

Copyright is owned by the Author of the thesis. Permission is given for a copy to be downloaded by an individual for the purpose of research and private study only. The thesis may not be reproduced elsewhere without the permission of the Author.

Modelling primary proteolysis in cheddar cheese in commercial cool stores

A thesis presented in partial
fulfilment of the requirements
for the degree
of master of technology in Bioprocess Engineering at
Massey University

Richard Edmonds

2000

ABSTRACT

One issue identified as a possible problem during the manufacture of cheddar cheese is the possibility of producing a non-uniform product. It was proposed that a pallet of cheese experiencing different time-temperature histories, depending on the position within the pallet, could cause the heterogeneity. This work involved the investigation of that issue.

The level of primary proteolysis observed in cheese was measured over time in cheeses of different compositions, stored at different temperatures. The remaining intact α_{s1} casein was measured using reverse phase high performance liquid chromatography. Several trends were observed during maturation. High temperatures caused a faster rate of disappearance of α_{s1} casein. The temperature relationship followed Arrhenius law. High moisture content caused a faster rate of the disappearance of α_{s1} casein. The level of rennet added to the milk during production had a directly proportional effect on the rate of the disappearance of α_{s1} casein. Salt had no observable effect in the range investigated here. From the data a kinetic model was developed that described the rate of disappearance of α_{s1} casein in terms of the temperature, the moisture content, and the level of rennet in the cheese.

The heat transfer occurring in the commercial pallet of cheese was mathematically modelled and solved numerically. The heat transfer model was then applied to produce data describing the time-temperature profile throughout a pallet of cheese for a variety of possible industrial storage conditions. The kinetic model developed was then used to predict the extent of proteolysis in each case.

It was found that there would be significantly different levels of proteolysis within a pallet of cheese that had undergone chilling. A 10% difference in the level of proteolysis between the surface and the centre was observed after chilling for 40 days. During freezing the difference in the level of proteolysis after freezing was complete ranged from 10-25%. It was found that the heterogeneity was reduced during the thawing process and that the greatest reduction in non-uniformity was observed when thawed at lower temperatures.

ACKNOWLEDGEMENTS

It has been said that the search for knowledge is like a blind man searching for a black cat in a darkened room. I thought I heard it meow, but that may have been the walls closing in. As they say however: It's *all* good. It is true also, that this project would not have been possible without the help (and patience) of a number of people.

I would therefore like to thank the following:

- Dr J.E. Bronlund, Dr L.K. Creamer, C. Honoré for their supervision and guidance
- New Zealand Dairy Research Institute, for the research monies which made this work possible and for the use of their facilities in making and analysis of the cheese.
- Dave Alger, Don Otter, and Carmen Norris, without their assistance the HPLC work would have been impossible or least very much more time consuming and unsuccessful.
- Paul Mulligan, Hamish McCook, and Kiwi Dairies for providing industrial data collection opportunities.
- Harriet Gibbs for her input and assistance in experimental work
- Vanessa Schofield for her input and assistance in experimental work and analysis in particular her work involved in the creation of figure 3.3
- Tracy Loye, for her infinite patience and assistance in a great deal of the experimental sample preparations and support throughout the project.
- Last but not least (at least for some) family and friends, they know who they are and what they did. I haven't forgotten either... so watch it.

CONTENTS

Abstract.....	ii
Acknowledgements	iii
List of Figures.....	ix
List of tables	x

PROJECT OVERVIEW

1.1 Background and problem definition.....	1
1.2 Project Objectives.....	2
1.2.1 Specific Objectives	2

PROTEOLYSIS OF CHEDDAR CHEESE

2.1 Introduction	4
2.2 Proteolysis	4
2.2.1 Proteolysis as an indicator of ripeness.....	4
2.2.2 Types of proteolysis.....	5
2.2.3 Moisture content	6
2.2.4 Starter used	7
2.2.5 Rennet and enzymatic proteolysis	7
2.2.6 Salt content	8
2.2.7 pH.....	8

2.2.8	Temperature	9
2.3	Literature conclusions	10
2.4	Method development	11
2.4.1	Methods and materials	11
2.4.2	Chromatogram result	16
2.4.3	Data discussion	18
2.4.4	Kinetic Discussion	18
2.4.5	Modeling the rate of proteolysis of α_{s1} casein.....	27
2.4.6	Empirical regression of rate relation	29
2.4.7	Comparison of model to raw data.....	30
2.5	Proteolysis conclusions	32

THERMOPHYSICAL PROPERTIES OF CHEESE

3.1	Introduction	34
3.2	Thermal property data in literature	35
3.2.1	Ice Fraction	35
3.2.2	Initial freezing point	38
3.2.3	heat capacity	38
3.2.4	Thermal conductivity.....	42
3.2.5	Density	47
3.3	literature Conclusions.....	49
3.4	Experimental determination of ice fraction from specific heat data	50

3.4.1	Method development	50
3.4.2	Real system validation	55
3.5	Summary of thermal properties	58
3.5.1	Heat capacity	58

HEAT TRANSFER

4.1	Freezing and Thawing effects	60
4.1.1	Freezing	60
4.1.2	Thawing	60
4.2	Heat Transfer models	61
4.2.1	Heat transfer equations	61
4.2.2	Model solution technique	63
4.2.3	Literature conclusions	64
4.3	Model development	64
4.3.1	Model Purpose	64
4.3.2	Assumptions	66
4.3.3	Equation formulation	68
4.4	Model Solution	69
4.5	System input parameters	69
4.5.1	Size	69
4.5.2	Heat transfer coefficient	69
4.5.3	Initial Conditions	70
4.5.4	Time frame	70
4.5.5	Thermal conductivity Data	70
4.5.6	Volumetric Heat Capacity	72

4.6	Numerical error checking	73
4.7	Model Validation.....	73
4.7.1	Numerical solution data visualisation	73
4.7.2	Model Performance	77
4.7.3	Predictions	79
4.7.4	Conclusions.....	81

PRODUCT VARIABILITY IN A PALLET OF CHEESE DUE TO HEAT TRANSFER

5.1	Introduction	82
5.2	model formulation	82
5.2.1	Variables	83
5.2.2	Assumptions	83
5.2.3	Equation formulation.....	83
5.3	Model solution.....	84
5.3.1	Model checking	84
5.4	Model Application.....	85
5.4.1	Proteolysis in a pallet of cheese being chilled.....	85
5.4.2	Proteolysis in a pallet of cheese undergoing a freeze thaw cycle	88
5.5	Conclusions	95

CONCLUSIONS

APPENDIX

7.1	Cheese Compositions	100
7.3	Casein vs time complete data set	102
7.4	Matlab Scripts.....	126
7.4.1	Model input file	126
7.4.2	Model Output file	127
7.4.3	Proteolysis script file	128
7.4.4	Thaw data input file.....	132
7.4.5	Proteolysis Integration file.....	133
7.5	RADS input file.....	135
7.6	Ice fraction data	139
7.7	Heat capacity data	141

NOMENCLATURE

REFERENCES

LIST OF FIGURES

Figure 2.1 Typical chromatogram.....	16
Figure 2.2 Example raw data for vat 3 (high rennet)	17
Figure 2.3 Example raw data for vat 7 (low rennet)	18
Figure 2.4 Example log plot vat 3 (high rennet)	19
Figure 2.5 Example log plot vat 7 (low rennet)	20
Figure 2.6 First order reaction rates at -2°C vs salt	21
Figure 2.7 First order reaction rates at -2°C vs moisture.....	22
Figure 2.8 First order reaction rates at -2°C vs rennet.....	22
Figure 2.9 Arrhenius plots for vat 3 and vat 7	26
Figure 2.10 Arrhenius plot for all cheese vats (-2°C to 15°C)	26
Figure 2.11 Vat 3 prediction of remaining intact α_{s1} casein.....	31
Figure 2.12 Vat 7 prediction of remaining intact α_{s1} casein.....	32
Figure 3.1 Specific heat model data from Equation 3-29	54
Figure 3.2 Ice fraction from model data	55
Figure 3.3 Effect of scanning rate on cheese melting thermogram	56
Figure 3.4 Heat capacity - real cheese data.....	57
Figure 3.5 Ice fraction for vat 3	58
Figure 4.1 Cheese pallet.....	65
Figure 4.2 Thermal conductivity used in heat transfer model	71
Figure 4.3 Heat capacity used in heat transfer model.....	72
Figure 4.4: a: Cheese pallet (plan), b: Cheese pallet (side elevation), c: Thermocouple positions (plan)	75
Figure 4.5 Temperature-Time profile for a pallet undergoing chilling	75
Figure 4.6 temperature-time profile for a pallet undergoing freezing	76
Figure 4.7 Center/surface temperature for a slab undergoing freezing	77
Figure 4.8 Model performance for experimental chilling data.....	79
Figure 4.9 Model performance for experimental 1D freezing data	79
Figure 4.10 Model performance for experimental freezing data	80
Figure 5.1 Time-temperature profile for a pallet of cheese chilled to 2°C.....	86
Figure 5.2 Level of proteolysis in a pallet undergoing chilling.....	87
Figure 5.3 Variation in proteolysis in a pallet of cheese undergoing chilling	88

Figure 5.4 Time-temperature profile for freeze thaw cycle 12 to -15 to 12°C	89
Figure 5.5 Casein remaining for a low k_o cheese in a freeze thaw cycle.....	90
Figure 5.6 Variation of proteolysis in low k_o cheese undergoing a freeze thaw cycle ...	91
Figure 5.7 Casein remaining for a high k_o cheese in a freeze thaw cycle.....	92
Figure 5.8 Variation of proteolysis in two cheeses undergoing a freeze thaw cycle.....	93
Figure 5.9 Time-temperature profile for freeze thaw cycle 12 to -15 to 2°C	94
Figure 5.10 Casein remaining for a high k_o cheese thawed at 2°C.....	94
Figure 5.11 Variation in the level of proteolysis for different thawing regimes	95

LIST OF TABLES

Table 2-1 Experimental design table	12
Table 2-2 Experimental design result levels	13
Table 2-3 HPLC program	15
Table 2-4 Summary of first order rate constant for all cheeses	21
Table 4-1 Heat transfer initial temperatures	78
Table 4-2 Heat transfer simulation times.....	78
Table 5-1 Cheese compositions used for freeze/thaw simulations	90

CHAPTER 1

PROJECT OVERVIEW

1.1 BACKGROUND AND PROBLEM DEFINITION

New Zealand produced approximately 245000 tonnes of cheese in the 1997/1998 season (Johnston, Luckman, Lilley, & Smale, 1998). As it is such a large part of New Zealand industry it is important that the process is understood and optimised.

Cheese making is a dehydration concentration process. Cheese curd is produced from standardised milk by destabilising the casein micelles present in the milk causing them to aggregate and precipitate out of solution. Destabilisation is carried out by acidulation or an enzymatic reaction transforming kappa casein proteins into non-stabilising para-kappa casein. The casein gel formed is then cut and cooked causing it to release water present in the gel as “whey”. This leaves behind concentrated casein and milk fat that forms cheese curd. Once the whey is drained from the curd, it is dry salted to the desired level and cheddared. The curd is left until the pH reaches approximately 5.2 and then pressed. Once pressed, the cheese is put in chillers to mature, between 2-18 months for mild to tasty cheese respectively (Fox, 1989).

During maturation one of the reactions occurring is the proteolysis of α_{s1} -casein. As the protein is broken down this changes the functional properties of the cheese. The flavour and odour also change as breakdown products of the proteolysis process begin to appear. The rate at which the maturation occurs is a function of the cheeses composition. For example the salt content effects the rate at which enzymes can work. The rate is also a function of temperature (Law et al. 1979). In some applications the functional properties of the cheese are important. (e.g. processed cheese). As an ingredient some of the cheese used is required to have a low level of α_{s1} -casein breakdown in order to provide the desired functionality.

Production of cheese that has a low level of proteolysis is possible by manipulation of those factors that effect the rate of proteolysis. That is; adjusting the composition or temperature at which the cheese is stored. In addition to this, when

cheese is made it is packed into $\cong 1\text{m}^3$ pallets and then chilled or frozen. This leads to the possibility of non-homogeneous time-temperature histories depending on the position in the pallet. It is not known whether this could cause a significantly non-homogeneous product in terms of the level of proteolysis or functionality.

The challenge therefore is twofold. Firstly, to determine a method that can be used to estimate the ripeness of cheese given different compositions and chilling regimes. Secondly, characterise the extent of and offer possible solutions to the problem of a non-uniform cheddar cheese after storage.

1.2 PROJECT OBJECTIVES

The solution to the problem outlined above is the development of a method to control the rate of proteolysis in cheddar cheese. This requires the understanding of a number of things including:

- The cheese composition. The influence of cheese composition and how this may effect the cheese and its ripening properties. (i.e. the aim of this work is to determine the impact of salt, rennet, and moisture on rate of proteolysis in the early stages of proteolysis).
- Low temperature during the early stages of proteolysis. This includes an understanding of the effects of low temperatures and freezing on the rate of proteolysis. This understanding would allow prediction of the level of proteolysis for any given cheese under any given time-temperature profile and would allow for the prediction of maximum storage times for these cheeses in terms of the amount of remaining intact α_{s1} -casein.
- The extent of the heterogeneity of the level of proteolysis throughout a pallet of cheese caused by variations in the temperature time history. This can be achieved through modelling the heat transfer through a pallet of cheese and combining this with the understanding of the rate of proteolysis.

1.2.1 SPECIFIC OBJECTIVES

To achieve the understanding outlined above the following objectives were proposed.

-
- 1) Experimentally measure the rate of proteolysis in cheddar cheese by measuring the rate of disappearance of α_{s1} -casein with time. The reaction rate was to be described mathematically in terms of temperature and composition. This work has been covered in chapter 2.
 - 2) To collect data on the thermophysical properties of cheddar cheese in the chilling and freezing range 15°C to -40°C. The results of this investigation are given in chapter 3.
 - 3) To construct and validate a mathematical model to predict the time temperature relationship with position in a pallet of cheese undergoing chilling or freezing. This would use the thermophysical properties described in chapter 3. The formulation and testing of that model is presented in chapter 4.
 - 4) To combine the chilling/freezing model for a pallet of cheese produced in chapter 4 with the proteolysis work carried out in chapter 2. This was to allow the prediction of positional variability of the level of primary proteolysis in a pallet of commercial cheese and identification of possible ways to control this variability. This work is outlined in chapter 5.