacterium may have inadvertently found its way into a formulated milk product from other sources. In fact, some recent work using a probabilistic assessment model predicted that contamination of a UHT product with *Cl. botulinum* might arise only once in 367 years (Pujol *et al.*, 2015). The release of product containing the thermophilic spore-former *Geobacillus stearothermophilus* was

Table 1.3 Some drinking milk products available commercially or being developed.

Milk types
Full-cream, skim, semi-skim – HTST, ESL, UHT, sterilised
Flavoured
Lactose-reduced
Carbonated
Goat's, sheep, buffalo's, horse and camel's milk
Microfiltered
Breakfast milks
A2 milk
Yogurt drink
Pet milk
Soy, almond, oat and other plant "milks"
Additives/fortifiers
Calcium and other minerals
Vitamins
Plant sterols and stannols
Omega-3, conjugated linoleic acid (CLA)
Microparticulated whey protein
Milk bioactive peptides
Dietary fibre (e.g., β-glucan, inulin)
Melatonin
Polyphenols
Oligosaccharides

calculated to be much higher than this, but this is not a food pathogen and will only be problematic where the temperature of the products during storage is allowed to reach >50 °C, such as in hot climates.

The ideal UHT milk product should be free of environmental contaminants and also be commercially sterile. This is the combined responsibility of the milk producer, the milk processor and the packaging technologist. However, this is by no means the end of the process because UHT milk will then be expected to be acceptable to the consumer and have a "best before date" of at least six months (Rysstad & Kolstad, 2006). There are sound scientific explanations why six months is a reasonable period and problems may be encountered if this is extended. Although it is possible to eliminate microbial activity, it is not possible to prevent chemical and physical reactions taking place; in some circumstances, enzymatic reactions such as proteolysis and lipolysis may also be encountered. Thus, there is in place a dynamic situation in UHT milk during storage, where its active components are reacting or interacting and, as a result, some of its important quality attributes are also changing. The rate at which these changes take place is

Table 1.4 Some high-temperature-processed milk products from different countries.

Product	Country	Brand Name	Format	Other comments
On-the-go snack	USA	Dynamoo	UHT, 8 fl oz, boxes	
Red-bean flavoured milk	Taiwan	Acacia Lover	UHT, 250 mL bottle	
Fruit and milk drink	France	Danoo Mon fruit prefere	1 L re-sealable carton	Acidic product
Milk shake	USA	Cold Stone Milk Shake	UHT, 12 fl oz plastic bottle	
Nutritious weight loss shake	UK	USlim	UHT, 250 mL plastic bottle	
Dairy based functional drink	Latvia	Lakto	100 g plastic pots	P24, digest, acidic product
Coffee milk	Finland	Kahvi Maito, Valio	1 L cartons	Claimed to foam well
Milk with real fruit pieces	China	Meng niu	UHT cartons	Available since 2007
Milk with cereal grain	China	Yi Li	UHT cartons	Thai rice and Euro wheat
Milk with oat cereals	China	Meng niu	UHT cartons, "Miao Dian"	
Black cereal milk	China	Guanxi Huangshi Dairy Co ltd	UHT cartons	Black sesame, rice and beans
Milk and peanut protein			UHT cartons	
Breakfast milks	Australia	Up and Go (sanitarium)	UHT cartons and plastic bottles	Many flavoured varieties
	UK	Weetabix	Tetra Prisma	2014
	UK	The Fuel Station		Caffe Latte
High protein drinks (whey based)	UK	Upbeat (Good Whey Company)	ESL, plastic bottles	Microparticulated whey protein (8%)
Milk based recovery and build drink		Maxi-Nutrition	UHT, plastic bottles	9% protein, fruit flavoured products
Infant and follow-on formulations	UK	SMA, first infant milk	Sterilised, glass bottle	Protein, 1.3%;fat 3.6%, C/H 7.3%
	UK	Aptamil, first milk	UHT, 100 mL bottle	Contains GOS, FOS
	UK	Aptamil, toddler milk	UHT, 200 mL bottle	Contains GOS, FOS

influenced by storage temperature. Within the life-span of a carton of UHT milk, it may be stored at temperatures from -10 °C to over 50 °C. For example, during transportation from UK to China it may go through fluctuating temperatures as it passes from the UK through the Gulf states and across the equator. Furthermore, large countries such as China, Australia and USA have several climatic zones, and ambient temperature may

Table 1.5 Composition of bulk raw milk from one farm collected over 15 months.

Composition/properties	Mean (n = 25)	Range	Seasonal variation
pH	6.79	6.73–6.87	SP > SM and A;W > A
Ca^{2+} (mM)	2.05	1.68–2.55	NS
Total solids (%)	12.78	12.31–13.31	A > SM
Protein (%)	3.29	2.89–3.56	SP > SM and A
Total casein (%)	2.36	2.08–2.52	SP > SM and A
Fat (%)	4.08	3.62–4.77	A > SP, SM and W
Lactose (%)	4.59	4.52–4.69	NS
Ash (%)	0.71	0.53–1.03	NS
Total Ca (mM)	29.29	24.53–31.53	NS
Total Mg (mM)	5.11	4.21–5.81	NS
Total citrate (mM)	9.04	8.22–10.09	NS
Total P (mM)	27.52	22.58–33.57	NS
Urea (%)	0.0237	0.016–0.033	NS
SCC ('000)	155	65–357	W > SP, SM and A

Source: Chen *et al.,* 2014. Reproduced with permission of Elsevier.
(SP=Spring; SM=Summer; A=Autumn; W=Winter; NS=Non-significant difference ($p > 0.05$) (from Chen *et al.,* 2014)

extend over a wide range, from below 0 °C to above 50 °C. Also, individual milk cartons from the same specific production batch may have had totally different temperature storage histories by the end of six months. The expectations are that each one of these individual cartons will be still acceptable to the consumer. In our opinion, this is a lot to expect from the product and there is no doubt that the number of complaints will increase from stored products where the best-before period exceeds six months. In fact, the expected best-before time period is now creeping up to nine months or even one year, which is posing some new challenges for the UHT milk producer.

Also, the consumer is becoming more discerning. For example, it is reported that the Chinese consumer spends more time than any other reading food labels. There are a number of things any consumer might notice which could result in their making a complaint. On pouring the product, any physical defects such as fat separation, gelation or sediment will be obvious. The more inquisitive consumer will also notice what is left in the carton after the contents have been removed. Sediment and fat may be left in the carton and, if observed, may be a source of complaint, although this is unlikely to be a safety issue. The colour of the milk may also give cause for concern, especially if it is browner that expected. On tasting, any physical defects which change the mouthfeel might be noticed, for example, increased viscosity or presence of sediment giving rise to a powdery or gritty mouthfeel. Finally, its flavour must be acceptable and not be too cooked, oxidized or lipolytically rancid, as well as free of any other off-flavours and taints. Overall, expecting UHT milk to have a best-before period of longer than six months under all possible conditions is taking it out of its comfort zones. Consumers do

allow some leeway for product imperfections such as a small fat layer on the milk but this cannot be pushed too far. One anecdote is that some consumers are prepared to accept a degree of fat separation, as this indicates that fat is actually present in the product.

1.2 Scope of the Book

This book aims to integrate the scientific information arising from several disciplines that needs to be considered in order to ensure that UHT and other highly heat-treated products are both safe and acceptable to the consumer. UHT processing requires an understanding of aspects of fluid flow and heat transfer, and a detailed knowledge of the properties of the food being processed and of the mechanisms of the various changes that occur during processing and storage. This includes knowledge of its chemical composition, the enzymes that are present and its microbial flora as well as an awareness of possible environmental contaminants.

When any food is subjected to UHT treatment, a large number of heat-induced reactions take place, which, if properly understood and controlled, ensure that the food is safe and that it will have a good appearance and taste for up to six months and probably for considerably longer. The material in this book is derived from the scientific literature related to UHT processing and the personal insights from two practitioners who have spent much of their working lives involved with UHT products and processes.

In order to put UHT processing and products in perspective in dairy processing, an overview of the heat treatments of milk is initially given (see Chapters 2 and 3). Furthermore, the microbiological aspects of these heat treatments and their associated products are provided (Chapter 4) as it has to be remembered that the basic reason why heat treatments are carried out is to destroy micro-organisms. A good understanding of the microbiological aspects is therefore fundamental to ensuring the safety and quality of the products. In these chapters, extended shelf-life (ESL) processing, a sub-UHT heat treatment, is covered in some detail because of the growing demand for ESL products.

1.3 Reasons for Heating Foods

In addition to inactivating micro-organisms, both pathogenic and spoilage, foods are heated to inactivate enzymes, as foods may change and become unacceptable due to reactions catalysed by enzymes. Milk contains about 60 indigenous enzymes (Fox, 2003), some of which, such as lipases and proteases, may cause flavour changes, whereas in fruit, browning may occur as a result of polyphenol oxidase activity. The process of heating a food may also induce physical changes and chemical reactions, such as starch gelatinisation, protein denaturation and Maillard browning, which in turn affect the sensory characteristics, such as colour, flavour and texture, either beneficially or adversely. For example, during the manufacture of canned evaporated milk, forewarming of milk prior to evaporation is essential for preventing gelation and thickening during the subsequent evaporation and canning steps. Heat treatment is also crucial in yogurt manufacture to achieve the required final texture in the product. However, such heating processes may result in loss of important nutrients, although these losses can be reduced by controlling the heating conditions.

Thermal processes vary considerably in their severity, ranging from mild processes such as thermisation and pasteurisation, intermediate processes such as used for ESL milk, through to more severe processes such as UHT and in-container sterilisation processes (see Chapters 2 and 3). The severity of the process affects both the shelf-life and quality characteristics of the product.

A UHT process contains heating, holding and cooling stages. After the product has been heated to the desired temperature, it is held for a short period of time to inactivate the microorganisms before being finally cooled and packaged under aseptic conditions. Continuous processes provide scope for energy savings, whereby the hot fluid is used to heat the incoming fluid; this is known as heat regeneration and saves both heating and cooling costs (see Chapter 5)

A wide range of products are heat-treated, ranging from low-viscosity fluids such as milk and fruit juices, through to highly viscous fluids. The process is more complicated when particles are present, as it becomes necessary to ensure that both the liquid and solid phases are adequately and, if possible, equally heated. A secondary issue is keeping the particulates suspended during storage, especially in transparent containers. The presence of dissolved air in either of the phases becomes a problem as air becomes less soluble as temperature increases and will come out of solution. Air is a poor heat-transfer fluid in comparison to steam and hence its presence affects the rate of heating of the food. For this reason, deaeration is sometimes used.

1.4 Brief History of Sterilisation Processes

Food sterilisation in sealed containers is often attributed to the pioneering work of Nicholas Appert. However, Cowell (1994, 1995) reported that investigations on heating foods in sealed containers were documented and took place earlier than this. He describes the commercialisation of the canning process in East London at the turn of the nineteenth century, which included the contributions not only of Nicholas Appert, but Peter Durand, Bryan Donkin, John Gamble and Phillipe de Girard. It is both noteworthy and worrying that bacteria which are the causative agents of food poisoning and spoilage were not understood until considerably later in the nineteenth century, through the work of Pasteur. He confirmed that the many food fermentations which were spoiling foods were not spontaneous but caused by microbial metabolism. He also discovered that both yeasts and *Acetobacter* could be destroyed by relatively mild heat treatments at about 55 °C. According to Wilbey (1993), Pasteur's work on producing beer, wine and vinegar laid the foundations for hygienic processing and the recognition of the public health implications of hygiene and heat treatment.

Early sterilisation processes were essentially of a batch nature and the food was heated in the container. Batch processing still has an important role in food processing operations and provides the small-scale food producer with a cheap and flexible means of heat-treating foods. The steps involved in a batch sterilisation process are shown in Figure 1.1. Continuous sterilisers had been patented and constructed and were able to heat milk to temperatures of 130-140 °C before the end of the nineteenth century, again well before the benefits of the process were understood. Hostettler (1972) recalls that in 1893, a continuous-flow heating apparatus with an output of up to 5000 L/h had been constructed which could heat milk to 125 °C, with a holding time of up to 6 min.

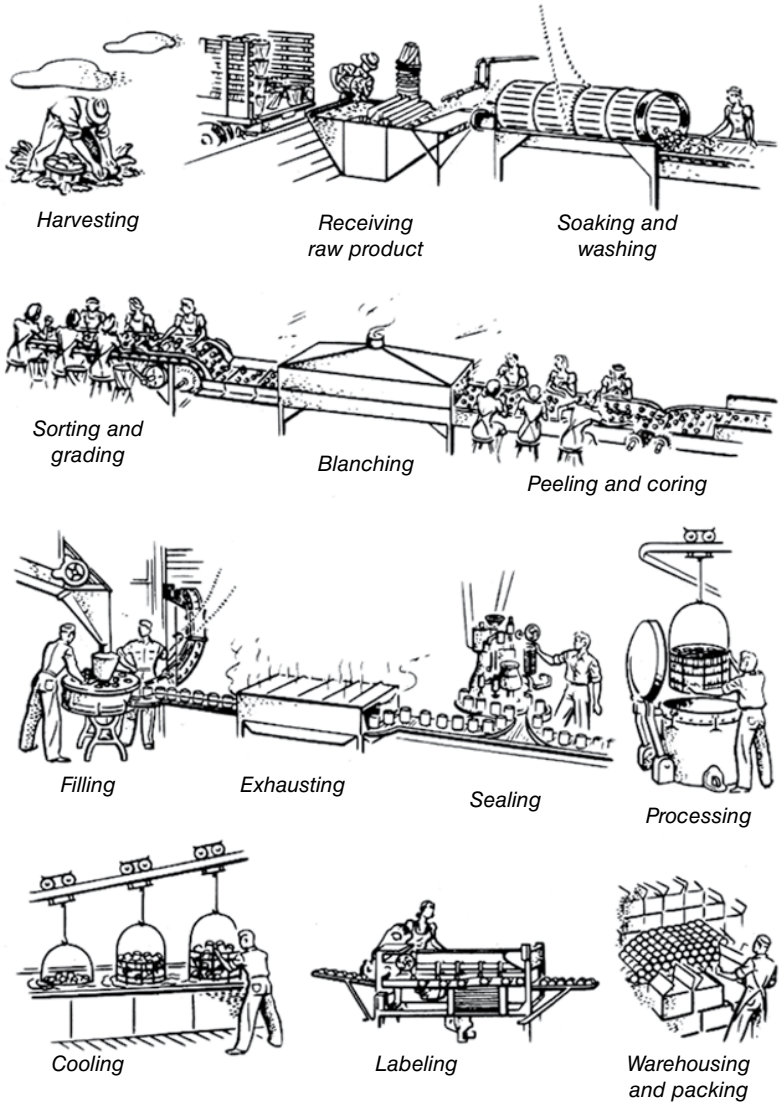

Figure 1.1 The batch canning process (from Jackson & Shinn, 1979).

Around 1909, a number of patents were registered which involved contacting milk with jets of hot air, gases and steam. Aseptically canned milk was produced in 1921 and a steam injection system was developed in 1927 by Grindrod in USA. However, the major initiatives leading to commercialisation of the UHT process began in the late 1940s, through the development of concentric-tube sterilisers and the uperisation steam-into-milk UHT system, which was developed in conjunction with the Dole aseptic canning system. UHT milk was not commercially available in the 1940s and early 1950s, as evidenced by the absence of information in both Cronshaw (1947) and Davis (1955). During the first half of the twentieth century, investigations took place side-by-side into

in-container sterilisation and UHT processing, but the unsolved difficulty of filling the sterilised milk, without recontamination, into containers caused the interest in continuous processes to wane, so sterilisation of milk in sealed containers retained its dominance at this time. It is also noteworthy that many of these early investigations involved direct heating and the only mention of UHT-type processing in Davis (1955) was to uperisation, a steam injection process. In fact the marketing of uperised UHT milk in cans was first practised in Switzerland in 1953, with milk heated by steam injection at 150 °C for 2.4 s and flash cooled. The dominance of sterilised milk around that time is also illustrated in Davis (1955).

As mentioned, early commercial aseptic filling machines filled milk into metal cans, which were usually sterilised by superheated steam, which could be used at atmospheric pressure and avoided condensation and wet cans. A shelf-life of 4 to 6 months was claimed for the product.

The main developments in getting UHT milk to the market place occurred between the early 1960s and 1972 and were rapid. A major development was the use of hydrogen peroxide to sterilise the packaging material. Typical conditions then were 17% w/v solution, with a wetting agent. Hydrogen peroxide was evaporated off with hot air at about 180 °C. Equipment using this procedure was first commercialised in 1961 and from this point availability of UHT products started to accelerate.

Regulations permitting UHT milk in the UK were introduced in 1965. In 1968 UHT milk was introduced in Germany and in 1969 it commanded less than 2% of the liquid milk market. Its success there is illustrated by the fact that now over 90% of milk consumed is UHT treated. In Australia, the first successful UHT operation commenced in 1968 although an earlier installation ceased operation after a few years due in part to technical difficulties such as age gelation (Zadow, 1998).

In 1970, Hsu published the first book on UHT processing of dairy products and this was followed in 1972 by the first International Dairy Federation (IDF) monograph on UHT milk and a revised version in 1981. These publications catalogued most of the technical challenges that had been recognized and investigated in order to produce sterile milk of long shelf-life by means of a continuous-flow process involving heating at a high temperature for a short time, followed by aseptic packaging. By that time it had become well accepted as a method for heat treatment of milk for consumption.

A more detailed account of the early development of UHT processing, before it was properly commercialized is given by IDF (1972). The history of the continuous sterilisation process has also been reviewed by Burton (1988).

It is interesting that in the early 1970s there was no clear statement about how long UHT milk should keep. However, it was probably quite short because of the numerous challenges in UHT processing and the lack of a good understanding of the technology and its effects on product quality. An indication of this was given by Singh and Patel (1988) who reported that the shelf-life of UHT milk in India was only 15 days although the expected shelf-life was three months. They identified numerous aspects of the UHT process which required attention to improve the shelf-life including the initial bacterial content of raw milk, selection of suitable time – temperature conditions, problems related to heat-resistant proteases, sedimentation and deposit formation, and problems with the packaging system; these would have been similar to those encountered by the early UHT processors. With the developments in technology and a better understanding of the key determinants of shelf-life, together with market demands, it is not

uncommon for the "best-before" period to be now set at nine months, and more recently 12 months, as discussed above.

At this point it is instructive to state two descriptions of a UHT treatment from the latest EU regulations (Hickey, 2009): "Continuous flow at a high temperature for a short time with not less than 135 °C for a suitable holding time such that there are no viable spores capable of growing in the treated product when kept in an aseptic container at ambient temperature" and "Sufficient to ensure that the products remain microbiologically stable after incubating at 15 days at 30 °C in closed containers, or 7 days at 55 °C in closed containers, or after any other method that demonstrates that appropriate heat treatment has been applied." The EU regulations no longer state what level of microbial activity would constitute microbial sterility after these incubation periods, whereas previous regulations stipulated it to be less than 100 cfu/mL, which seems to be a reasonable standard. This was illustrated by Quratulain and Saeed (2004) who found two brands of commercial UHT milk had mesophile counts of 75 and 96 cfu/mL after storage for 40 days; they commented that the milk met the "requirements of the standard". The current Australia and New Zealand Food Standards match the EU regulation and state that UHT milk and cream "should comply with a test for commercial sterility" (FSANZ, 2011).

In conclusion, it is worthwhile considering what factors have changed over the past 15 years since the publication of the Lewis and Heppell (2000) book. The basic processing technology and heat exchanger configurations have changed little although improvements continue to be made. There is now more recognition of the roles of the heating and cooling profiles. This has led to a wider use of the concept of bacterial and chemical indices (see Chapter 3) for characterising the process and understanding the effects of different processing conditions on the quality of the products.

The processing run times that can be achieved have increased considerably. It is now claimed that it is possible to obtain runs of 40 h. The main way of achieving this has been to include a protein stabilisation tube. One explanation is that this does not eliminate fouling but it causes the fouled deposit to accumulate in the protein stabilisation tube, which is away from areas where its build-up may be more critical.

The control and instrumentation has improved and information on when the plant needs to be cleaned and also when cleaning has been completed is more readily available. One possible consequence of longer run times is that the cleaning times may be longer, although this has not been reported to be the case. Also, a lot more information is now available to UHT process operators to provide them with a better understanding of the performance of the heat exchanger.

There have been other more subtle changes, such as improvements in homogeniser valve design, which should lead to an improvement in emulsion stability. This is crucially important as the "best-before" date for many products is now nine months or twelve months.

The product range continues to expand and there is now more emphasis on environmental considerations; for example, how much water and energy is used and how much waste is generated. One of the advantages that UHT processing offers is that the product does not need to be refrigerated during transportation or storage, although refrigeration or some form of temperature control may be beneficial in hot climatic conditions.

It has been difficult selecting a concise title for this book to reflect its entire content. However, we have chosen high temperature processing of milk and milk products. One reason for this is the dominance of white milk and other milk-based products in the

Table 1.6 Volumes of liquid dairy and dairy-like products sold worldwide in 2015 (Source: Reproduced with permission of Tetra Pak Compass).

Product	Volume (billion litres)
White milk	216.8
Baby and toddler milk	19.6
Flavoured milk	18.4
Soy milk	17.8
Drinking yogurt	9.4
*RNGS milk	8.3
Dairy cream	4.0
Sweetened condensed milk	2.5
Buttermilk	2.3
Evaporated milk	1.6

* RNGS is rice, nuts, grains and seeds products

global dairy products market, as shown in Table 1.6. Almost all of the beverages listed are subjected to thermal processing of some kind and many of them to UHT processing. Non-dairy products such as the rice, nuts, grains and seeds (RNGS) products are making inroads into the nutritive beverage market. These and most of the other products listed in Table 1.6 are discussed in Chapter 9 while some emerging technologies which have potential for processing these products are covered in Chapter 10.

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2

Heat Treatments of Milk – Thermisation and Pasteurisation

2.1 Introduction

This chapter explains the important principles and procedures for producing heat-treated milk which is safe and of high quality. It includes information gained by the authors through their combined experiences of teaching, pilot plant work, research and troubleshooting.

Raw (or untreated) milk consumption has fallen considerably worldwide over the past 30 years and in some areas it is now illegal to sell raw milk for direct human consumption. In the UK, raw milk consumption now accounts for less than 0.1% of liquid milk consumption and in many countries, for example, Scotland and Australia, its sale is prohibited. Consequently most milk for consumption is now heat treated.

The two main treatments are pasteurisation and sterilisation, with treatments somewhere between these for extended shelf-life (ESL) products. The main aims of heat treatment of raw milk are to reduce the microbial population, both pathogenic and spoilage, to inactivate enzymes and to minimise chemical reactions and physical changes during storage. Such heating may also alter the sensory characteristics of the milk, for example, its overall appearance, colour, flavour and texture, as well as its nutritional value, but will make it safe for consumption and improve its keeping quality.

Most of the milk destined for conversion to dairy products is also heat treated at some point, an exception being those cheeses which are made from raw milk. Such processes include thermisation, which is milder than pasteurisation and used for extending the storage time of raw milk, preheating or forewarming applied to milks prior to evaporation and powder production, a high pasteurisation treatment used in yogurt manufacture, as well as pasteurisation, ESL treatment and sterilisation. The most common heat treatments, in order of increasing severity, are thermisation, pasteurisation, ESL treatment, ultra-high temperature (UHT) treatment and in-container sterilisation. The heating conditions for these are summarized in Table 2.1 together with their bactericidal effects and their effects on selected enzymes. These treatments and their effects on milk are reviewed in this chapter. As high-temperature treatments are the focus of this book, an overview only is given of sterilisation treatments, especially UHT processing, here as these are covered in detail in subsequent chapters.

High Temperature Processing of Milk and Milk Products, First Edition. Hilton C. Deeth and Michael J. Lewis.

Table 2.1 Heat treatments used for milk (in increasing order of severity).

Heat treatments	Temperature–time conditions	Bactericidal effect	Effect on selected enzymes
Thermisation	57-68 °C for 5-20 s	Destroys most non-spore-forming psychrotrophic spoilage bacteria	Does not inactivate milk alkaline phosphatase, lipase, lactoperoxidase, plasmin or bacterial proteases/lipases
Pasteurisation	63 °C for 30 min; 65 °C for 15 min (batch) 72-82 °C for 15-30 s[1] (continuous, HTST)	Destroys non-spore-forming pathogens and psychrotrophic spoilage bacteria	Inactivates milk alkaline phosphatase and lipase but not lactoperoxidase, plasmin or bacterial proteases/lipases
ESL (extended shelf-life) processing	123-145 °C for <1-5 s	Destroys all non-spore-forming bacteria and most psychrotrophic and mesophilic spores	Inactivates milk alkaline phosphatase, lipase and lactoperoxidase but not plasmin or bacterial proteases/lipases
UHT (ultra high temperature) processing	138-145 °C for 1-10 s	Destroys all non-spore-forming bacteria and all spores except highly heat-resistant spores	Inactivates milk alkaline phosphatase, lipase, lactoperoxidase; and most plasmin but not all bacterial proteases/lipases
In-container sterilisation	115-120 °C for 10-30 min (conventional) 125 °C for 4 min (e.g., Shaka™ technology)	Destroys all non-spore-forming bacteria and all spores except highly heat-resistant spores	Inactivates virtually all enzymes

[1] 72 °C for 15 s are the regulated minimum conditions in most countries

2.2 Thermisation

Thermisation is the mildest heat treatment given to milk. It is used to improve the keeping quality of raw milk when it is necessary for the milk to be held chilled for some time before being further processed. Thermised milk is subsequently used for other heat-treated milk or converted into various milk products. The aim of thermisation is to reduce the growth of psychrotrophic bacteria which may release heat-resistant proteases and lipases into the milk if allowed to reach high levels. These enzymes will not be totally inactivated during subsequent heat treatments and may give rise to off-flavours in processed milk or in subsequently manufactured cheese or milk powders. Conditions used for thermisation are 57 to 68 °C for 5-20 s, followed by refrigeration. Humbert *et al.* (1985) recommended 65 °C for 20 s as these were the minimum conditions for extending the shelf-life by four days at 4 °C. According to IDF (1984), thermised raw milk can be stored at a maximum of 8 °C for up to 3 days. Similarly, Stadhouders (1982) found that thermisation at 64-68 °C for 10 s extended the shelf-life by 3 days at 4-7 °C. Thermised milk is phosphatase-positive which distinguishes it from pasteurised milk, which is phosphatase-negative. Thermisation causes virtually no whey protein

denaturation, does not affect the milk's heat stability as measured by the heat coagulation time at 130 °C (Coghill *et al.,* 1982) and reduces lipase activity by about 50% (Humbert *et al.,* 1985). While thermisation reduces psychrotrophic bacterial growth, it may activate and initiate germination of bacterial spores and accelerate the build-up of the thermoduric bacterium *Streptococcus thermophilus* in the regeneration section of the pasteuriser (Stadhouders, 1982).

2.3 Pasteurisation

2.3.1 Introduction

"Pasteurisation of milk represents one of the singularly successful contributions to the safety of foods of animal origin" (Holsinger *et al*., 1997). Pasteurisation was first practised on wine, prior to 1857 and slightly later on beer. In terms of milk processing, the history of pasteurisation between 1857 and the end of that century came chiefly from the medical profession interested in infant feeding. The first commercial positive holder pasteurisation system for milk was introduced in Germany in 1895 and in the USA in 1907. A most important principle was recognised as early as 1895 that an effective pasteurisation process "will destroy all disease germs" and "a thoroughly satisfactory product can only be secured where a definite quantity of milk is heated for a definite period of time at a definite temperature. Then too, an apparatus to be efficient must be arranged so that the milk will be uniformly heated throughout the whole mass. Only when all particles of milk are actually raised to the proper temperature for the requisite length of time is the pasteurisation process complete" (Cronshaw, 1947). This remains the main guiding principle underpinning current heat treatment regulations for ensuring a successful pasteurisation process. The description of pasteurisation given by the IDF (1986) remains very appropriate: "a process applied with the aim of avoiding public health hazards arising from pathogenic microorganisms associated with milk, by heat treatment which is consistent with minimal chemical, physical and organoleptic changes in the product". This implies that pasteurised milk should be little different to raw milk in terms of its sensory characteristics and nutrient content (Deeth, 2005).

Pasteurisation of milk is now universally accepted, although it did meet with resistance when first introduced (Satin, 1996). There are still devotees who prefer to drink raw milk and many artisan cheesemakers do not use pasteurisation. Pasteurisation is now mostly performed as a continuous process, which is known as the high-temperature, short-time (HTST) process. This allows it to benefit from economies of scale with capacities of modern HTST units of up to 50,000 L/h. These units operate at high heat regeneration efficiencies (>95%) and are capable of long run times of up to 20 h before cleaning is required. A recently opened dairy in the UK has a capacity of pasteurising 1.3 billion litres of milk each year, which is about 10% of all the raw milk produced in the UK. Pasteurised milk does require refrigeration to ensure a long shelf-life, which incurs substantial energy requirements. In many countries it remains the preferred option to UHT milk, for example, UK, Scandinavia, Greece, USA, Australia and New Zealand.

The conditions used in pasteurisation are designed to inactivate the most heat-resistant, non-spore-forming pathogenic bacteria in milk, *Mycobacterium tuberculosis* and *Coxiella burnetii*. According to Codex Alimentarius (2003), pasteurisation is designed to achieve at least a 5-log reduction of *C. burnetii* in whole milk. It therefore results in

very substantial reduction in populations of pathogens that might be present in raw milk with the exception of the spore-former *Bacillus cereus* of which some strains can be toxigenic (Juffs & Deeth, 2007) (see Section 3.2.2.2.1) for more information on *B. cereus*).

There are now some alternative non-thermal processes which have been developed to replace or augment thermal pasteurisation. These include microfiltration, high pressure processing, high pressure homogenisation, pulsed electric field technology, and UV and gamma irradiation (Deeth *et al.,* 2013). These are discussed in Chapter 10. However, a major factor preventing these alternative technologies gaining widespread acceptance is that thermal processes, especially pasteurisation, are firmly established and accepted as being capable of producing safe, high quality and highly nutritious foods in large volumes and at relatively low processing costs. Many of the alternative technologies also face regulatory hurdles. To date, they have not been able to compete in terms of scale of operation, length of processing runs and energy efficiency for high-volume products like milk.

2.3.2 Historical Background

To chart the developments that have taken place with milk pasteurisation, it is interesting to note what was known about the process 50 to 60 years ago, by reference to publications such as Cronshaw (1947) and Davis (1955) which are well worth consulting. Halfway through the twentieth century (~1950), batch pasteurisation was still widely used but the principles of HTST processes were well established. Continuous pasteurisers were available, processing, on average, just under 10,000 L/h. As mentioned in Section 2.3.1, it was well recognised that every element of milk being pasteurised needed to be sufficiently heat-treated. Although from the start pasteurised milk had to satisfy a plate count requirement of less than 10^5 cfu/mL, in the 1940s it became evident from emphasis on keeping quality that these plate count standards had shortcomings. From 1946, the official test for pasteurisation efficacy became the phosphatase test and, as an indicator of milk keeping quality, the methylene blue reduction test was used. The phosphatase test remains in use throughout the world but the methylene blue test is seldom used now. The methylene blue test is a simple way of providing a rough estimate of the bacterial state of a milk sample. Although it is much less used now, it was recently reported being used for assessing the microbial quality of UHT milks imported into Iraq (Al-Shamary & Abdalali, 2011). Traditionally, it has been used more for assessing the bacteriological quality of raw milk.

When pasteurised milk was first introduced, its keeping quality was poor and its shelf-life was short. Household refrigeration was not widespread and usually milk was stored in the larder. A satisfactory keeping quality meant that it would remain sweet and palatable for 24 h after delivery to the consumer and up to 48 h if the consumer was lucky. If milk with a longer shelf-life was required, the only alternative was milk which had been sterilised in the bottle, with its strong cooked flavour and brown colour. UHT milk was not then available. Even in the 1960s, the choice of milk products was limited (UK Milk Marketing Boards, 1964). There was hardly any mention of skim milk. In the UK, 69% of milk produced went to liquid sales, 31% to manufacture, 6.2% was consumed raw, 18% went into condensed milk, and less than 2.6% was used for other products; fermented products such as yogurt received no mention. No breakdown was provided

of what proportion of milk was pasteurised or sterilised and, at this juncture, the heat treatment regulations for UHT milk had just been introduced. Thus considerable commercial interest arose in UHT milk between 1950 and 1965 (see Section 3.4.2).

HTST continuous processes were developed between 1920 and 1927 and for some time the ability of this process to produce safe milk was questioned. The importance of flow control and temperature control was known and it was appreciated that there was a distribution of residence times. Scales of operation were fairly substantial; Davis (1955) quotes HTST plants between 50 and 5,000 gal/h, although the most favoured were about 2,000 gal/h. (note that Imperial units were widely used: 1 gal/h = 4.54 L/h). Run times were cited as being up to 5 h. Milk was cooled to below 43 °F (5 °C) for distribution after pasteurisation, and brine cooling was popular. Energy regeneration up to 72% was achieved and Davis (1955) reported that 75% of liquid milk was processed (pasteurised) using HTST methods. Gaskets were a problem on the early equipment. Milk was not often homogenised, as a visible cream line was a popular feature. Where homogenisation was used, the pasteuriser was run at a slightly higher temperature. Scale formation was also mentioned as being a problem, most likely occurring when poorer quality milk was being processed. If temperatures were not well controlled, a cooked flavour may have resulted and/or the cream line been diminished. Time – temperature conditions which induce a cooked flavour and result in loss of cream line were well known. According to Cronshaw (1947), momentary heating at 169-172 °F (76.1-77.8 °C) or 30 min hold at 158-162 °F (70-72.2 °C) would cause the cooked flavour to appear.

The role of pasteurisation in inactivating *M. tuberculosis* was well established. A key development was in 1927, when North and Park established a wide range of temperature – time conditions to inactivate tubercle bacilli (Cronshaw, 1947). These experiments were performed by heating milk heavily infected with tubercle bacilli at different conditions and injecting them into guinea pigs. A selection of conditions where negative results were found, that is, those where the animals survived, were: 212 °F (100 °C) for 10 s; 160 °F (71.1 °C) for 20 s; 140 °F (60 °C) for 10 min and 130 °F (54.4 °C) for 60 min.

The phosphatase test was in widespread use as an index of correct heat treatment of milk, in particular to ensure that no milk was under-treated. It was developed from pioneering work reported by Kay and Graham (1935) and was based upon the finding that the naturally occurring alkaline phosphatase in milk had similar inactivation kinetics to the inactivation of *M. tuberculosis*. It is interesting that about 70 years later the bacterium *Mycobacterium avium* subsp. *paratuberculosis* (MAP) became of concern to the dairy community (Griffiths, 2006). One procedure recommended to ensure its destruction was to increase the pasteurisation holding time from 15 s to 25 s (Grant *et al.*, 2005). Hickey (2009) pointed out that while this recommendation has been adopted widely by the UK industry, and supported by many retailers, it is a recommendation that is voluntary and is not a legal requirement for HTST pasteurisation, which still remains at 72 °C for 15 s. MAP is discussed in more detail in Section 4.4.1. Further developments were made in the classification of tests for evaluating the pasteurisation process; these included tests for raw milk quality (the platform test); pasteurisability (survival of thermodurics); efficiency of pasteurisation (pathogens and phosphatase); recontamination (thermophilic and coliform bacteria and the methylene blue test); and general bacterial quality, including organisms surviving pasteurisation plus contaminating organisms (plate count).

It was also recognized that it would be more difficult to inactivate microorganisms in situations where clumping of bacteria occurred, although this is not discussed much now. The role of thermoduric and thermophilic microorganisms was recognised and it was fully appreciated that some microorganisms would survive pasteurisation. It is noteworthy that the role of thermoduric bacteria has started to be questioned again (Gleeson *et al.,* 2013); this is discussed in more detail in Chapter 4. Maintaining the cream line was important as most milk was packaged in glass bottles where the cream line was clearly visible. In fact, taking the temperature up to about 78 °C was one method of losing the cream line. Odour and taste were also important quality characteristics. The role of post-pasteurisation contamination (PPC) was recognised, although this became more fully appreciated once pasteurised milk was stored in domestic refrigerators. Davis (1955) reported that when pasteurised milk soured or deteriorated rapidly it was almost invariably due to post-pasteurisation contamination. The situation today is very similar.

A number of installations were introduced for batch pasteurising milk sealed in bottles. Although the keeping quality was comparable to that of HTST pasteurised milk (Davis, 1955), there were some major technical problems and costs were considered to be higher. Consequently, this innovation was relatively short-lived.

"In considering the history of pasteurisation, it is important to remember that, although scientists everywhere agreed fairly closely on the necessary degree of heat treatment, the process itself was loosely (less well) controlled in commercial practice. Milk was frequently either over-heated or under-heated so that it either gave a cooked flavour or was found to contain viable tuberculosis bacteria. In addition, pasteurised milk was often so badly contaminated by unsterile plant, that its keeping quality was decreased" (Davis, 1955).

Several changes have influenced heat treatment of milk over the last 50 years. Some of these are:

- A much wider variety of milk products is available, including skim, semi-skim, flavoured, lactose-reduced, calcium-fortified and a range of speciality milk products with added nutritional and health benefits.
- Milk from species other than cows is more widely available and in the UK goat's milk has increased in popularity. An interesting phenomenon that both authors have encountered is that pasteurised goat's milk has a better keeping quality than pasteurised cow's milk; this still remains a curiosity.
- Scales of operation have increased, with dairies handling upward of 5 million litres of milk a day, most of which is heat-treated in some way.
- Considerable advances have been made in understanding the role of raw milk quality and the role of PPC in keeping quality.
- Domestic refrigeration is much more widely available and the cold chain, involving refrigerated transport and storage systems, has improved. The role of low temperatures in extending shelf-life is better understood.
- With improvement in refrigeration, there has emerged a better understanding of the role of psychrotrophic bacteria, as raw milk remains refrigerated for longer periods prior to pasteurisation and pasteurised milk remains acceptable for longer.
- Homogenisation is now widespread.
- There is a wider variety of packaging options.

- Much less milk is sold in glass bottles; it was 95% in 1975 but is now less than 5% in the UK.
- There is a demand for extended-shelf-life products.
- Environmental issues have become more important in terms of reducing energy and water use, reducing product waste, minimising effluent, reducing detergent usage and minimising the carbon footprint.

2.3.3 Pasteurisation Equipment

2.3.3.1 Holder or Batch Heating

Cronshaw (1947) and Davis (1955) both provide excellent descriptions of equipment for the holder or batch process – individual vessels (heated internally) and externally heated systems with one or more holding tanks. These processes are more labour-intensive than continuous processes and involve filling, heating, holding, cooling, emptying and cleaning. Temperatures attained are between 63 and 65 °C for 15-30 min. They are still used, particularly by small-scale producers who require flexibility and the ability to treat relatively small volumes of a wide variety of products. They are relatively time-consuming and heating and cooling times are considerable; the total time for one batch may be up to 2 h. The time required to reach the pasteurisation temperature can be determined from the following equation:

$$t = \frac{Mc}{AU} \ln\left(\frac{\theta_h - \theta_i}{\theta_h - \theta_f}\right)$$

t = heating time (s) c = specific heat ($J\,kg^{-1}\,K^{-1}$)
M = mass batch (kg) A = surface area (m^2)
U = overall heat transfer coefficient ($W\,m^{-2}\,K^{-1}$): θ = temperature, i, initial: f, final; h, heating medium temperatures (see Section 5.2.1.8.3.3).

The dimensionless temperature ratio represents the ratio of the initial temperature driving force to that of the final approach temperature. The same dimensionless ratio can be used to evaluate cooling times, which tend to be longer than heating times, because of the limitations of chilled water temperature and hence a lower approach temperature. Cooling times can be shortened by using glycol systems, but this adds to the complexity. These factors have been discussed in more detail by Lewis (1990). One major advantage of the batch system is its flexibility, that is, it is easy to change from one product to another. Also, provided the product is well mixed, there is no distribution of residence times (see Section 5.2.1.8.4).

An interesting question is whether HTST pasteurisation produces a better quality product that the holder process. Yale in 1933 (cited in Cronshaw, 1947) concluded that one method of pasteurisation is as good as the other when sound methods are used and when conditions are comparable. The authors are unaware of anything of late to contradict this, although most pasteurised milk is now produced by the HTST process. Homogenisation just prior to or after pasteurisation is simple in a continuous flow system. However, it is more difficult to link homogenisation with batch pasteurisation as the time delay between homogenisation and when the milk reaches pasteurisation temperature can result in an unacceptable amount of lipolysis (Deeth, 2002). However, this problem can be largely overcome by homogenising the milk at ~60 °C.

2.3.3.2 Continuous Heating

HTST pasteurisation permits the use of continuous processing, regeneration of energy and long run times. The main types of indirect heat exchanger for milk are the plate heat exchanger and the tubular heat exchanger. Plate heat exchangers (PHE) are most widely used for pasteurisation of milk, cream and ice-cream mix. They have a high overall heat transfer coefficient (OHTC) and are generally more compact than tubular heat exchangers. Their main limitation is pressure, with an upper limit of about 2 MPa. The normal gap width between the plates is between 2.5 and 5 mm but wider gaps are available for viscous liquids to prevent large pressure drops. In general, PHEs are the cheapest option and the one most widely used for low viscosity fluids. Maintenance costs may be higher than for tubular heat exchangers, as gaskets may need replacing and the integrity of the plates also needs evaluating regularly as pin-holes may appear in the plates of older heat exchangers. This may lead to pasteurised milk being recontaminated, for example, if such plates are in the regeneration section, a cracked or leaking plate may allow raw milk to contaminate already pasteurised milk. They are also more prone to fouling, but this is a more serious problem in UHT processing (see Section 6.2.2).

Tubular heat exchangers have a lower OHTC than plates and generally occupy a larger space. They have slower heating and cooling rates with a longer transit time through the heat exchanger. In general, they have fewer seals and provide a smoother flow passage for the fluid. A variety of tube designs are available to suit different product characteristics. Most tubular plants use a multi-tube design. They can withstand higher pressures than PHEs. Although they are still susceptible to fouling, high pumping pressures can be used to overcome the flow restrictions. Tubular heat exchangers give longer processing times than PHEs with viscous materials and with products which are more susceptible to fouling. Thus they may be used with more viscous milk-based desserts. They are also widely used in UHT processing of milk and milk products.

The viscosity of the product is a major factor that affects the choice of the most appropriate heat exchanger and the selection of pumps. Viscosity will influence the pressure drop causing a problem in the cooling section and when phase transitions such as coagulation or crystallization take place. For more viscous products or products containing particulates, for example, starch-based desserts or yogurts with fruit pieces, a scraped-surface heat exchanger may be required. Viscosity data for a range of milk products at different temperatures were presented by Kessler (1981).

One of the main advantages of continuous systems over batch systems is that energy can be recovered in terms of heat regeneration. The layout for a typical regeneration section is shown in Figure 2.1. The hot fluid can be used to heat the incoming fluid, thereby saving on heating and cooling costs. Regeneration efficiencies over 90% can be obtained.

In terms of the temperatures at different locations, the regeneration efficiency (RE) is given by:

$$RE = \frac{\theta_2 - \theta_1}{\theta_3 - \theta_1} x100$$

θ_1 = inlet temperature; θ_2 = temperature after regeneration; θ_3 = final temperature

Although higher regeneration efficiency results in considerable savings in energy, it necessitates the use of higher surface areas, resulting from the lower temperature driving force, and a slightly higher capital cost for the heat exchanger. This also means that the heating and cooling rates are slower, and the transit times longer, which may affect product quality.

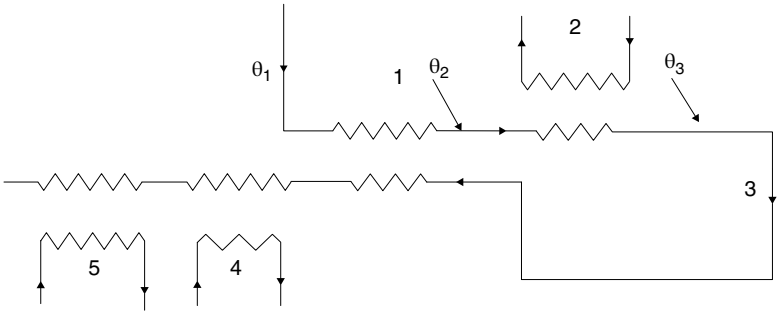

Figure 2.1 Heat exchanger sections for HTST pasteuriser: 1 regeneration; 2, Hot water section; 3, Holding tube; 4 mains water cooling; 5 chilled water cooling. (Source: Lewis, 1994. Reproduced with permission of Elsevier.)

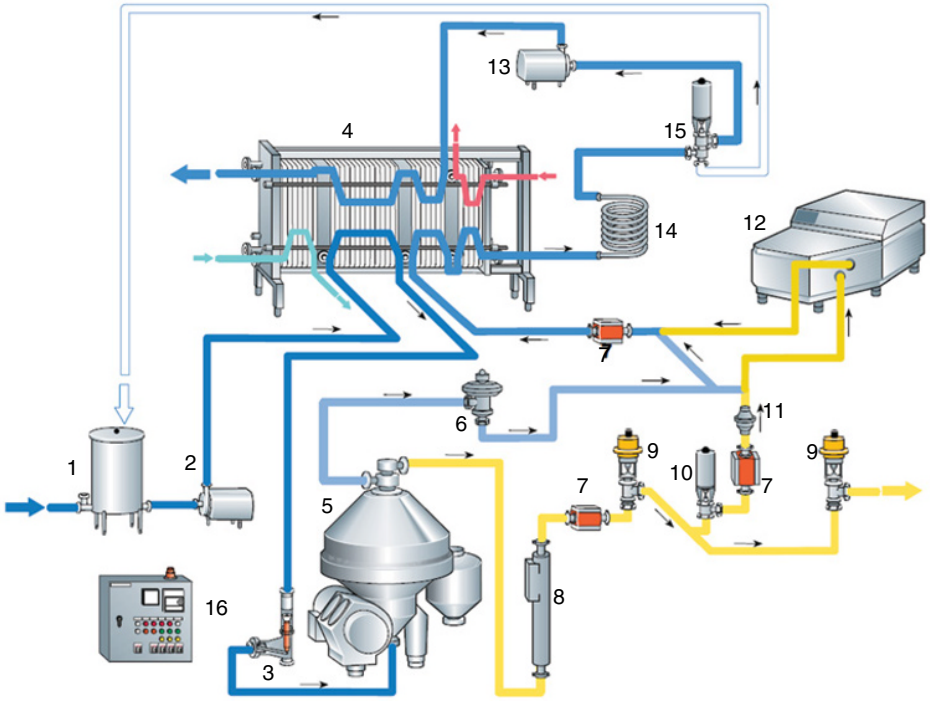

Figure 2.2 Production line for pasteurised milk with partial homogenisation. 1 Balance tank; 2 Product feed pump; 3 Flow controller; 4 Plate heat exchanger; 5 Separator; 6 Constant pressure valve; 7 Flow transmitter; 8 Density transmitter; 9 Regulating valve; 10 Shut-off valve; 11 Check valve; 12 Homogeniser; 13 Booster pump; 14 Holding tube; 15 Flow diversion valve; 16 Process control. (Source: Reproduced with permission of Tetra Pak.)

For milk containing substantial fat and for various cream products, homogenisation must be incorporated to prevent fat separation. However, as homogenisation of raw milk is a very effective way of initiating lipolysis (Deeth & Fitz-Gerald, 2006), it must be carried out immediately before or after pasteurisation, which inactivates the native lipase.

The layout of a typical HTST pasteuriser and its ancillary services is shown in Figure 2.2. The holding time is controlled either by using a positive displacement pump

or by a centrifugal pump linked to a flow controller, and the temperature is usually controlled and recorded. Note that a booster pump can be incorporated to ensure that the pasteurised milk is at a higher pressure than the raw milk in the regeneration section to eliminate PPC in this section. A flow diversion valve diverts under-processed fluid back to the feed tank. In continuous processing operations there will be a distribution of residence times, and it is vital to ensure that the minimum residence time, that is, the time for the fastest element of the fluid to pass through the holding tube, is greater than the stipulated time in order to avoid under-processing. In a fully developed turbulent flow, the minimum residence time is about 0.83 × average residence time (t_{av}). This will usually be the situation for milk, but it may be different for more viscous fluids. In this situation, the minimum residence time will only be $0.5 \times t_{av}$ and the distribution of residence times will be much wider (see Section 5.2.1.8.4).

Since most HTST pasteurisers are of the plate type, the plates themselves should be regularly tested for pinhole leaks, as discussed earlier. Consideration should be given to ensuring that if leaks do occur, they do so in a safe fashion, that is, pasteurised milk is not contaminated with cooling water or raw milk in the regeneration section. This can be achieved by making sure that the pressure on the milk side (downstream of the holding tube) is higher than on the water side, or on the raw milk side in the regeneration section. The control instrumentation, diversion valves and other valves should be checked regularly.

2.3.4 Process Characterisation

A number of parameters have been used to characterise heat treatment processes in order to allow comparisons of different temperature – time profiles. Two which have universal application are D-value and z-value. A parameter used specifically for pasteurisation is the pasteurisation unit (PU).

2.3.4.1 D-value

The time required at a particular temperature to reduce the bacterial population by 90%, that is, 1 log cycle or decimal reduction.

The number of decimal reductions is equal to the heating time divided by the decimal reduction time (*D*-value). For example, if the original bacterial count is 10^3 cfu/mL, the remaining count after various heat treatments will be as shown in Table 2.2.

Table 2.2 Remaining bacterial counts after heat treatments causing one to nine decimal reductions. Initial count = 10^3/mL.

		Remaining count	
Log reduction	**Percentage reduction**	**Per mL**	**Per L**
1D	90	100	100,000
2D	99	10	10,000
3D	99.9	1	1,000
6D	99.9999	0.001	1
9D	99.9999999	0.000001	0.001

The practical inference from this table is that the higher the initial count, the higher will be the count of remaining bacteria after heat treatment.

2.3.4.2 z-value

The change in temperature required to produce a tenfold change in the decimal reduction time (D-value).

The z-value is therefore the slope of the semi-logarithmic curve of D-value versus temperature.

The reported z-values for a range of pathogenic non-sporeforming bacteria that can contaminate milk are shown in Table 2.3 assembled from Juffs and Deeth (2007). Lovett *et al.* (1982) quoted the expected range of these z-values to be 5.56 ± 1.1 °C for heat treatments in the range 54.4 to 71.1 °C.

These z-values are lower than those for destruction of bacterial spores which, in turn, are considerably lower than those of chemical reactions (see Table 3.1). The practical significance of this is that the lower the *z*-value, the greater will be the effect of raising the temperature of processing. This is highly significant for UHT processing as it is the basis of one of the two fundamental principles on which UHT technology is based (see Section 3.4.2).

2.3.4.3 Pasteurisation Unit (PU)

One PU results from heating at a temperature of 60 °C (140 °F) for 1 min. The equivalent z-value is 10 °C (18 °F), which is high for vegetative bacteria (see Table 2.2 for z-values of

Table 2.3 z-values (°C) of selected pathogenic vegetative bacteria.

Bacterium	z-value
Brucella abortus	5.3, 4.7-4.8, 4.3-4.8, 4.4-5.5
Campylobacter jejuni	5.1, 4.94-5.6, 7.0, 8.0
Coxiella burnetii	4.4-5.5
Enterobacter (now *Cronobacter*) *sakazakii*	5.82, 3.1, 3.6, 5.7
Escherichia coli	5.07, 4.61, 3.2-3.4, 4.72
E. coli 0157:H7	1.8, 3.1
Listeria innocua	4.8-5.9
L. monocytogenes	6.1-7.4
Mycobacterium tuberculosis	4.4-5.6
M. bovis	4.8, 4.8, 4.9, 5.2
M. avium subsp. *paratuberculosis*	7.11, 8.6 (mean of 5 strains)
Salmonella Typhimurium	5.3
Staphylococcus aureus	6.04, 5.1, 4.4-6.7, 9.46, 4.83, 4.5
Streptococcus pyogenes	4.4-6.7
Yersinia enterocolitica	5.78, 5.22, 5.11

Data extracted from Juffs and Deeth (2007)

vegetative pathogenic bacteria). Thus, the number of pasteurisation units for a heating temperature (T,°C) and heating time (t, min) is given by:

$$PU = 10^{(T-60)/10}.t$$

Thus, a temperature of 63 °C for 30 min would have a value of approximately 60 PU (Wilbey, 1993), whereas HTST conditions (72 °C/15 s; originally 161 °F or 71.7 °C/15 s) would give only 3.96 PU. One might expect the values to be similar, and the discrepancy probably arises from the large z-value. Perhaps a lesson to be learnt is that it may not be meaningful to extrapolate this to continuous pasteurisation processes.

2.3.4.4 p*

Another parameter, introduced by Kessler, is p*. This is based on a reference temperature of 72 °C and a z-value of 8 °C. Processing conditions of 72 °C for 15 s are designed to provide a safe pasteurisation process for milk and are given an arbitrary p* of 1.

It can be calculated from:

$$p^* = \frac{10^{\frac{T-72}{z}}}{15}t$$

T = temperature (°C); t = time (s)

Figure 2.3 shows the time-temperature combinations that correspond to a p* value of 1 (normal pasteurisation) as well as other p* values (0.1 to 10).

This simplified equation ignores the contribution of the heating and cooling section. Both these factors provide an additional measure of safety (in terms of inactivating pathogens) but they may not help to extend shelf-life; they are further discussed by Kessler in IDF (1986). Knowledge of the heating and cooling profiles will enable their contribution to be determined. The procedure for this is, from the temperature – time profile, to plot p against time and determine the area under the curve. Alternatively, the activation energy (285 kJ/mol) can be used. As mentioned earlier, it is important to check the minimum residence time and that this exceeds the residence time required by regulations. Dye injection methods can be used to check this. It is also important to calibrate temperature probes at regular intervals. Note from earlier discussions that subjecting milk to higher p* than 1 will ensure that the milk is safe but it will not necessarily extend its shelf-life (see also Section 2.3.5).

It is worth reiterating that these different pasteurisation parameters make use of different z-values: If the holder process (63 °C/30 min) is considered to be equivalent to HTST conditions (72 °C/15 s), in terms of microbial inactivation, this would give those microorganisms a z-value of about 4.3 °C. It is probably not to be recommended to extrapolate PU from a batch to a continuous process, or p* from continuous to batch processes. The z values used are:

Comparing holder/HTST process	4.3
p*	8
PU	10

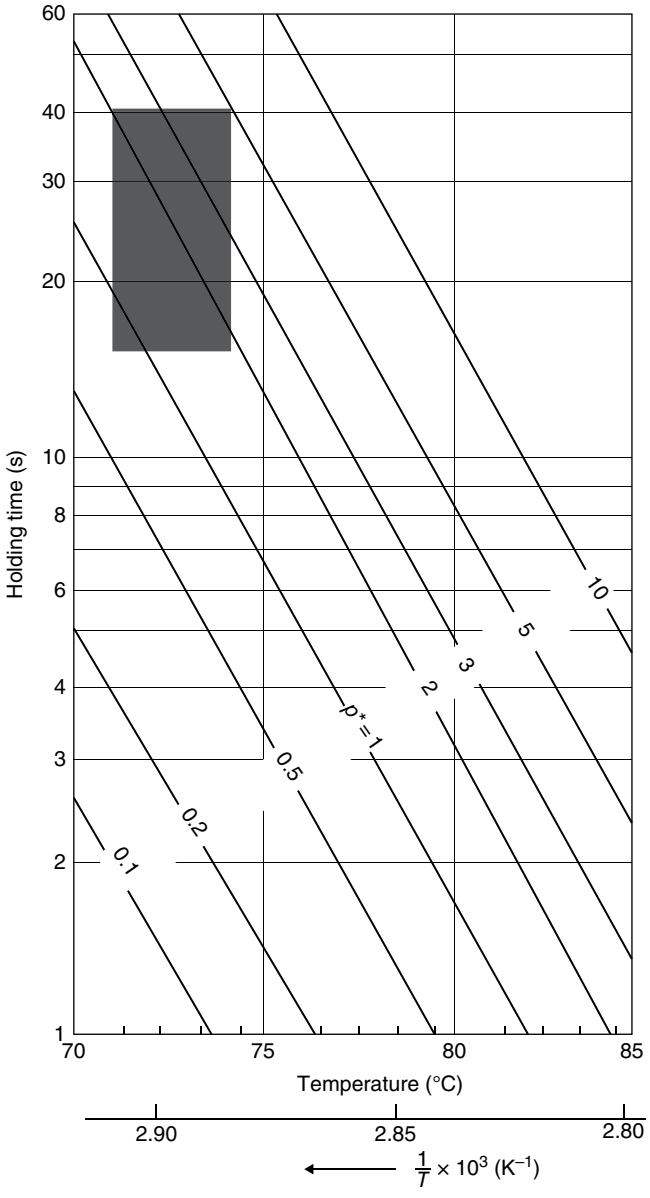

Figure 2.3 Temperature–time combinations required to give p* values in the range of 0.1 to 10. (Source: Kessler, 1989. Reproduced with permission of Elsevier.)

2.3.5 Processing Conditions

While the specified minimal pasteurisation conditions for milk are generally 72 °C for 15 s, in some situations they are exceeded. From the above discussion, it is not clear what the rationale is for selecting higher processing temperatures. It may be that they are being used to overcome problems related to poor quality milk. This is further discussed in the following section.

However, pasteurisation conditions do vary from one country to another. In the USA, a wide range of conditions are used including 63 °C for 30 min, 77 °C for 15 s, 90 °C for 0.5 s and 100 °C for 0.01 s. Similarly in Australia, a range of conditions are used for commercial pasteurisation of milk. Of 10 batch pasteurisation plants surveyed, the conditions ranged from 63 °C for 30 min to 80 °C for 5 min while of 61 HTST plants, most used conditions in the range 72-82 °C for 15 to 30 s; no holding times were <15 s (Juffs & Deeth, 2007).

Normal HTST pasteurisation conditions for milk are a minimum of 72 °C for a minimum of 15 s. As many processes are more severe than this, it is worthwhile considering the positive and negative effects associated with these more severe processing conditions. One surprising finding is that increasing the severity for pasteurisation of milk reduces its keeping quality at low temperature. This has been identified by Kessler and Horak (1984), Schroder and Bland (1984), Schmidt *et al.* (1989), Gomez Barroso (1997) and Barrett *et al.* (1998). It seems counterintuitive as it could be expected that a more severe heating process (e.g., at ~80-90 °C) would result in an improvement in keeping quality. Because these higher pasteurisation heat treatments do not result in milks with better keeping quality, a relatively new class of milk known as ESL milk which is mostly produced by heating at >120 °C has been developed and is commercially produced in several countries. ESL technology is described in Section 3.3.

The earliest explanation for the poorer keeping quality of milks subjected to these higher pasteurisation heat treatments was that the more severe conditions caused activation of the spores and that their subsequent germination and growth reduced the keeping quality. This is possible as it is known that such conditions activate *B. cereus* spores, some of which can grow at low temperatures (see Section 4.2.2.2.1). Another explanation is that there is reduced competition from other spoilage bacteria which would be destroyed at the harsher processing conditions. A third explanation is that the natural antibacterial lactoperoxidase system (LPS) in milk is inactivated. The LPS system involves the enzyme lactoperoxidase (LP), hydrogen peroxide and thiocyanate, all of which are present in raw milk. The oxidation products, for example, hypothiocyanite, exhibit strong anti-microbial activity by oxidising sulfydryl groups of bacterial cell walls (Reiter and Harnulv, 1982). The LPS system can be further activated in raw milk by small additions of thiocyanate and hydrogen peroxide and can be used to keep raw milk longer in countries where refrigeration is not widespread (IDF, 1988). LP inactivation is very temperature-sensitive with z-values of about 4 °C. Marks *et al.* (2001) showed that pasteurisation at 72 °C for 15 s resulted in active LPS remaining in the milk which greatly increased the keeping quality of milks inoculated with *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Str. thermophilus*, when compared to heating at 80 °C for 15 s.

The LPS also appeared to affect the shelf-life of pasteurised milk manufactured from raw milk which had been kept for 1 to 7 days at refrigeration conditions prior to pasteurisation (Ravanis & Lewis, 1995). The longest shelf-life was found for milk that was pasteurised after 3 or 4 days' storage and not for milk stored for 1 day. It was found that lactoperoxidase activity changed during storage of raw milk and was usually higher later in its storage period.

2.3.6 Changes During Pasteurisation

2.3.6.1 Microbiological Aspects

Raw milk from healthy animals has a very low microbial count, but it easily becomes contaminated with spoilage and sometimes pathogenic microorganisms. These need

to be inactivated and this is readily achieved by heat treatment. From a milk processor's standpoint, raw milk composition and its microbial loading will vary from day to day.

The primary reason for pasteurisation is to destroy the small numbers of pathogenic microorganisms in raw milk which may be picked up from the farm environment. These include non-spore-forming bacteria such as *St. aureus, Campylobacter jejuni, Salmonella* spp, *Escherichia coli* including *E. coli* O157:H7, *Yerisinia enterocolitica, Listeria monocytogenes, C. burnetii* and *M. tuberculosis,* together with spore-formers such as *Bacillus cereus* and *Clostridium* spp. An excellent review of pathogenic bacteria in raw milk was published by Griffiths (2010). The non-spore-forming pathogens can be effectively controlled by pasteurisation; according to Codex Alimentarius (2003), pasteurisation achieves a 5-log reduction of *C. burnetii*, so inactivation of the above non-spore-forming pathogenic bacteria will be greater. A summary of heat resistance data for various pathogens was provided by Lewis and Heppell (2000).

The secondary but very important microbiological reason for pasteurisation is to destroy spoilage organisms, particularly psychrotrophic bacteria which can grow and cause spoilage during storage of milk at low temperatures. *Pseudomonas* species are the major psychrotrophic bacteria in cold-stored raw milk but several other psychrotrophs also occur (see Section 4.2.1). Since these bacteria increase in numbers over time, raw milk should be processed as quickly as possible. The commercial reality is that some raw milk is pasteurised within 24 h of milking, but some may be up to one week old before it is pasteurised. In addition, it is important for the raw milk to be maintained at a low temperature, preferably ≤4 °C. In countries where it is not possible to refrigerate raw milk, its keeping quality can be extended by activation of the naturally occurring milk lactoperoxidase system. This has been demonstrated by Fweja *et al.* (2008) and discussed in recent reviews by the FAO/WHO (2006).

While pasteurisation destroys the pathogenic and most of the spoilage non-spore-forming bacteria in raw milk, some thermoduric bacteria remain. A count of the microbial flora in pasteurised milk determined soon after processing provides a measure of these thermoduric bacteria. High thermoduric counts are sometimes found in raw milk and these give rise to high counts in freshly pasteurised milk. Milk processors occasionally experience spikes in thermoduric counts, which are generally spasmodic and short lived. These most probably arise from brief lapses in hygiene during milking, poor temperature control or unfavourable climatic conditions. Thermoduric bacteria play a minor role in spoilage of pasteurised milk.

The thermodurics include non-spore-formers and spores of spore-formers which are more heat-resistant than the non-spore-formers; the spores all survive 80 °C for 30 min and some survive 100 °C for 30 min. Some non-spore-formers such as coryneforms also survive 80 °C for 30 min. The main non-spore-forming thermoduric genera are *Microbacterium, Micrococcus, Enterococcus, Lactobacillus, Corynebacterium* and *Streptococcus* and the main spore-forming genera are *Bacillus, Geobacillus, Paenibacillus* and *Clostridium.* While the thermoduric non-spore-forming bacteria are not pathogenic, some sporefomers are. Of particular interest are *Clostridium* species and some strains of *B. cereus*. These are further discussed in Section 4.2.2.

2.3.6.2 Enzyme Inactivation

Milk contains an abundance of indigenous milk enzymes. These have been extensively reviewed in Fox and McSweeney (2003) and Kelly *et al.* (2006); the heat stabilities of several of them were reviewed by Griffiths (1986) and Andrews *et al.* (1987). Some of

these enzymes have particular relevance to the heat treatments discussed in this book. These include alkaline phosphatase, lactoperoxidase, lipase and plasmin (alkaline protease).

The role of alkaline phosphatase has been discussed earlier in this chapter. The original phosphatase test for assessing the adequacy of pasteurisation was based upon the reaction of phosphatase with disodium phenyl phosphate. It was claimed to be able to detect the presence of about 0.2% raw milk (addition) in pasteurised milk, as well as under-processing, for example, heating at 62 °C instead of 63 °C for 30 min or 70 °C rather than 72 °C for 15 s. Since then, more automated tests based on fluorescence measurement (e.g., the Fluorophos® ALP Test System) have increased the sensitivity of the method, being able to detect the presence of 0.006% added raw milk. This is a very useful quality assurance test procedure as it detects low levels of post-pasteurisation contamination by raw milk, and helps to minimise the incidence of pathogens in pasteurised milk.

Griffiths (1986) determined the heat resistance of several indigenous milk enzymes. Alkaline phosphatase was the most heat labile of those measured ($D_{69.8} = 15$ s; $z = 5.1$ °C), compared to lactoperoxidase ($D_{70} = 940$ s; $z = 5.4$ °C). Acid phosphatase was much more heat resistant (about 100 fold) than alkaline phosphatase. Some discrepancies were also noticed between data obtained from capillary tube experiments and those obtained from HTST conditions using plate heat exchangers. Lactoperoxidase activity was thought to provide a useful indicator of over-processing. Activities determined on a PHE for 15 s were generally lower than those obtained from the capillary tube method. Using a PHE, lactoperoxidase activity was almost all destroyed at 78 °C for 15 s and completely destroyed at 80 °C for 5 s. The enzyme appeared sensitive to temperatures above 75 °C, with a z-value of 5.4 °C. Therefore, if milk is processed at or slightly above the specified minimum heating conditions of 72 °C for 15 s, it should test negative for alkaline phosphatase but positive for lactoperoxidase.

Ribonuclease was found to be more heat resistant than lactoperoxidase. Again there were discrepancies between laboratory studies and PHE studies. No loss of activity was observed at 80 °C for 15 s (lab), whereas a 40% loss of activity was found when using a PHE at 80 °C for 15 s.

Andrews *et al.* (1987) determined the following percentage retentions of enzyme activities in milk samples heated for precisely 15 s at 72 °C in glass capillary tubes; acid phosphatase, >95%; α-D-mannosidase, 98%; xanthine oxidase, 78%; γ-glutamyl transpeptidase, 75%; α-L-fucosidase, 26%; N-acetyl-β-D-glucosaminidase, 19%; and lipoprotein lipase, 1%. It was recommended that N-acetyl-β-D-glucosamiindase could be used for more detailed studies between 65 and 75 °C and γ-glutamyl transpeptidase (GGTP) between 70 and 80 °C. N-acetyl-β-D-glucosaminidase (NAGase) has been used as an alternative to somatic cell counts as a measure of mastitic infection (Kitchen *et al.,* 1978).

Patel and Wilbey (1994) recommend measuring GGTP activity for assessing the severity of HTST heat treatments above the minimum for whole milk, skim milk, sweetened milks, creams and ice-cream mixes. There was a good correlation between the reduction in GGTP activity, destruction of streptococci and water activity (Lewis, 1994).

Zehetner *et al.* (1996) investigated endogenous milk enzymes as indicators of heat treatment and concluded that α-fucosidase and phosphohexose isomerase were useful

for thermisation, phosphodiesterase and γ-glutamyltransferase were suitable for pasteurisation and α-mannosidase was an appropriate indicator for high-temperature-pasteurised milk.

Lipolysis by indigenous lipoprotein lipase produces free fatty acids which give rise to soapy, rancid off-flavours and reduce the foaming capacity of milk, an important consideration for making cappuccino coffee. This topic has been reviewed by Deeth (2006) and Deeth and Fitz-Gerald (2006). Lipolysis can be initiated on the farm or in the factory if raw milk is subjected to mechanical actions such as agitation or homogenisation which disrupt the milk fat globule membrane and allow access of the lipase to the fat. For example, this can happen when mixing flavoured milks or other similar products, using raw milk, at temperatures of about 50 °C before pasteurisation (Fitz-Gerald, 1974). Fortunately, pasteurisation destroys virtually all of the natural lipase in milk. If this did not happen, all pasteurised (homogenised) milk would have a high level of free fatty acids and taste rancid (Pearse, 1993).

Plasmin is an indigenous alkaline protease in milk. It is very heat-resistant and survives pasteurisation and even some higher-temperature treatments such as UHT processing. However, plasmin-related problems are not reported in pasteurised milk; this is probably due to the short shelf-life of pasteurised milk and the very low activity of plasmin at low temperature. Problems related to residual plasmin activity are more serious in UHT milk. The plasmin system in milk and its effects in UHT milk are addressed in Chapters 6 and 7.

Enzymes may also arise from psychrotrophic bacteria. Many of these are very heat-resistant and survive pasteurisation. However, it is unlikely that residual bacterial lipases and proteases will cause problems in pasteurised milk because of its relatively short shelf-life, the refrigerated storage conditions and the very low activity of the residual protease. In general, however, it is best to avoid using aged milk for pasteurisation because of the risk of its containing bacterial enzymes but also because of its higher microbial count, higher acidity (lower pH), reduced heat stability, and greater likelihood of having off-flavours.

2.3.6.3 Other Changes

There are some other important changes that take place during pasteurisation. As far as chemical reactions are concerned, pasteurisation can be considered to be a mild process. Between 5 and 15% of the whey protein is denatured in milk. This is not sufficient to significantly release levels of volatile sulfur compounds which cause the development of cooked flavour as occurs with higher temperature treatments (see Section 6.1.6.1). Whey protein denaturation is higher during pasteurisation of skim milk concentrates produced by ultrafiltration, increasing with the increase in the concentration factor (Guney, 1989). There is some suggestion that the holder process may cause slightly more whey protein denaturation than the HTST process (Painter and Bradley, 1996). Pasteurisation results in little change in the renneting properties of milk and little association of whey proteins with casein; as a result, good quality cheddar cheese can be produced from pasteurised milk and the majority of milk for cheesemaking is subjected to pasteurisation. No dephosphorylation and no significant reduction in pH and ionic calcium occur during pasteurisation and there is very little effect on the heat-sensitive water-soluble vitamins. Overall, pasteurisation results in little change in texture, flavour and colour, compared to raw milk (Deeth, 1986).

Wilson (1942) reported that it was clear that the majority of people are unable to distinguish between raw and pasteurised milk. Also, the difference in taste between different raw milks appears to be as great as or greater than the difference between raw and pasteurised milks. There is no evidence to suggest that this observation has changed over the past 65 years. However, as the sale of raw milk for human consumption is now prohibited in most countries, comparison of the flavour of raw and pasteurised milk is generally not possible. Nursten (1995) reported that pasteurisation barely alters the flavour of milk and that the volatile flavours responsible for cooked flavour are negligible.

2.3.7 Changes During Storage

2.3.7.1 Changes Due to Post-Pasteurisation Contamination (PPC)

PPC with Gram-negative psychrotrophic bacteria is the major cause of spoilage of pasteurised milk and is a very important determinant of the keeping quality of milk. Muir (1996a,b) describes how this became widely recognised for both milk and cream in the early 1980s, although Davis (1955) had drawn attention to this much earlier. PPC encompasses the recontamination of the product anywhere downstream of the end of the holding tube. It can occur in the regeneration or cooling sections, in storage tanks and in the final packaging of the product, due to poor hygienic practices. It is much reduced by ensuring that all internal plant surfaces in contact with the product are heated at 95 °C for 30 min before processing is commenced. It can only be completely eliminated by sterilising and employing aseptic techniques downstream of the holding tube. One of the main safety concerns is recontamination of the product with pathogens from raw milk; this could occur via by-passing of the holding tube by a number of possible routes, including pinhole leaks in plates. The presence in pasteurised products of high counts of microorganisms (e.g., coliforms and pseudomonads) which should be inactivated by pasteurisation is indicative of PPC. A number of different tests which can be used to determine the extent of the problem are catalogued in IDF (1993). In practical situations where the keeping quality of milk starts to deteriorate or is below expectations, the most likely explanation is an increase in PPC and this should be the first factor to be investigated. Bintsis *et al.* (2008) pointed out that the test for Enterobacteriaceae instead of coliforms is a more sensitive test for post-pasteurisation contamination, since the test detects all of the heat-sensitive, non-spore-forming, Gram-negative rods and provides good evidence that contamination has occurred. In this case the media used for the test must contain glucose instead of lactose.

2.3.7.1.1 Ropiness – a PPC Issue

In 2016, an interesting post-processing contamination of pasteurised milk resulted in the formation of ropy milk in the UK. The most striking feature was the production of thin strings when the milk was poured or when a spoon was placed on the surface and pulled away. The ropiness could be dispersed by mild agitation. We had not encountered this before and we later found that many experienced dairy colleagues had also never seen it. It had apparently been common when pasteurisation was first introduced but is much less common now. Davis (1955) reported that these defects occurred occasionally in pasteurised milk stored about a week and Hubbell Jr. and Collins (1962) reported that a few dairy plants in California had occasionally encountered ropiness in pasteurised milk. However, these authors commented that

improved equipment, upgraded cleaning methods, and better sanitizing procedures had reduced its incidence to a low level.

The milk also appeared to be more viscous and it had a distinct fruity aroma. Its pH was 6.50, compared to 6.69 for unspoilt pasteurised milk and its FPD was 535 m°C, compared to 513 m°C for unspoilt milk. However, its viscosity was lower than that of unspoilt milk, at 2.4 cP, compared to 2.6 cP. It was unstable in 70% alcohol, whereas unspoilt milk was stable in 80% alcohol. On centrifugation (3000 *G* for 30 min), there was no noticeable separation in the ropy milk and no noticeable difference to centrifuged unspoilt milk. As for its taste, it was still sweet, but slightly acidic, fruity with a slightly bitter after taste.

The best account of ropy milk was found in Davis (1955). One pertinent observation from that source was that ropy milk was of bacterial origin and the microorganisms involved would be inactivated by efficient pasteurisation. A small amount (1 mL) was added to 20 mL unspoilt milk and incubated at 5 °C and 20 °C to see if ropiness could be induced. The inoculated milk samples showed signs of ropiness after 24 h incubation at 5 °C and at 20 °C. The pH had fallen slightly and a fruity aroma was not apparent but the milk produced strings on pouring. This suggested that ropiness was bacterial in origin and that the offending bacteria grew well at 5 °C. Davis (1955) also mentioned that low temperatures also favour the formation of the slimy material by the bacteria. Thus, it was possible to produce ropy milk by inoculating a small amount into unspoilt milk, within 24 h in the refrigerator. It was also shown that it was inactivated by pasteurisation (63 °C for 30 min) and is therefore most likely to be a post-processing contaminant. In this case there were fewer than 20 consumer complaints about the product, so it was assumed that it was a very low level post-processing contamination, which was very quickly rectified. Nevertheless, it also interesting when something which is rarely seen, reappears. Coincidentally, another case of ropy milk was found in an ESL chocolate milk about one month later, from a completely different location in the UK.

In these recent cases, the bacteria responsible for the defect were not identified; however, several pyschrotropic bacteria have been reported to cause ropiness. These include *Alcaligenes* (Gainor & Wegemer, 1954, Samaras *et al.,* 2003, Morton & Barrett, 1982), *Escherichia intermedia* (Marth *et al.,* 1964), *Ps. aeruginosa, Klebsiella oxytoca* and others (Cheung & Westoff, 1983), *Acinetobacter* spp (Morton & Barrett, 1982) and *Bacillus aerogenes* and *B. lactis viscous* (Davis, 1955). The rope consists of long chains of bacteria, held together by their capsules. The capsules are polysaccharides which one report identified as levans which are polymers of fructose (Wegemer & Gainor, 1954).

2.3.7.2 Other Changes

It is instructive to outline the changes which lead to spoilage of pasteurised milk. These are illustrated by the results of Gomez Barosso (1997) who compared the shelf-life (days) for two batches of milk which were pasteurised at 72 °C for 15 s and 80 °C for 15 s and stored at 8 °C. The shelf-life was assessed by the aroma, tendency to form a clot on boiling, ethanol stability, titratable acidity, pH and dissolved oxygen, and the results were compared with those corresponding to a total viable count (TVC) of 10^7 cfu/mL. The results are summarised in Table 2.4. It is apparent that even when the TVC reaches 10^7 cfu/mL, the milk may still be acceptable by other criteria.