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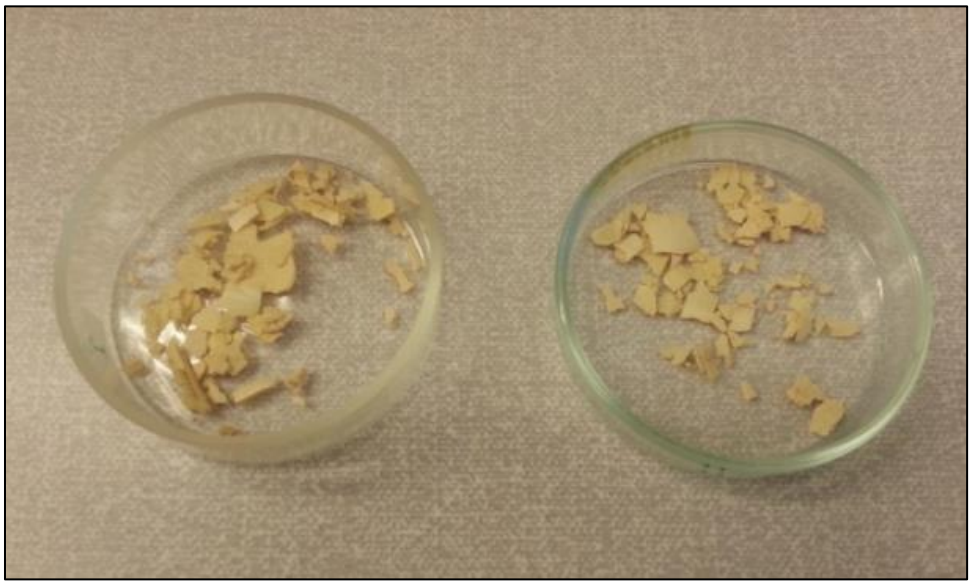

Method Development for Measuring Cleaning Agent Efficiency at a Laboratory Scale.

Cleaning agent efficiency in dissolution of UHT fouling at different concentrations and temperatures.

Master's thesis in Biotechnology

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Gothenburg, Sweden 2014

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Cover:
Picture showing dried UHT fouling placed on bottom half of glass petri dishes.

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Abstract

In the milk industry, the production must be stopped daily in order to clean the process equipment from deposits that are formed at the inside surface when the products are heat treated. This leads reduced production time since no products can be treated when the equipment are being cleaned. The heat treatment of milk products is necessary to ensure that the product is safe to consume. When heat treating the milk, certain compounds in the milk become unstable and form a deposit called fouling inside the heating equipment. This fouling layer reduces the heat transfer from the heat equipment to the product that is supposed to be heat treated. In order to remove the fouling deposit cleaning with first alkaline and then acidic detergent are used. It can take up to a few hours before the equipment is cleaned depending on the degree of fouling.

The aim of this master's thesis is to develop a laboratory method for measuring cleaning efficiency of UHT fouling by using cleaning detergents at different temperatures and concentrations. The method was created by looking on how a Cleaning-In-Place process is performed and imitates these steps at a laboratory level, by performing experiments on storage, drying, size and separation of fouling. The cleaning method was evaluated according to repeatability and ability to distinguish cleaning effects of the different cleaning liquids. A cleaning method for dissolution of UHT fouling was achieved where it was possible to detect trends of degree of dissolved fouling when different temperature and concentration of cleaning liquids were used in the sodium hydroxide step. The repeatability of the cleaning method was sufficient to be able to detect the cleaning trends. The cleaning effect trials showed that higher temperature and concentration leads to more fouling dissolved. It was also seen that with higher concentration of alkaline detergent a lower temperature could be used to achieve a similar cleaning compared to lower concentration of alkaline detergent.

Keywords: Fouling, cleaning, mineral, milk

Table of contents

1. Introduction

Heat treatment of dairy products is important to eliminate microorganisms that could spoil the product, produce a safe product for the consumer and achieve a prolonged shelf life for the product. During the heat treatment of milk products a deposit called fouling consisting of mainly proteins and minerals are formed at the surface of the process equipment. The fouling layer has an undesirable effect on heat treatment since it reduces the heat transfer from the heat exchanger walls to the product and increases the pressure drop inside the system (T.J. M. Jeurnink, Walstra, & de Kruif, 1996). The heat treatment of milk products are often performed with indirect heating at either pasteurization temperatures or in ultra-high temperatures (UHT), around 75°C and above 130°C respectively. At these temperature intervals it is distinguished that two different types of fouling are formed: protein rich fouling and mineral rich fouling (Burton, 1968).

The formation of fouling leads to that the production must be interrupted daily in order to clean the processing equipment from the fouling deposit. The cleaning is often performed by cleaning in place (CIP) where the equipment is first rinsed with alkali cleaning liquids followed by acidic cleaning liquids. With this method no time is required for dismantling of the process equipment, but also makes it hard to see when the surface is clean. To be sure that the surfaces are cleaned the cleaning processes are often performed with suboptimal cleaning liquid concentrations and temperatures. With optimization it could be possible to reduce the down time and reduce temperature and concentrations on the cleaning liquids which would do the cleaning process more environmentally friendly and production more cost effective. Around 80% of the total production costs in the dairy industry today are due to of fouling and cleaning (Bansal & Chen, 2006).

The cleaning of fouling formed at pasteurization temperatures has extensively been studied, where both formation and removal of this type of fouling has been investigated. Less research has been performed at fouling formed during UHT milk treatments and the cleaning efficiency of the cleaning liquids when used for cleaning UHT fouling. The efficiency in CIP is mainly dependent on four parameters: time, concentration, temperature and flow of the cleaning liquids (Lorenzen, 2005). In this master´s thesis the focus will be to develop a method for measuring the influence of concentration and temperature of the cleaning liquid for the dissolution of mineral rich fouling at a laboratory level.

1.1 Objective

The aim of this master's thesis is to develop a laboratory method to evaluate the dissolution (cleaning efficiency) of mineral rich fouling under different concentration and temperatures for the cleaning liquids.

1.2 Specification of the objective

- How could cleaning efficiency be measured and evaluated in a laboratory scale?
    - Weight change
    - Calcium concentration,
- How does the pre-treatment of the fouling affect the cleaning?
    - Drying and size
    - Storage
- What is the composition of the fouling?
    - Content of minerals and protein
- How do the cleaning liquid conditions influence the dissolution of fouling?
    - Temperature
    - Concentration

1.3 Delimitations

This project will only look at dissolving mineral rich fouling, formed during ultra-high temperature treatment. The internal structural change of fouling during treatment will not be investigated. Factors that are relevant for cleaning in place others than temperature and concentration of cleaning liquids will not be investigated. Fouling pieces scraped off from the fouling units will be used.

2. Theory

In the theory part the basic constituents of milk and their properties relevant for fouling and cleaning will be presented. The industrial heating methods commonly used for milk heat treatments will be shortly described. The basic understandings on how fouling are formed and built up will be described so that the cleaning methods can be understood.

2.1 Milk constituents

The cow´s milk is an aqueous solution with a pH of 6.5 - 6.7 at room temperature which consists of several different compounds like lipids, minerals, proteins, lactose and water (Bylund, 1995; Coultate, 2009). The weight percent of each constituent are shown in Table 1. The chemical composition of each compound will be described and further information will be presented for compounds of interest in the formation and cleaning of fouling. During processing of milk and other dairy products there will be formation of fouling in the processing equipment.

Table 1 Average composition of cow's milk in weight percent (Bylund, 1995).

Component	Concentration, wt%
Water	87
Fat	4
Lactose	4.7
Proteins	3.5
Minerals	0.8

2.1.1 Fat

The amount of fat in milk is around 4% and is mainly composed by triglycerides (98%) in a complex mixture and exists in milk as small fat globules (Bylund, 1995). Other lipids present in the milk are; phospholipids, cholesterol, free fatty acids, mono- and diglycerides (Walstra, Wouters, & Geurts, 2005c). Fat is the largest and lightest particle in the milk which makes it easy to separate the fat from the milk, if the fat is removed it is called skim milk (Bylund, 1995; Coultate, 2009).

2.1.2 Sugars

Lactose is the main carbohydrate in milk; it is composed of D-glucose and D-galactose. In milk around five percent of the total weight is from lactose (Walstra et al., 2005c). At high temperatures the lactose can react with protein in a Maillard reaction which forms a brown and caramel tasting compound (Bylund, 1995).

2.1.3 Protein

The proteins in milk can be divided into two major groups which have different properties in the milk, casein and whey proteins. These proteins also have a role in the fouling and the subsequence cleaning process. The typical casein and whey protein composition for cow´s milk are shown in Table 2.

Table 2 Typical protein distribution of skimmed cow´s milk (Coultate, 2009).

Protein	Concentration, wt%
Casein (total)	**80%**
α_S-Casein	31-45%
B-Casein	24-34%
K-Casein	3-5%
γ-Casein	1-1.5%
Whey protein (total)	**20%**
α-Lactalbumin	3.3-5.0%
β-Lactoglobulin	6.6-13.3%
Immunoglobulins	2.3-2.7%
Others	3-7%

The casein part of the milk is comprised by three major caseins; α_S-, β- and κ-casein which all has a fibrous structure that is relative hydrophobic and has a net negatively charge at normal pH in milk (Kelly & Bach Larsen, 2010; Walstra et al., 2005c). In the aqueous milk the caseins associates and form micelles which consist of several thousands of caseins. The casein micelles contain more than 95% of the casein in the milk (Kelly & Bach Larsen, 2010). Micelles are spherical with a diameter up to 600 nm but about half of the micelles are found to be in a size of 130-250 nm in diameter the rest is evenly distributed over and under this range. Casein micelles also contains a large amount of colloidal calcium phosphate, around 6% on dry basis (Coultate, 2009). The physical stability of milk during different treatments such as heating, concentrating and storage is to a large extent determined by the casein micelles (Walstra, Wouters, & Geurts, 2005a).

The structure of the casein micelles is very complex and not fully understood but a model of the casein micelle is proposed by Carl Holt and further developed by David Horne (Coultate, 2009). According to that model the micelle consists and is built up by a mix of submicelles. The submicelle contains 20 to 25 casein molecules and each submicelle is at a size of 12 to 15 nm. The nanocluster around 3 nm in size is constituted of calcium phosphate and most of the colloidal calcium phosphate present in the casein micelle is also located in the nanocluster which prevents precipitation within the micelle. The calcium phosphate nanoclusters are linked to α- and β-casein through a cluster of phosphoserine groups (esters of serine and phosphoric acid). This interaction forms a stable three dimensional network. The "hairy layer" at the surface of the micelle (Figure 1) consists of κ-caseins which only have a single phosphoserine group and a C-terminal end which is hydrophilic and negatively charged which leads to that it is only able to integrate to the network via the N-terminal end. The hydrophilic C-terminal ends will then prevent expansion in that region and give the characteristic "hairy layer" on the surface of the casein micelles. This layer is an important part to provide colloidal stability. The forces that keep the submicelles together during normal conditions in milk are hydrophobic bonds between proteins and cross-links between peptide chains by the nanoclusters (Coultate, 2009; Walstra et al., 2005a). A schematic cross-section figure of the model is shown in Figure 1.

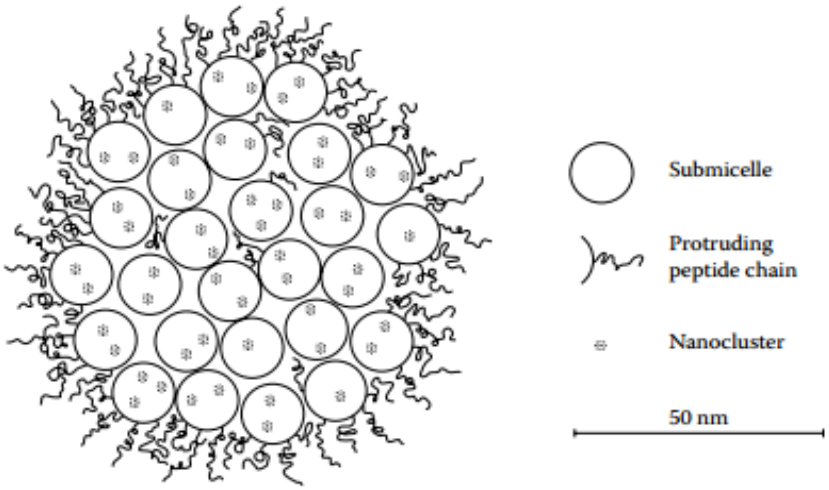

Figure 1 Schematic cross section for the model proposed by Horne and Holt (Walstra et al., 2005a).

The casein proteins are important as a transport system for calcium and phosphate, without this large insoluble crystals of calcium phosphates would be formed which could cause problems for secretion and digestion (Coultate, 2009).

The whey protein also called serum protein is consisted of α-Lactalbumin (α-LA), β-Lactoglobulin (β-LG), immunoglobulin and serum albumin. Whey proteins constitute to 20% of the total proteins in milk, where β-LG is most abundant (Bylund, 1995). The β-LG contains five cysteine residues and all of these has one –SH group, of these five –SH groups four of them are bonded into S-S bridges while the fifth is sheltered inside the structure under normal temperatures. The β-LG is the most sensitive protein in milk to alteration in temperature and pH (Visser & Jeurnink, 1997). Little is known about the biological function of these proteins in milk other than that they have a high nutritional value (Bylund, 1995; Coultate, 2009).

2.1.4 Minerals and salts

The salt constitutes of about 1% of the milk. The most important salts are calcium, potassium, sodium, magnesium, chloride and phosphate (Table 3). The salts can be distributed between the serum phase and the casein micelles in milk. The casein micelles can hold undissolved salts as well as colloidal calcium phosphates and counter ions to the negatively charged caseins. This leads to a complex distribution of the salts where the salts can be associated with the casein micelle (undissolved salts) but also be dissolved in the serum (Walstra et al., 2005c).

Table 3 Average concentration in weight percent of important salts in cow's milk (Walstra et al., 2005c).

Compound	Concentration, wt%
Sodium	0.05
Potassium	0.15
Calcium	0.12
Phosphate	0.20
Chloride	0.11
Citrate	0.17

2.2 Methods for heating of milk

The most common process in food processing plants are heating and cooling of food products. This involves heat transfer between product and heating/cooling medium. The reasons for heat treatment of milk is to kill pathogenic microorganism present in the milk, but also to inactivate enzymes and other microorganisms and this prevent microbial and enzymatic degradation (Bylund, 1995; Singh & Heldman, 2009).

The heat treatment process is performed in a unit called heat exchanger. The heat exchanger can be of two types either indirect heating or direct heating. In direct heating the product comes in direct contact with the heating medium. In indirect heating the product and the heating media are physically separated by a thin wall. Two of the most common indirect heat exchangers are shortly described below (Singh & Heldman, 2009).

- **Plate Heat Exchanger**: Consists of a series of parallel, closely spaced stainless-steel plates in a tight frame. Liquid product are flowing on one side of the plate and heating media on the other side. Heated surfaces in milk production can cause deposition of solid material on the surface which decreases the heat transfer rate from heating media to the product (Singh & Heldman, 2009).
- **Tubular Heat Exchanger**: Consists of a double-pipe where one pipe is located inside the other pipe and product are flowing in one pipe and heating medium in the other (Singh & Heldman, 2009).

2.3 Fouling

Fouling is the solid deposition formed at the surface on the heat exchanger equipment during processing of a product. The formation of fouling is unwanted since it reduces the heat transfer rate due to that fouling has low thermal conductivity and when the fouling layer increase a pressure drop in the system occur (Awad, 2011; Bansal & Chen, 2006). Reduced heat transfer in the heat exchanger leads to that more energy is demanded to achieve sufficient pasteurization or sterilization process.

Fouling deposits are formed in both pasteurization and UHT treatment processes, but the composition of the fouling formed differs. The composition of the fouling deposits also differs from the composition of cow´s milk (Theo J. M. Jeurnink, 1992)). It has been commonly accepted that two main types of fouling are formed; type A and type B. The type A fouling also known as protein rich fouling deposit is formed at pasteurization and preheating temperatures of 75°C up to 110 °C. The protein fouling deposit is white, soft and spongy and has a composition of 50-70% proteins, 30-40% minerals and 4-8% fat (Bansal & Chen, 2006). The Ca/P ratio in the fouling is ~1.5, which indicates presence of tricalcium phosphate (Changani, Belmar-Beiny, & Fryer, 1997; Lalande, Tissier, & Corrieu, 1984). The major protein present in the deposit is β-LG which are the most heat sensitive protein (Visser & Jeurnink, 1997). Type B fouling or mineral rich fouling is formed at UHT treatment temperatures, 110°C -140 °C. This deposit composition is 70-80% minerals, 15-20% proteins and 4-8% fat (Burton, 1968). The mineral consists of around 80% calcium phosphates (Lalande et al., 1984). The protein fraction consists mainly of β-casein (50%) and α-casein (27%) (Visser & Jeurnink, 1997). The mineral fouling is hard, compact, and granular in

structure and greyish in color(Changani et al., 1997). The molar Ca/P ratio in mineral fouling has experimentally been found to be around 1.2-1.35, which would indicate that crystallites of calcium phosphates has been formed in the deposit (Burton, 1968; Foster & Green, 1990; Lalande et al., 1984).

One of the most abundant proteins β -LG is also one of the most heat sensitive, in temperatures over 65 °C the β-LG will start to unfold and denature. The unfolding will lead to the previously buried –SH group will be exposed and reactive. The activated β -LG can form S-S cross-links with other β-LG or other proteins that has a –SH group available. This leads to formation of protein aggregates in the bulk phase of the liquid that may be transported to the surface of the heating equipment and form a deposit. The aggregates can contain associated casein micelles and calcium phosphate (Xin, Chen, & Özkan, 2002).This is the proposed reason for how formation of protein fouling starts (Figure 2), it should be stated that a WCP model was used.

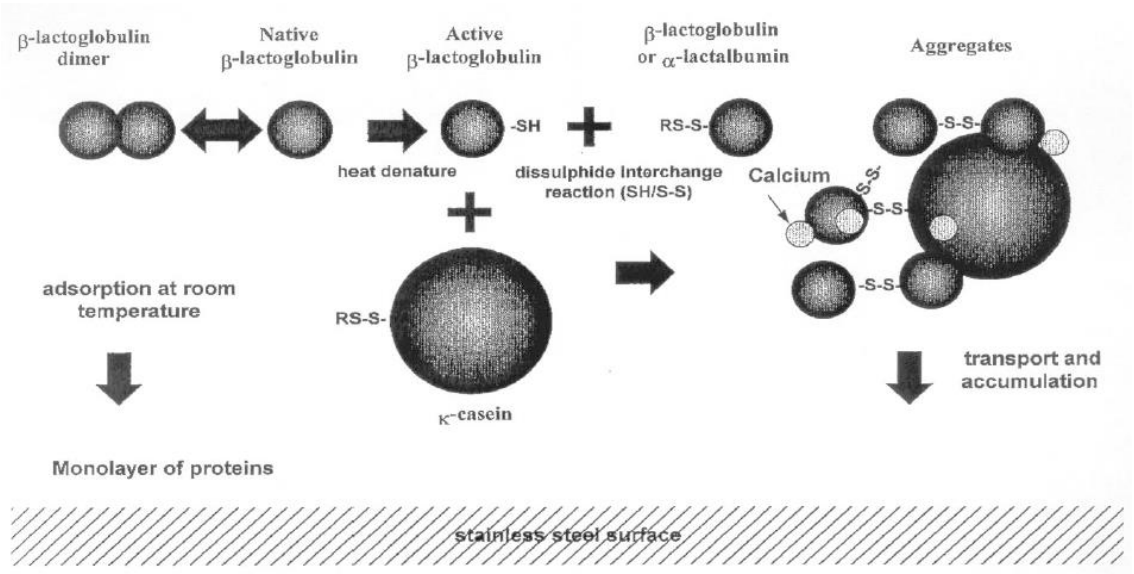

Figure 2 A shematic view of the mechanism of protein deposit built up. (Xin et al., 2002).

In mineral fouling the β-LG content is minor and the major protein constituent is caseins (Changani et al., 1997). However, it is not clear what the caseins role in the mineral fouling is. The higher casein content in mineral fouling can be due to lowering of the pH in milk at increasing temperatures which could decrease the colloidal stability of the casein micelles and that the most β-LG has been deposited in the preheater or aggregated to such size that they are transferred away with the flow (Foster & Green, 1990). The higher content of minerals like calcium phosphate can be due to the lower solubility of the salts when temperature is increasing (Burton, 1968; Changani et al., 1997). The decrease in pH during heating could make the calcium phosphate to be removed from the disturbed micelles, but precipitated again due to low solubility with the rise in temperature (Foster & Green, 1990) . Caseins are suggested to be in the fouling either via the β-LG process described previously or when the colloidal stability is reduced (Theo J. M. Jeurnink & Brinkman, 1994). This induced

instability of the casein micelles could be an explanation of why the deposit composition is changing with temperature.

Investigations on the structure of the mineral fouling layer has shown that fouling layer closer to the heating surface consist of more minerals and that protein is mainly present in the outer layer (Foster & Green, 1990). Even though the fouling layers closest to the heating surface contain mostly minerals, there is still discussion on what that adhering first to the heating surface. The main theory is that protein deposit first and minerals diffuse through the proteinaceous layer. Studies performed by Belmar and Fryer using whey proteins models shows that first a protein film is formed. After a short period calcium was detected and then just after one hour calcium phosphate was detected in the fouling which support the theory about protein being first to adhere (Changani et al., 1997). Foster and Green studied fouling formed at 140°C and suggested that minerals can diffuse through the deposit and that the minerals and protein layer are built simultaneously (Foster & Green, 1990).

The deposit of protein and mineral fouling are believed to be formed via different mechanisms. The mineral fouling is proposed to follow a three stage process where first the whey protein are denatured by the heating and forms aggregates with other proteins and adhere to the surface. Then when temperature increases more calcium phosphate becomes insoluble and precipitates to the surface where it forms the mineral deposit. In the third stage different layers are formed depending on the fluid composition (Changani et al., 1997). The fouling process theories involve both reactions in the bulk and at the surface, where the surface reaction is the final step. But it could be that the rate limiting step is reactions that occur in the bulk. Three mechanisms have been presented to be probable to be involved in transferring fouling compounds to the surface. The first is that the temperature gradient formed between the heating surface and liquid close to the wall causes reactions to occur faster at the surface. This leads to a concentration gradient near the wall which drives more compounds to the wall (T.J. M. Jeurnink et al., 1996). The second mechanism involves reactive molecules and particles that have been formed in the bulk are transferred to the surface via diffusion. Continuous supply of products leads to that after prolonged runtimes there is a deposit formed. This could explain why there are fouling formed also in sections of the product plant where the temperature of the wall and liquid is the same (Walstra, Wouters, & Geurts, 2005b). The third mechanism is that presence of air and vapor bubbles strongly enhance the fouling deposition. The air bubbles can be formed at the heating surface when the milk is heated and the air dissolves. If the bubbles remain at the surface particles will adsorb to the gas/liquid interface, especially casein micelles. The bubble will cause water to evaporate and later the bubble will collapse with the concentrated particles, which will deposit at the heating surface (T.J. M. Jeurnink et al., 1996).

There are several factors and processing conditions that are affecting the fouling in the heat exchangers the most important of them are shortly described: (Walstra et al., 2005b)

- **Temperature**: Below temperatures of 60°C very little to none deposit is formed (Walstra et al., 2005b).
- **Preheating:** Formation of protein fouling is mainly caused by denaturation of β-Lactoglobulin. Preheating can be used to denature the β-LG before UHT treatment and this will decrease the fouling significant in the heat exchanger (Walstra et al., 2005b).
- **Acidity:** Fouling deposits faster when the pH of the milk decreases. This is believed to be caused by the lower solubility of the denatured whey proteins. Meanwhile the calcium phosphate content in fouling during lower pH is reduced this is caused by the higher solubility of the calcium phosphate with lower pH (Walstra et al., 2005b).
- **Cold storage:** If the milk is stored cold the fouling deposits is reduced during heat treatment there is no know explanation for this (Walstra et al., 2005b).
- **Other milk properties:** Enzymatic degradation of protein in the milk can also induce or enhance the fouling deposits (Walstra et al., 2005b).

2.4 Cleaning

In order to remove fouling deposits from the heating surface a cleaning of the process equipment is required; this is usually performed with a cleaning in place (CIP) method. The main advantage of using a CIP method is that no dismantling of the process equipment is necessary, which was needed before when manual cleaning of the equipment was performed. So when comparing these methods, CIP saves both time and money (Walstra et al., 2005b). The CIP method builds on the concept to minimize the deposition mass flux and maximize removal mass flux. This is achieved by changing the product which causes the fouling (milk) into a cleaning solution. The cleaning solutions then alter the fouling layers together with increased temperature and mechanical forces which helps in the dissolving and removal of the fouling from the surface.

Cleaning of the process equipment is performed daily in the dairy industry and a summary of a typical CIP cleaning sequence used are presented in Table 4.

Table 4 An general example of a cleaning procedure used for removal of fouling(Bylund, 1995).

Cleaning step	Purpose
Prerinse with water	Remove non-deposited material
Cleaning with NaOH	Dissolve and remove proteinaceous fouling
Rinse with water	Remove alkali cleaning agent and dissolved fouling
Cleaning with HNO_3	Remove deposited mineral fouling
Rinse with water	Remove acid cleaning agent and dissolved fouling
Disinfection or Sterilization	Microbial load reduced to a low level
Final rinse	Remove sanitizer (not needed for sterilization)

A thorough *prerinsing* with water will remove 80% to 90% of the non-deposited residual material. To reduce the loss of product and excessive waste water production the milk residues should be removed before prerinsing (Walstra et al., 2005b). Depending on the process the cleaning step can differ a bit, in milk cleaning processes the common way is to first apply an alkali such as sodium hydroxide which should dissolve the proteinaceous part of the fouling. Water is used to remove the alkali cleaning solution before the acidic solution is applied; often nitric acid or phosphoric acid is chosen. The two-step cleaning is performed after heating processes where serious fouling deposits has been formed (Walstra et al., 2005b). A *final rinse* with water is needed after cleaning steps to remove cleaning residuals and cleaning liquids, so there is no cleaning residues left when a new batch of product are heat treated.

The cleaning mechanics of the cleaning process is yet not fully understood but can be viewed as an heterogeneous reaction between cleaning solution and the fouling layer, the reaction can be divided into two parts the mass transport and reaction processes (Plett & Grashoff, 2006). The cleaning detergent must be transferred to the surface of the fouling deposits through laminar boundary layer, which will be due to turbulence or concentration gradient. The cleaning detergent then come into contact with the fouling deposits surface where reaction starts and penetration into the fouling begins. The penetration into the fouling deposits is believed to be driven by either molecular diffusion or capillary action depending on the structure of the fouling (Plett & Grashoff, 2006). The smaller reaction products from the fouling and cleaning detergent are then transferred across the boundary layer into the bulk solution of the cleaning detergent in use forced by concentration gradient or turbulence. Meanwhile larger pieces can be detached by shear force after reactions has weakened the soil-soil or soil-surface bonds (Changani et al., 1997; Plett & Grashoff, 2006). The degree of effectiveness of a CIP cleaning is depending on four parameters time, concentration, flow and temperature (Lorenzen, 2005).

- **Time**: Depending on production different times is needed to achieve a clean result. (Walstra et al., 2005b). Producers want to be sure that the equipment is clean, so the cleaning process is often run longer than necessary
- **Temperature**: The temperature has significant impact on the cleaning efficiency which the cleaning liquid is producing. Since the chemical reactions can proceed more rapidly at higher temperatures also the viscosity of the liquid becomes lower at higher temperatures, hence Reynolds number and diffusion coefficients become larger. There are some limitations temperatures of cleaning detergent. Higher temperature than the temperature used for the product are not recommended since increased denaturation of proteins can occur (Plett & Grashoff, 2006).

- **Flow**: A turbulent flow in the tubes is important to achieve sufficient shear stress to induce cracks in the fouling which helps in the removal of fouling and delivery of detergents into the fouling. A common recommendation is that a velocity of 1.5 m/s is enough to have a turbulent flow, and higher turbulence leads to thinner laminar boundary layer which leads to increased overall reaction. The flow contributes to transfer cleaning detergents to the fouling surface, initiate cracks and transfer fouling residues away from fouling layer (Walstra et al., 2005b).
- **Concentration:** Higher concentration of detergent must not necessarily mean that the cleaning efficiency is better. In protein rich fouling it has been found that a concentration of 0.5% sodium hydroxide was optimal. And concentration higher than 0.5 wt% showed that the NaOH caused a glossy film over the fouling surface which prevented the mass transfer of cleaning detergent. Also in the acid cleaning step too high concentration can lead to corrosive action of the metal surface. The optimum concentration for UHT fouling has not been found (Visser & Jeurnink, 1997).

2.4.1 Cleaning of fouling with alkali

The alkaline cleaning detergent is used because it allow for dissolution of the protein in the fouling. The process of the cleaning of protein fouling can be divided into three stages: Initial swelling stage, uniform stage and final decay stage (Xin et al., 2002). Most of the data for describing alkaline cleaning are from experiments with whey proteins models. In the initial swelling stage swelling occur when the alkaline detergent has reached into the matrix of the fouling, this is caused by rearrangement of the original structure and forms void structure which in its case causes a concentration gradient and water to be drawn into the void matrix. In the second step, uniform stage, the fouling layer starts to crack and pieces of fouling are removed from the area until, after a while the decay stage is reached where the dissolution almost stops, and there is only a thin layer of mineral rich fouling left on the heating surface. Bird and fryer found that an optimal concentration for sodium hydroxide in protein fouling removal was 0.5 wt% and that temperatures above 50°C increased the cleaning efficiency (Bird & Fryer, 1991).

2.4.2 Cleaning of fouling with acid

Acid cleaning detergents are used for the removal of the mineral part of the fouling, and often applied after the alkali step where only a thin layer mineral layer is left at metal surface. This thin layer is important to be removed since it can act as nucleation area for new fouling (Bird & Fryer, 1991). Acid detergents such as nitric acid can transform insoluble minerals into water soluble minerals (Plett & Grashoff, 2006). An example reaction is shown in Figure 3 below.

$$Ca_3(PO_4)_2 + 4HNO_3 \rightarrow Ca(H_2PO_4)_4 + 2Ca(NO_3)_2$$

Figure 3 Example reaction of nitric acid transforming water insoluble tricalcium phosphate into water soluble salt (Plett & Grashoff, 2006).

The settings for a nitric acid solution used in cleaning are recommended to be a concentration between 0.5 wt% – 1 wt% and temperature of 70°C (Bylund, 1995).

2.4.3 Formulated cleaning detergents

If only a single stage cleaning processes are performed formulated cleaning detergents are used. The formulated cleaning detergent contains wetting agents, surface agents and chelating compounds in a mixture to perform the cleaning. Formulated cleaning detergents would allow for shorter cleaning times because only one cleaning step is needed but the cleaning chemicals are more expensive (Changani et al., 1997). Formulated alkali solutions is often used in two stage cleaning aswell to achieve a good contact with the fouling layer, a wetting agent is added to lower the surface tension of the solution and chelating agents for prevention of re-deposition of dissolved compounds (Bylund, 1995).

3. Experimental

The experimental part is divided into three main areas Production and analysis of fouling, Preparation of cleaning liquid and Basic cleaning method.

3.1 Production and analysis of fouling

To produce fouling a method from Hagsten et al. was used where two set-ups exist one for production of fouling and one for cleaning, for this master's thesis project only the production part of the method was used (Hagsten et al., 2013). The production part of the set-up is located in-line at Arla Foods dairy plant in Esbjerg, Denmark. Where the equipment is constructed so that indirect heating of low pasteurized skim milk can be performed at both UHT and preheating temperatures. The equipment is divided into two fouling units one connected after the preheater and the other connected after the final heater, with temperatures of 100°C after the preheater and 137°C after the final heater. The fouling is formed inside six rectangular units (Figure 4), which can be mounted into one test unit.

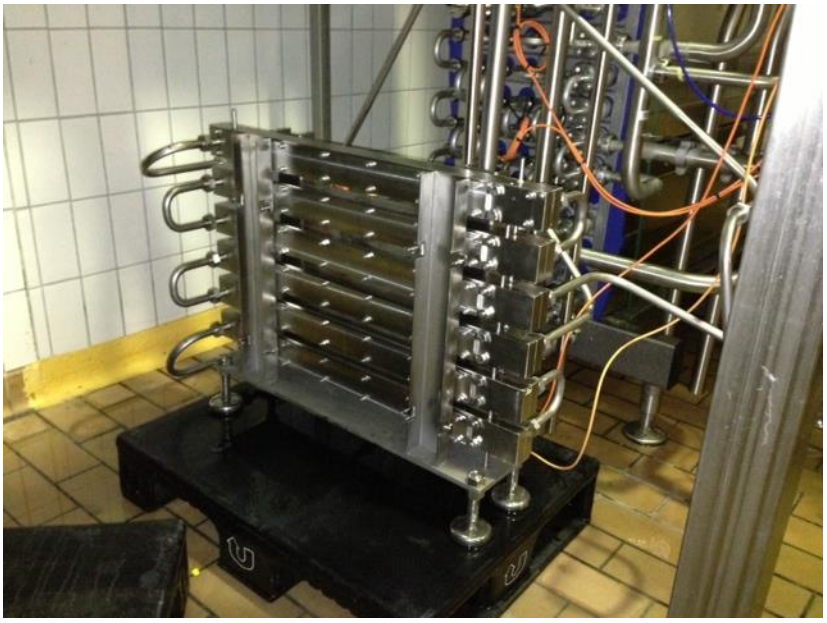

Figure 4 The six fouling units mounted on top of each other into one test unit. Test unit is connected to a heat exchanger present at Arla Foods in Esbjerg.

Each of these six units has 20 coupons that could be removed and used in the cleaning pilot plant equipment but that was not done in this project. Indirect heat treatment of skim milk was performed for 11 hours. The composition of the skim milk used in the heating process is presented in Table 5.

Table 5 Composition of the skim milk used in the production of fouling. Presented in volume percent.

	Fat, v%	Protein, v%	Dry matter, v%	Lactose, v%
Skim milk	0.07	3.52	9.29	4.88

After transportation of the fouling units to SIK it got dismantled so the fouled surfaces could be reached. The areas outside of the coupons were carefully scraped off so pieces of fouling could be gathered and put in a container either UHT or Preheat depending on which unit the

fouling was scraped off from. The two containers were stored in room temperature with a small opening for air to allow for the fouling to dry and prevent mould growth.

To be sure that the fouling produced in the pilot plant was of the correct composition to be categorized into protein rich or mineral rich fouling a chemical analysis was performed. Tests were performed on three samples randomly taken from the UHT part of equipment that would be expected to be mineral rich fouling. One sample was collected from the preheating section of the fouling equipment that would be expected to be similar to protein rich fouling. All samples were dried in room temperature and 1 gram of each sample was selected and put in plastic petri dishes and sent to Ekologihuset at Lunds University for protein and mineral analysis.

The protein analysis was performed by measuring the nitrogen content, the fouling samples was dried at 50°C for 24 hours and then put in a Vario MAX where an elemental analysis was performed. From the nitrogen content the total protein content was calculated by using a protein conversion factor of 6.38 according to the Kjeldahl method (Theo J. M. Jeurnink & Brinkman, 1994) The mineral content was defined to be the content of calcium, phosphorous and magnesium. The samples was dried at 50°C for 24 hours and then dissolved by microwave digestion in high concentrated HNO_3 at high temperature and high pressure. The concentration of the dissolved minerals was detected and measured by using an Induced coupled plasma- atomic emission spectroscopy.

The fouling produced had some differences in appearance depending on if it was made in the UHT section or in the preheater section of the fouling production plant. The UHT fouling was a bit grey–yellow in color and gritty structure except at the surface that was in contact with the heating wall that surface was smooth and white. The fouling from preheater fouling had a more white color and voluminous look.

The chemical analysis was performed in order to verify that mineral rich fouling are produced and used in the experiments later on. This is important to know since the cleaning process of protein and mineral rich fouling are believed to be different due to the different composition in the two fouling types. The result from the chemical analysis can be seen in Table 6. The protein content is similar for the three UHT fouling samples 12.4%, 13.3% and 12.1%. The protein content in mineral fouling are expected to be around 15-20% so the protein content in the UHT fouling are a bit lower than the values presented by Burton (section 2.3). The mineral content was defined to be calcium, magnesium and phosphorus which would be expected to contribute to around 90% to the total ash content so the real ash content could differ some from the measured and calculated and should be viewed as a well based estimation what the mineral content are. The ash content for the UHT fouling samples are 80.7%, 79.4% and 78.6% which is also was similar and close to the mineral values of 70-80% presented by Burton. The Ca/P molar ratio in the UHT fouling where 1.43, 1.43 and 1.45 which is a bit lower than the theoretical 1.5 for the tricalcium phosphate, but it is higher than the 1.3 presented by Burton. This would indicate the crystallites of tricalcium phosphates are formed in the fouling.

For the preheat sample the protein content is 34.1%, mineral content 50.5% and a Ca/P molar ratio of 1.65. These values compared to those presented by Burton (section 2.3) for protein fouling you can see that the protein content is lower and mineral content higher also the Ca/P ratio of 1.65 is higher than 1.5 Ca/P ratio proposed for tricalcium phosphate which would indicate that other crystallites of the calcium phosphate are present in the fouling.

Table 6 Results from chemical analysis of fouling taken from UHT and preheat part of the heat exchanger. The values are presented as weight percent.

	Protein, %	Ash, %	Ca/P molar ratio
UHT fouling 1	12.4	80.7	1.43
UHT fouling 2	13.3	79.4	1.43
UHT fouling 3	12.1	78.6	1.45
Preheat fouling	34.1	50.5	1.65

The difference between the UHT and preheat fouling produced here is likely to do with run time and the temperature differences. The fouling composition is believed to change with time, so run time is important to notice for similar heating conditions. The difference between the preheat and UHT fouling is most likely to do with the temperature here since the time was the same, protein fouling consist mainly of β-LG and calcium meanwhile mineral rich fouling would consist of casein and calcium phosphate (Changani et al., 1997). It can be that most of the denatured and aggregated β-LG are situated at the preheat part and when the temperature increases the aggregated β-LG are becoming too big to deposit and get flushed away with the flow. That leads to less protein content in the UHT deposit. Meanwhile the casein micelle becomes instable and dissociates with the colloidal calcium phosphate in the nanocluster which then may deposit at the surface. This could be one explanation why the mineral content is higher and protein content less in UHT fouling.

The UHT fouling produced in this project was similar enough to the type B (mineral rich) fouling presented by Burton (section 2.3) to be characterized as mineral rich fouling. The preheat fouling is less clear which part it would be characterized in, it is more similar to protein rich fouling in both looks and composition. So for the experiment the fouling taken from UHT sector can be used as investigation of cleaning mineral rich fouling meanwhile the fouling from the preheater is not used.

3.2 Preparation of cleaning liquids

Cleaning liquids were prepared from 50 wt% sodium hydroxide (VWR International, France) and 53 wt% nitric acid (Swed Handling AB, Sweden) solutions. The cleaning solutions were diluted with water to get the desired cleaning liquid concentration for the experiments. The amount of water and stock solution needed was calculated as follows;

$$V_1 = \frac{C_2 * V_2}{C_1}$$
$$V_w = V_2 - V_1$$

C_1	Nitric acid or sodium hydroxide start concentration in wt%.
V_1	Volume needed from start concentration to prepare new cleaning liquid in mL.
C_2	Wanted concentration of the cleaning liquid in wt%.
V_2	Volume wanted for the cleaning liquid in mL.
V_w	Volume water to dilute the start solution with in mL.

To verify that the wanted concentrations of the cleaning liquids were achieved a titration method was performed to measure and calculate the actual concentrations in the cleaning liquids, see Appendix A for titration method and calculation equations.

The real concentrations of the cleaning liquid prepared were as follows; Nitric acid cleaning liquids were 1.07 wt% for the 1 wt% HNO_3 and 1.59% for the 1.5 wt% HNO_3.The concentration for the sodium hydroxide liquids were 0.58 wt% for the 0.5 wt% NaOH and 1.54 wt% for the 1.5 wt% NaOH.

3.3 Basic cleaning method

In order to assess how different preparations and selections of fouling influence the cleaning efficiency a basic cleaning method was used. The basic method was based on a previously performed thesis work at SIK by Lasse Ropers and basic understandings how a CIP process is performed. The method was aimed to simulate the cleaning (dissolving) of fouling during different temperature and concentrations during the alkaline and acidic cleaning steps. The cleaning method involves several steps and the importance and influence of some of them are being evaluated to present a new cleaning method. With the new cleaning method screening trials is performed to see if it repeatable and if it is possible to distinguish the influence of temperature and concentration effects for dissolution of fouling.

Basic cleaning method;

1. Add 100 mL of cleaning liquid to a 250 mL beaker
2. Heat cleaning liquid to wanted temperature with a heating plate (Janke & Kunkel GmbH & Co) controlled by a thermostat (ETS-D4, IKA-Werke GmbH & Co).
3. Add fouling sample into the cleaning liquid after the wanted temperature is reached.
4. Clean the fouling sample for 10 minutes with magnetic stirrer (2.5cm) on low rotational speed.
5. Separate the remaining fouling from the cleaning solution by vacuum filtration. Using water suction and büchner funnel with filter paper (d=11cm and pore size 11µm, Whatman)
6. Rinse the remaining fouling present on the filter paper with distillated water.
7. Dry the sample and then weigh the dry fouling

This is the first cleaning step, cleaning with alkaline. The second cleaning step is similar and differs only in amount of fouling used, where the solid fouling remaining from the first part is used and nitric acid is used as a cleaning liquid.

4. Results & discussion

In the first section of the results the new cleaning method will be presented. The section after that will present the experiments performed and results obtained in order to achieve the new cleaning method. In the last section the cleaning effects measured from the new cleaning method will be presented to view the influence of concentration and temperature of the cleaning liquids on dissolution of fouling.

4.1 Cleaning method

The new cleaning method is presented in this section. In section 4.2 the experiments performed and results gathered to reach this cleaning method are presented. The method is done to measure the dissolution of fouling under different temperatures and concentrations of cleaning liquid. Dissolution is measured by weighing the fouling samples after each cleaning step. It is divided into to two sections "Selecting and preparation of fouling" where treatments of fouling prior the cleaning stage are presented and "Cleaning procedure" where the steps performed in order to dissolve the fouling are presented.

Selecting and preparation of fouling:

I. Take UHT fouling from the storage container and sieve between 5.6 mm and 0.71 mm mash grid size sieves.
II. Put the sieved samples in vacuum oven (Gallenkamp) at 80°C and 850 mbar and dry the samples overnight.
III. Weigh up 0.125g of small and large sized fouling each, total of 0.250g fouling. Place prepared sample in a desiccator until it is needed for the cleaning procedure.

Cleaning procedure:

1. Add 50 mL of NaOH cleaning solution into a 100 mL beaker, put it on a heating plate (Janke & Kunkel GmbH & Co) connected to a thermostat (ETS-D4, IKA-Werke GmbH & Co) and heat until set temperature is reached.
2. Add the prepared fouling into the heated cleaning liquid. Put the magnetic stirrer (1.5 cm) on low stirring speed. Clean the fouling for 10 minutes.
3. Pour the cleaning solution and remaining solids into a 50 mL falcon tube. Centrifuge (Hettich universal 2S) the sample for 5 minutes and 3200 rpm.
4. Pour out the cleaning liquid and rinse the solid fouling with water. (Keep the supernatant for back titrations with EDTA) Centrifuge again with the same settings.
5. Pour the water away carefully and scrape the solid fouling out from the tube and put on a glass dish.
6. Put the fouling in vacuum oven 80°C and 850 mbar overnight for drying.
7. Weigh the dried fouling on the glass dish.
8. Add 50 mL of HNO_3 cleaning solution to a 100 mL beaker, put it on a heating plate and heat until desired temperature is reached.
9. Transfer the sodium hydroxide cleaned and dried fouling to the heated nitric acid cleaning liquid. Clean the fouling for 10 minutes with low speed on the magnetic stirrer.

10. Pour the cleaning solution and remaining solids into a 50 mL falcon tube. Centrifuge the sample for 5 minutes and 3200 rpm.
11. Pour out the cleaning liquid and rinse the solid fouling with water. (Store the supernatant for back titrations with EDTA) Centrifuge again with the same settings.
12. Scrape out the pellet from the falcon tube and put it on a glass dish. The solid fouling is then put in vacuum oven overnight for drying.
13. Weigh the dried and cleaned fouling.

4.2 Method development

In the method development the different experiments performed to develop the new cleaning method will be described. The results from the experiments will then be explained in how that contribute to the way the cleaning method is performed. The repeatability of the cleaning method will also be evaluated.

4.2.1 Dry content

In order to evaluate the cleaning efficiency the weight change of the fouling after each cleaning step are measured. The weight change could be influenced by other factors others than dissolved fouling. If the samples can hold fluids this could affect the measurements. An experiment is performed to see if the fouling can hold water that could affect the measurements.

Fouling (0.8g) stored in room temperature was placed in lukewarm water for 10 min in a 100 mL beaker. The excessive water was then carefully poured off so there was only wet fouling left in the beaker. The wet fouling was then divided into two samples and transferred onto glass dishes and weighed (wet weight) and then put in vacuum oven at 80°C and ~850 mbar for 15 hours. The samples were then taken out of the vacuum oven and cooled; the weight (dry weight) is measured. Samples were dried for another two hours with same temperature and pressure and weighed again, to verify that the sample was completely dry. The dry content was then calculated as the dry weight divided by the wet weight.

Dry matter of the fouling was measured to be around 45%, see Table 7, which means that that it can hold a lot of fluids. If it can contain around 60% water it is likely that the fouling matrix contains channels and hollows where the water can reach. If there are channels and hollows in the matrix that the water can reach it is possible for the cleaning liquids to reach these cavities and channels.

Table 7 Results from the dry content experiments.

Sample	Dry content, %
#1	43.63
#2	46.92
#3	50.00
#4	37.57

These experiments where performed on fouling that had been stored in room temperatures for a couple of days and therefore cannot be seen as a measurement of the dry matter of fresh fouling. The fouling can hold a lot of fluids which could influence the measurement of the

cleaning efficiency, since the dry weight is needed to compare the different samples. Further experiments on treating fouling for receiving consistent weight measurements of the samples was performed in section 4.2.2.

4.2.2 Storage and drying

Storage condition of the fouling was investigated to see if different temperatures and relative humidity changed the weight of the fouling and therefore a need for a drying step in the cleaning method. Since the weight should be measured on the fouling and not the water change in the fouling. For the cleaning method it is important that consistent dry weights for the samples are measured. Three storage conditions were evaluated; Laboratory Room (LR), Climate Chamber (CC) and Climate Room (CR) each had different relative humidity and temperature which can be seen in Table 8.

Table 8 Storage conditions for the three storage places.

	Relative Humidity, %	Temperature, °C
Laboratory room	28	22
Climate chamber	21	71
Climate room	53	21

The experiment was performed in duplicates for each storage condition and fouling of similar shape and weight were used. The method of measure the changes are presented below.

1. Weigh up twelve samples of 0.3 gram each and place on filter paper (d=11cm and pore size 11µm); soak six of the samples in lukewarm water for 10 minutes.
2. Place two dry and two soaked samples in the three storage conditions.
3. Weigh the samples after 24 hours of storage.

The results from the experiments are presented in Table 9. Where the weight change after 24 hours storage are shown in percent.

Table 9 Difference in weight percent of the fouling after 24h stored under different storage conditions.

Samples	Laboratory room	Climate chamber	Climate room
Dry #1	0.4%	-3.3%	2.1%
Dry #2	-0.4%	-3.6%	2.7%
Wet #1	7.5%	-0.3%	10.2%
Wet #2	5.6%	0%	11.8%

The dry samples stored in laboratory room should not show any difference since the samples were taken from the fouling container stored in the same laboratory. The result shows that the weight difference for these is very little and may be due measurement equipment. For the samples soaked in water prior storage to simulate wet fouling after a cleaning step shows that the samples were not dried to the start weight after 24 hours for the LR and CR samples, which can be seen in Table 9. There is probably still some moist in the matrix of the fouling after this time. The dry and wet samples stored in climate room both showed an increase in weight from the start weights. The dry samples increased the weight by around 2.4% and the wetted samples around 11%. The slow drying process for the wet samples is likely due to the

high humidity. In the climate chamber the weight decreased for the dry by 3.5% and the wet samples dried to the start weight. The samples are not consistent in weight when comparing dry and wet samples after 24 hours, which means that there still are water present in the fouling.

From the results it can be shown that the storage condition has an influence of the weight and drying process, which means that a drying step is needed. It showed that high relative humidity was bad in order to achieve dry samples. Meanwhile the climate chamber that had a temperature around 70°C and 20 RH% showed most promising for drying samples. But even the climate chamber did not dry the wet samples entirely. In order to get consistent dry samples, a faster and more reliable drying is needed for the cleaning method. If there is still water present in the fouling when weighed, it will disturb the interpretation of the amount of dissolved fouling. The result suggests that higher temperature and lower relative humidity increases the effectiveness of the drying process.

From the results obtained above it was decided that a drying step is needed. Experiment to dry the samples in vacuum oven was performed. Three fouling samples with similar size and shape was taken from UHT fouling container and soaked. The wet samples were then put in vacuum oven (Gallenkamp) at 80°C and a pressure of 850 mbar overnight. The samples were weighed the morning after and then put in the vacuum oven for 2.5 hours more. They were weighed again to verify that the samples were dry. The result from these measurements showed that the weight of the samples in the first measurement was 0.259g, 0.256g and 0.253g. The second measurement for the same fouling was 0.259g, 0.255g and 0.253g. The difference weight between the second samples in the drying has probably to do with measurement error rather than drying occurring between the measurements. Consistent dry samples can be achieved by drying the fouling in vacuum oven at 80°C and a pressure of 850 mbar overnight. These results show that in order to measure the actual weight change of the fouling a drying step is needed before cleaning and after each cleaning step. This is why the vacuum oven drying is applied in the cleaning method before and after each cleaning step.

4.2.3 Capillary forces or diffusion forces

From the results obtained in section 4.2.2 a drying step is necessary for the cleaning method in order to be able to measure the weight change of the fouling. Comparison between the dissolution of wet samples and dry samples was performed to see if the cleaning is dominated by the diffusion forces (wet) or capillary forces (dry) fouling in the cleaning process, since the fouling are dried in vacuum oven prior cleaning.

Four fouling samples of 0.5 gram each was taken out from the storage container. Two of the samples were then used for wet fouling and the other two as dry fouling. The wet samples were put in lukewarm water 10 minutes prior each cleaning step. The experiments were otherwise performed according to the basic cleaning method (section 3.3) and done in duplicates with the settings presented in Table 10.

Table 10 Settings used to evaluate if there are any difference in cleaning efficiency using wet or dry fouling in the cleaning process.

Cleaning liquid	Concentration, wt%	Temperature, °C	Time, minutes	Separation method
NaOH	1.5	80	10	filtration
HNO_3	1.5	75	10	filtration

Results from the soaked samples compared to dry fouling cleaning shows that there are no big difference in the dissolved fouling over the entire cleaning processes, see Figure 5. During the first cleaning step with NaOH, there is a small difference in the remaining fouling for the two types. For the dry samples the first cleaning step with 1.5% NaOH the remaining fouling were 90% and 91% and for the wet samples these values were 88% and 84%. The difference between the soaked and dry sample here could be due to that the soaked samples were put in water for 10 minutes and then the water carefully poured out which could have brought small particles of fouling with it. For the second cleaning step, where acid is used, the dry values are 7% and 6.9% and wet samples 7.5% and 6.8%. All four samples are close to each other, and a difference cannot be seen in cleaning stage 2.

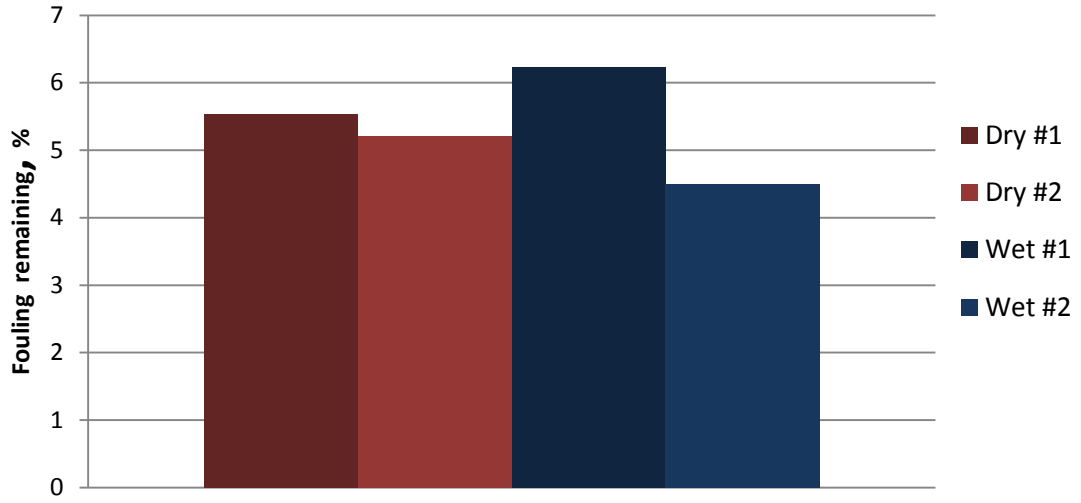

Figure 5 Comparison of soaked (diffusion forces) and dry (capillary forces) fouling remaining after cleaning with alkali and acidic liquids performed in duplicates.

Over the total process, the fouling removed were 94.5% for both wet and dry samples. For the method it does not seem to have any significant difference in dissolved fouling by using fouling that has been soaked for 10 minutes in lukewarm water prior each cleaning step. For the cleaning method, dried fouling can be used without a soaking step prior the cleaning. Fresh non-dried fouling was not investigated. To get a method that is more similar to the ones in production plant it could have been good to investigate fresh fouling also in this method to see if there are any difference.

4.2.4 Size of fouling

The diffusion and masstransfer in smaller and larger samples can be limiting factor in the cleaning process. Different sizes of the fouling will result in different surface area which could influence the cleaning result. To compare if the size (surface area) of the fouling has any significant effect of the cleaning result of fouling in this method the fouling was divided into smaller and larger pieces. Separation of fouling into smaller and larger pieces was performed by visually dividing them (Figure 6). From the divided parts four samples of 0.5g each was prepared, two from the smaller size and two from the larger size.

Figure 6 Comparison of large and small pieces of fouling. Large fouling pieces are placed in the top half of the petri dish and small fouling in the bottom half.

The basic cleaning method (section 3.3) was used to evaluate if there was any difference in cleaning efficiency due to different sizes. The settings for the experiments are listed in Table 11.

Table 11 Settings used to determine if there are any differences in cleaning efficiency by using smaller or larger fouling pieces.

Cleaning liquid	Concentration, wt%	Temperature, °C	Time, minutes	Separation method
NaOH	1.5	80	10	Filtration
HNO_3	1.5	75	10	Filtration

Samples were divided by taking the smallest and largest pieces from the storage container. The final cleaning degree (remaining fouling) for the large pieces was 8.2% and for the small pieces it was 7.6% (Figure 7). After the first cleaning step with sodium hydroxide 84.9% of the large fouling was remaining while 89.5% of the small fouling remained. For the second cleaning step with nitric acid 11.2% of the large fouling is remaining and 10.5% of the small fouling.

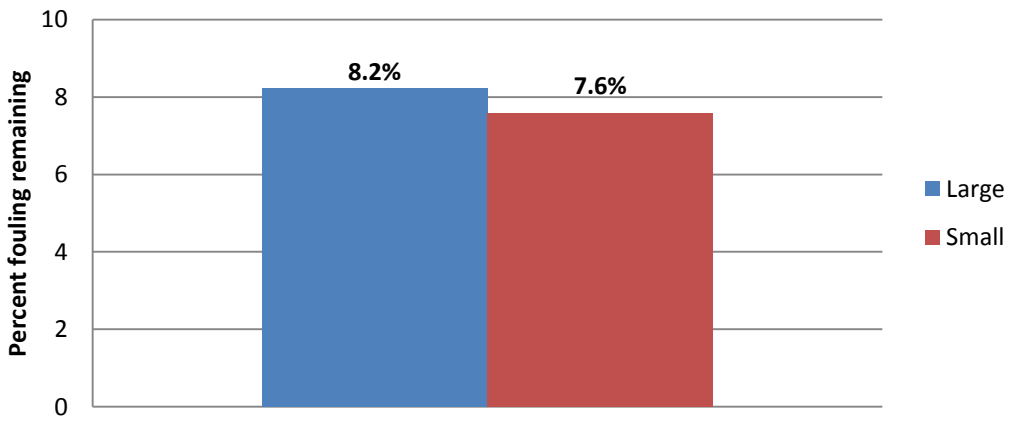

Figure 7 Comparison of final cleaning efficiency of large and small pieces of fouling performed in duplicates.

It seems like there is a small difference between the amounts of dissolved fouling in the first cleaning step, but this difference is then reduced in the second cleaning step. For the overall process, the choosing of small or large fouling does not seem to have any large influence. So from these results it was chosen that fouling from both sizes should be selected for the new cleaning method since it could be that with different cleaning times the size may still have an influence of the results. The two sizes of the fouling were chosen to be selected by sieving them between two mash grid sizes where the sizes were selected after the appearance of the fouling, so there would exists enough large and small pieces. The sizes of the sieves were 5.6 mm and 0.71 mm mash grid and fouling stuck on these layers were selected too be used in the cleaning method.

4.2.5 Separate solid fouling and cleaning liquid

A problem that aroused during the separation of the fouling and cleaning liquid was that fouling got dried into the filter paper. This was problematic since the fouling is needed for another cleaning step. To scrape the fouling of the filter paper cause a risk for filter paper to be scraped off, this will influence the measurement of the weight. In Figure 8a pieces of filter paper can be seen lie in the middle with fouling around it, this causes problem when measuring the remaining fouling when both the filter paper and fouling are being weighed. In the Figure 8b the fouling are stuck onto the filter paper which causes risk of filter paper being scraped of when trying to gather the dried fouling for the weight measurements. These problems forced a test of a new separation method, and the alternative method found to be a viable option was centrifugation. Several trials were performed with cleaned fouling and cleaning liquids to achieve enough separation so that the cleaning liquid could be poured out of the falcon tubes used in the centrifugation (Hettich Universal 2S) without risk of bringing visible particles of fouling with it. The result from these trials were that 5 minutes at a rotational speed of 3200 rpm was the best too be sure that all visible fouling particles are located in the pellet (Figure 8c). Centrifugation was found to be more practical than filtration.

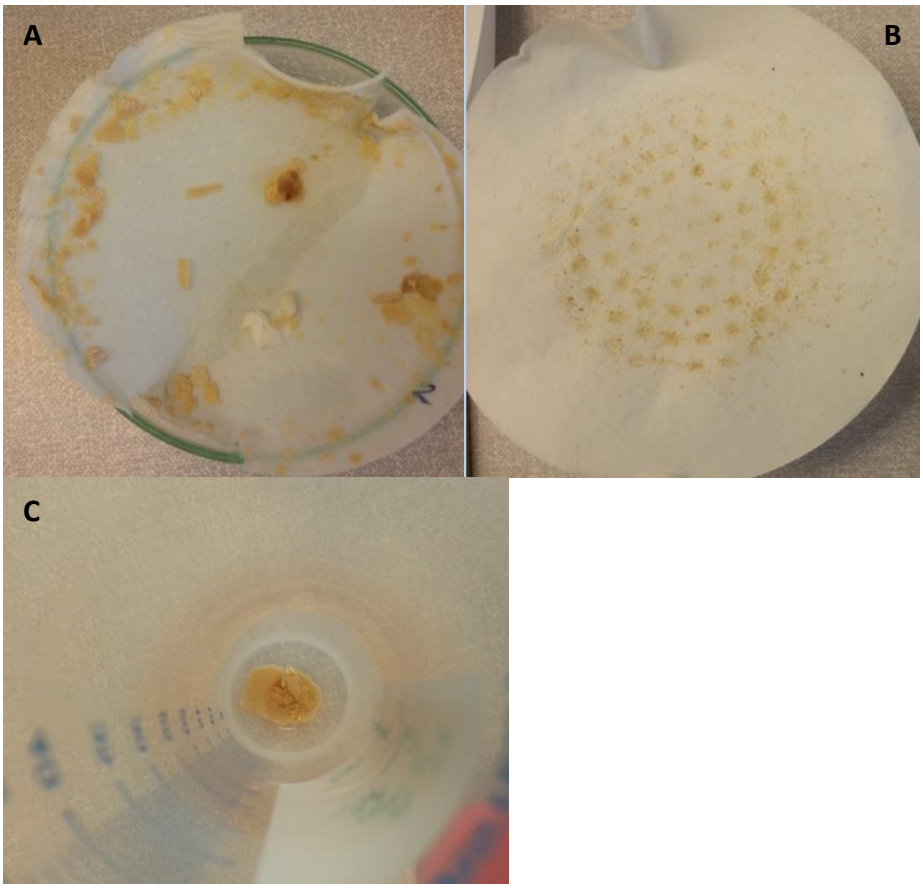

Figure 8 A) Fouling on a filter paper after cleaning with NaOH where the filter paper is broken and pieces present in the middle. B) Dried in fouling after cleaning with HNO_3, where risk for scraping filter paper off when trying to scrape off fouling. C) Fouling in 50 mL falcon tube after centrifugation and the cleaning liquid poured off.

4.2.6 Volume cleaning liquid, amount fouling and size of magnetic-stirrer

From the results obtained in section 4.2.5 the separation of fouling and cleaning liquid are performed with centrifugation. The falcon tubes used for centrifugation were limited to an amount of 50 mL cleaning liquid. To achieve separation of cleaning liquid and fouling in one separation process the amount of cleaning liquid used for the cleaning method was chosen to be 50 mL. Since the cleaning liquid are limited to 50 mL the amount of fouling must be altered to this amount since there should be enough active agents in the cleaning liquid to perform the dissolution of fouling. According to the example reaction in Figure 3, 4 mol of nitric acid needed for transforming 1 mol of calcium phosphate into water soluble substances. The amount of nitric acid in 1 wt% 50 mL cleaning liquid is 0.502gram. For dissolving 1g of fouling 0.8g of nitric acid is needed. The amount of 0.250g of fouling would then be enough to achieve a theoretical dissolution of all the fouling if it was consisted of only tricaclium phosphate. Since around ~80% of the fouling is of mineral type the total amount of cleaning liquid is about 3-4 times higher than the theoretical necessary value. So the weight of 0.250 g fouling was chosen in the cleaning method since it is sufficient to achieve a cleaning without the cleaning liquid being a limiting factor. For the cleaning with sodium hydroxide there is no overall process that could explain the reactions but since the amount of protein is small it is not likely that the amount of 50 mL NaOH at different concentrations (0.5 wt% - 1.5 wt%) would be too little. Dissolution trials performed at Tetra Pak where the dissolution of fouling

at different concentration of NaOH were compared no result indicated that the NaOH limited the dissolution at lower liquid/fouling ratio than used here.

Another problem that appeared in the cleaning method development was that there was a risk of the magnetic stirrer (2.5 cm) to grind fouling against the wall of the beaker. The rotation in the fluid is necessary to achieve a good flow of cleaning liquid to the boundary layer of the fouling surfaces. In order to minimize the risk of visible grinding of the samples against the beaker wall the magnetic-stirrer size was reduced to a 1.5 cm long one. The magnetic-stirrer were then placed in the middle of the beaker were it could stir the solution with reduced risk of grinding the solid fouling against the wall.

4.2.7 Determine calcium and magnesium ion concentrations in the cleaning liquid

After each cleaning step the fouling is separated from the cleaning liquid. This cleaning liquid can be used to indicate the dissolved calcium and magnesium and give a hint on how much of the minerals in the fouling that has been dissolved. The cleaning liquids gathered in the testing of the new cleaning method (section 4.2.8) were used to measure the dissolved amount of calcium during different conditions. To measure the dissolved calcium and magnesium ions a backwards titration containing EDTA2Na (International Biotechnologies Inc, New Haven, Connecticut) and magnesium chloride (Scharlau, Spain) procedure was adapted from University of Canterbury (Science, 2014) and performed according to the following steps;

Rinse all the equipment with distillated water before usage.

1. Take 10 mL of the used cleaning liquid and add to an Erlenmeyer flask of 250 mL.
2. Add either 20 mL of 0.01M EDTA2Na if it is from the NaOH solution or 5 mL of 0.1 M EDTA2Na if it is from the HNO_3 solution.
3. Add 10 mL of Ammonia buffer solution, pH 10 (Fluka, Germany) and 50 mL of distillated water. Stir the solution with magnetic stirrer.
4. Check the pH of the solution so it is in the around 10 pH, with a pH meter. If not add more Ammonia buffer solution. This is done to verify that the dye indicator Eriochrome Black T (Fluka, Germany) is able to shift color correctly which it does in the 8-11 pH range.
5. Add a spoon tip of Eriochrome Black T to the solution; continuously stir the solution with magnetic stirrer. The solution is now supposed to be light blue, if it is pink or purple add extra EDTA until the solution are light blue. There is now an excess amount of EDTA in the solution. Note total volume of EDTA2Na added.
6. Titrate the light blue solution with magnesium chloride solution of 0.025 M until it turns to stable pink color. Note the volume of added magnesium chloride.

The dissolved calcium and magnesium ions can then be determined by first calculate the total moles of EDTA added to the solution and then calculate the moles of magnesium chloride used for the titration. The excess amount of EDTA can be calculated from the equivalence to the moles of magnesium ions. The ratio of $(Ca^{2+}+Mg^{2+})$: EDTA is 1:1. The moles of calcium and magnesium ions complexed with EDTA are determine by subtracting the excess EDTA from the total amount of EDTA. From this the moles of calcium and magnesium ions in

solution is determined. From the moles from this the concentration of it in the sample can be calculated. For the simplicity and result from chemical analysis (Appendix B) where the ratio of calcium and magnesium was 20:1 the concentration is calculated on the calcium ions which would give the values a bit skewed, but not impact the interpretation of the results.

From the complexometric titration with EDTA and magnesium chloride the result are presented as calcium dissolved in cleaning liquid (mg/L).The dissolved amount of calcium ions in the different cleaning steps can be seen in Table 12.

Table 12 Measured calcium ions concentration in the cleaning liquid after cleaning stap 1 and 2.

Sample	Cleaning step 1 Calcium (mg/L)	Cleaning step 2 Calcium (mg/L)
1.5 wt% 50°C	120	1390
1.5 wt% 50°C	30	1490
1.5 wt% 70°C	80	1550
1.5 wt% 70°C	20	1490
1.5 wt% 90°C	100	1600
1.5 wt% 90°C	10	1470
1.5 wt% 90°C	20	1790
0.5 wt% 50°C	40	2300
0.5 wt% 50°C	10	1580
0.5 wt% 70°C	50	1510
0.5 wt% 90°C	40	1500

From the values of the titration it is not possible to distinguish any difference of fouling cleaned with different temperature and concentration. There is still possible to see that significantly more calcium has been dissolved in the acidic cleaning step compared to the alkaline, 1400-2300 mg/L and 10-120 mg/L respectively. More fouling has been dissolved in the nitric acid step and therefore the calcium concentration is higher since nitric acid dissolve the mineral fraction. In the alkaline step the dissolved fouling is little and it is supposed that mainly parts from the protein fraction has been dissolved which could explain the small amount of measured calcium ions (Table 12). There are only measurement values from trials 8-18 since the titrations before were performed with a direct titration method where the liquid became turbid and there was not possible to see any clear and stable color shift. The titration method to determine the calcium and magnesium ion content used was a back titration with magnesium chloride there a clear and stable color shift could be found. The titration is very sensitive and must be performed really carefully with more precise volumetric measurements than used in this experiment where the cleaning liquid was poured into a measuring jug and the EDTA2Na and magnesium chloride was pipetted with 1mL pipettes. If a more precise measurement is performed it may be possible to use this titration method to compare cleaning efficiency between different concentration and temperatures in the acidic cleaning step. To conclude, the results shows that complexometric titration can be used to compare the calcium concentration between the alkaline and acidic cleaning step but need further improvements to

be able to use in comparison of temperature and concentration within the same type cleaning liquid.

4.2.8 Repeatability

The new cleaning method (section 4.1) was tested for repeatability and to see if it can be used to differentiate cleaning effects (section 4.3) under varying concentration and temperature of the cleaning liquids. The rotational speed of the magnetic-stirrer (flow) and time was fixed in these trials. Due to limited time, two concentrations of sodium hydroxide were tested in the first cleaning step, 1.5% and 0.5%. These were selected since they are commonly used in fouling experiments. Temperatures in the sodium hydroxide cleaning step are 50°C, 70°C and 90°C, the higher temperatures were chosen since increased cleaning effect should occur. Temperatures over 100°C would be hard to achieve with the experimental set-up used since no pressure vessel are used. In the second cleaning step, cleaning with nitric acid, only one level are used for cleaning the fouling 1 wt% HNO_3 at 80°C. The concentration and temperature was chosen to be similar to those used in industrial dairy plants. The experiments are also performed in triplicates on each setting to get a more reliable result. A total of 18 trials were performed with the new cleaning method in a randomized order according to Table 13.

Table 13 Run order and settings used to test the repeatability and effect of concentration and temperature in dissolving UHT fouling.

Sample, Run order	Concentration, wt% NaOH	Temperature, °C	Concentration, wt% HNO_3	Temperature, °C
#1	0.5	50	1	80
#2	0.5	90	1	80
#3	0.5	90	1	80
#4	0.5	70	1	80
#5	1.5	70	1	80
#6	0.5	70	1	80
#7	1.5	50	1	80
#8	1.5	50	1	80
#9	0.5	50	1	80
#10	1.5	70	1	80
#11	0.5	90	1	80
#12	1.5	90	1	80
#13	0.5	70	1	80
#14	1.5	90	1	80
#15	1.5	90	1	80
#16	1.5	70	1	80
#17	0.5	50	1	80
#18	1.5	50	1	80

To see that the result for the process is not influenced too much by increased skill and knowledge in how to execute the different steps in the cleaning method, the mean value of the remaining fouling for each setting are subtracted from each sample and plotted according to the run order (Figure 9). The linear regression has a slope of -0.1442 which would indicate

that there is a little improvement by the laborants cleaning skills over time. The last four samples run showed improvement of the cleaning compared to the mean value. Even though these four samples showed improved cleaning, the samples are from separate cleaning settings and the effect would not affect the result significantly since they are "grouped in" with the other measurements. There seems like it is an improvement occurring in the laborants skills and knowledge in performing the cleaning. This improvement of laborant does not seem to influence the overall cleaning results, the same conclusion from the experiments would be reached weather or not improvement of the laborant would be occurring. The results obtained can be seen as the influence of the process parameters; concentration and temperature.

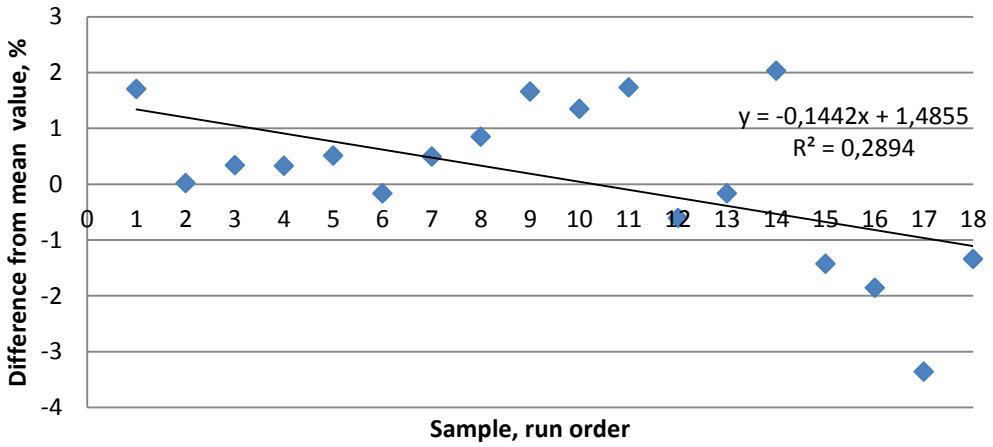

Figure 9 Difference from mean value for each sample after cleaning with NaOH and HNO_3 sorted according to run order.

In order to assess the repeatability of the method the different treatment has been grouped together according to concentration and temperature used in the alkaline cleaning step. These are then plotted into two separate graphs; one for the first cleaning step and one for the second cleaning step (Figure 10 & 11).

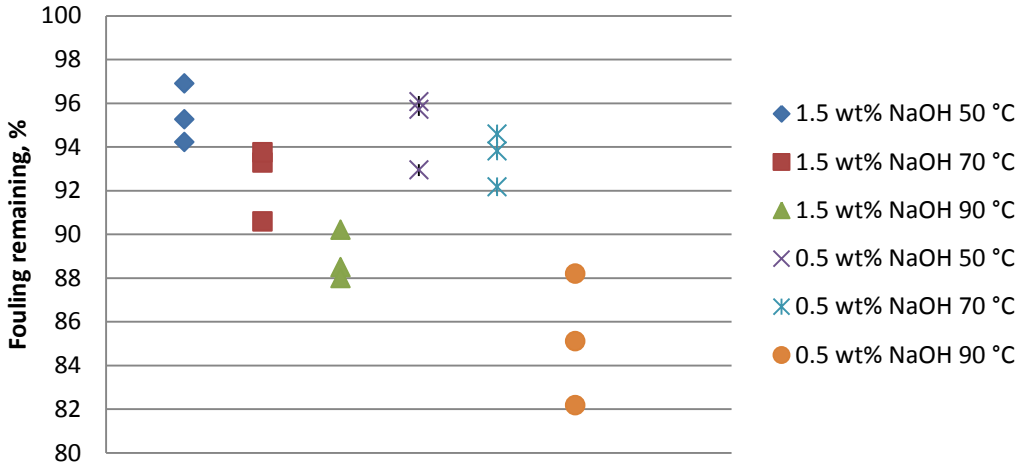

Figure 10 Fouling remaining after cleaning with sodium hydroxide. Grouped after which concentration and temperature the sample was cleaned with.

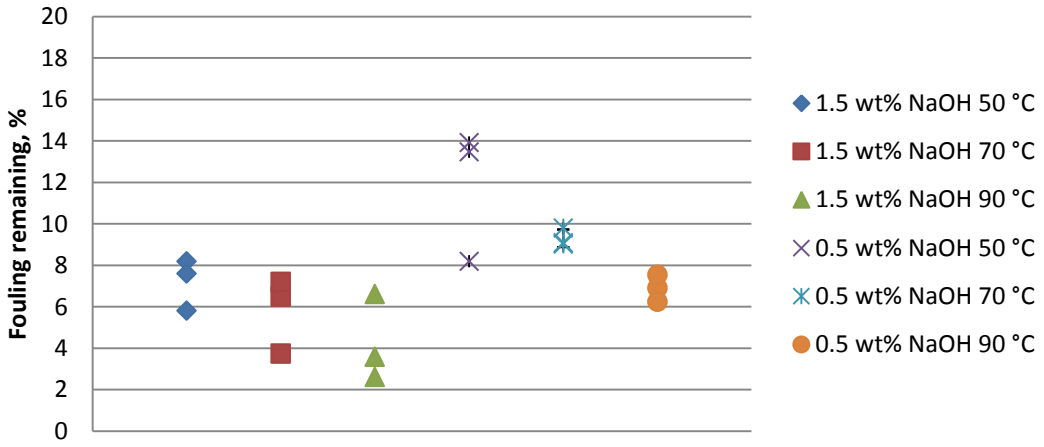

Figure 11 Fouling remaining after cleaning with nitric acid at 1 wt% and 80°C. Sorted after how they were cleaned in the sodium hydroxide step.

From Figure 10 it can be seen that for the first five cleaning conditions the results are in a range of 3-4% meanwhile for the sixth cleaning condition the result are in a range of 6% when cleaning with NaOH. This is not a big difference within the samples of the same group. The same patterns occur where the samples of the same group are within a range of 3-4% except for the fourth one which is in a range of 6% (Figure 11). The difference in cleaning conditions in Figure 11 is not clearly distinguishable as in Figure 10 it is probably because all of them are cleaned with 1 wt% nitric acid at 80°C. So for the both cleaning steps the triplicates are in a range 3-6% and that seems to be enough resolution to be able to distinguish effects of different temperature and concentration in the cleaning liquid (Figure 12 and 13). The cleaning trend starts occurring in Figure 12 after dissolving fouling at different concentration and temperature with sodium hydroxide. In Figure 13 a trend is visible for when fouling is cleaned with nitric acid at the same temperature and concentration after cleaned with sodium hydroxide. The cleaning trends and effects will be presented and discussed further in section 4.3. For a definite and statically significant result of the repeatability more experiments, must be performed for samples within the same condition group.

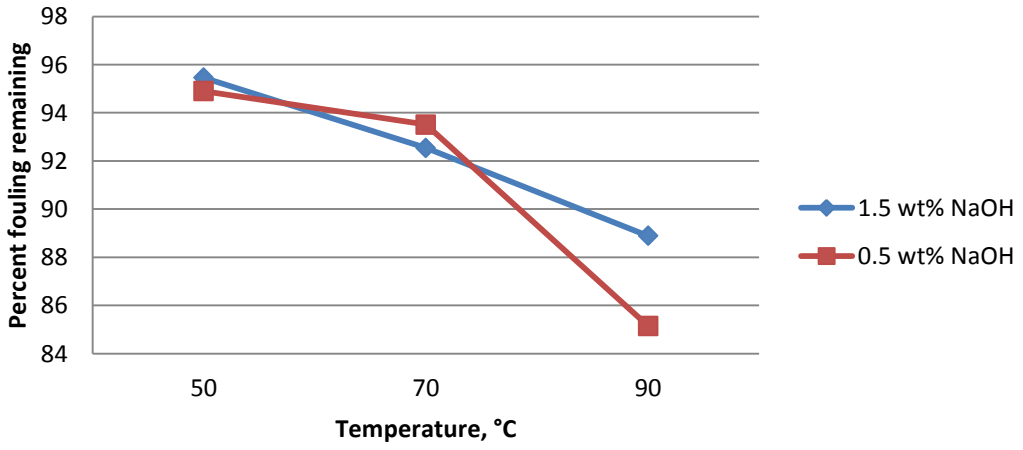

Figure 12 Cleaning trends occurring after dissolving fouling with NaOH at 1.5wt% and 0.5 wt% with 50°C, 70°C and 90°C. Mean values for the remaining fouling are plotted for the different conditions.

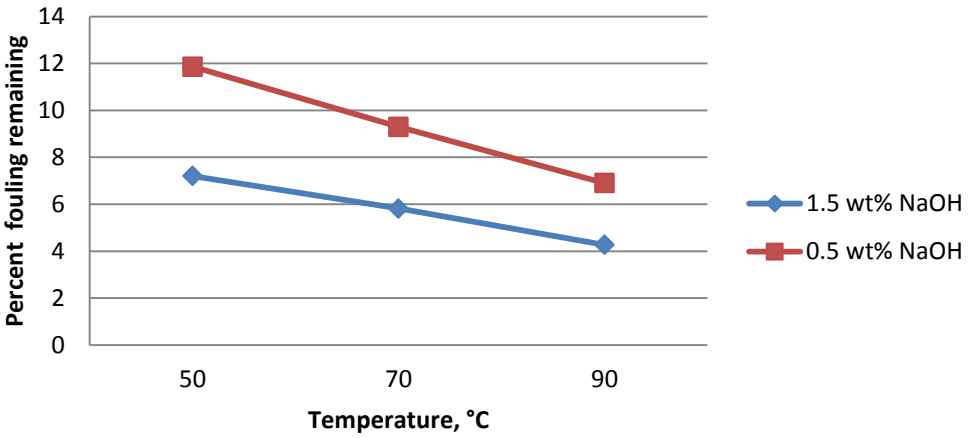

Figure 13 Cleaning trends that show up after dissolution of fouling in 1 wt% HNO_3 at 80°C after it has been cleaned in NaOH. Mean values for the remaining fouling are plotted after how the samples were treated in cleaning step 1.

4.3 Cleaning effects

The new cleaning method was evaluated by performing triplicate experiment with temperatures 50°C, 70°C and 90°C and concentration of 0.5 wt% and 1.5 wt% for the sodium hydroxide cleaning step. For the cleaning with nitric acid a fixed temperature of 80°C and concentration of 1 wt% was used. Time and rotational speed of the stirrer was fixed. In the following section the cleaning effects of different temperature and concentration of the cleaning liquid will be investigated from data collected from the same experiments to test the repeatability (section 4.2.8).

In the cleaning process the form, size and color of the fouling changed after each step. In Figure 14 the change occurring can be seen which is representative for all the trials. In Figure 14a the fouling is shown after the drying process where it is a bit grey-yellow and gritty. In Figure 14b after the fouling has been cleaned with NaOH the fouling pieces has become smaller and pale but still has a gritty surface. In Figure 14c the fouling are being formed as a pellet and the color has changed to be darker and brownish. The pellet is formed because of the centrifugation of the samples, when filtration was used to separate fouling and cleaning liquid the fouling clumped together and formed small isles on the filter paper with the same brown color.

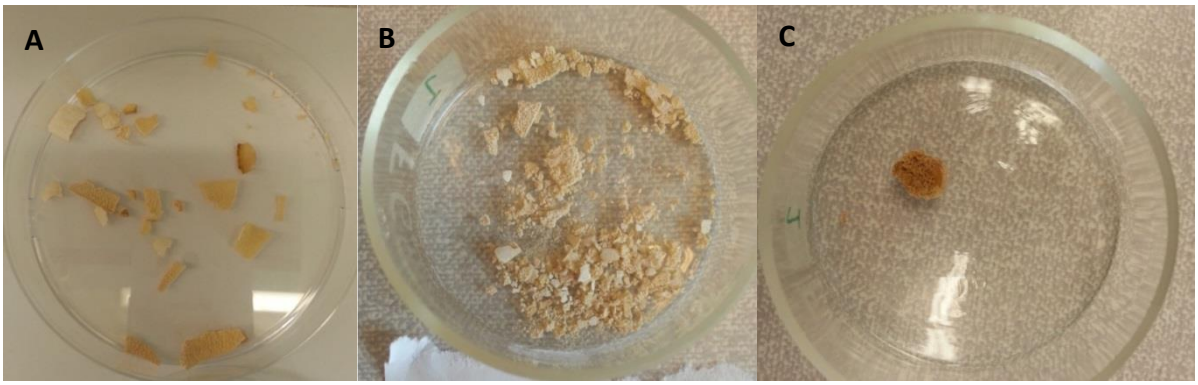

Figure 14 Pictures of the fouling for the first run. A) fouling used for the cleaning B) fouling after cleaning with NaOH C) fouling after cleaning with HNO_3.

The final cleaning is calculated from the start weight of the fouling and the weight after cleaning with nitric acid. The result from the six different conditions are presented and compared in Figure 15. The results are divided after which cleaning condition they were treated in the first cleaning step since the second cleaning step was the same for all samples.

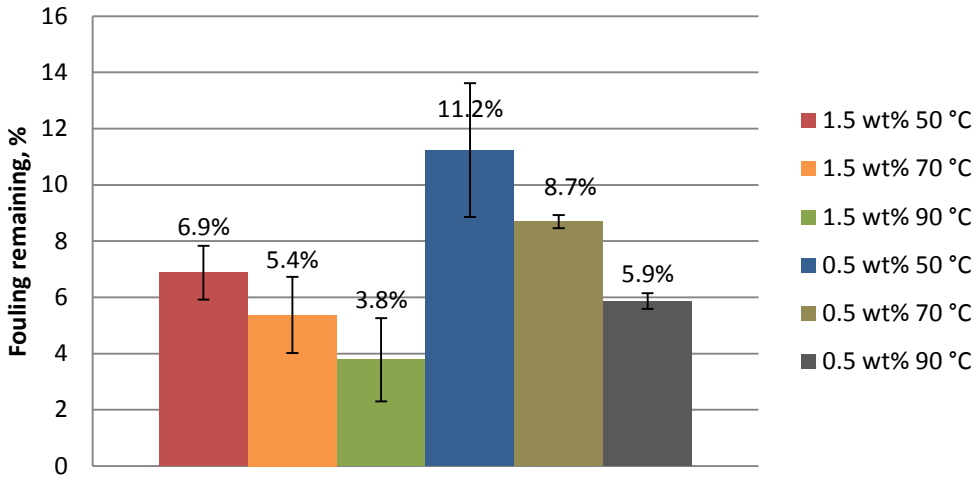

Figure 15 Comparison of the total cleaning for the fouling samples.

The treatment with 1.5 wt% NaOH the fouling remaining for 50°C, 70°C and 90°C are 6.9%, 5.4% and 3.8%. This indicates that the cleaning processes under 1.5 wt% NaOH improves a little with higher temperature. There is not a clear distinction between the 50°C and 70°C or the 70°C and 90°C but the difference between 50°C and 90°C seems big enough to say that 90°C has better cleaning effect than 50°C. So it is reasonable that the values for the 70°C lie between the lowest and highest temperatures this shows that the cleaning effect increases with temperature.

For the 0.5 wt% NaOH the same trend can be seen were the remaining fouling are smaller at 90°C compared to the 50°C and 70°C. The value for fouling remaining for 50°C, 70°C and 90°C are 11.2%, 8.7% and 5.9%. The spreading of the samples treated at 50°C is quiet big and cannot be really separated from those at 70°C. But the tendency is that during 50°C the fouling remaining is a bit higher than for 70°C. For the overall process using 0.5 wt% NaOH the higher temperature perform a higher dissolution.

When comparing the 0.5 wt% and 1.5 wt% against each other the result shows that for each temperature the fouling remaining at 1.5 wt% is lower. The similar degree of dissolved fouling is reached with 1.5 wt% at 70°C as with 0.5 wt% at 90°C. So for the overall cleaning process it seems like a higher concentration and temperature of the cleaning liquid is preferable to dissolve the most mineral rich fouling when using 1 wt% HNO_3 at 80°C as a second cleaning step. In order to assess the effect of the different cleaning steps the results obtained after each cleaning stages are presented in the following two parts.

Cleaning with sodium hydroxide

The results presented in Figure 16 show the percentage of the remaining fouling after the cleaning with 0.5 wt% and 1.5 wt% NaOH at temperatures 50°C, 70°C and 90°C. It is calculated from the start weight and weight after cleaning step 1. For the 0.5 wt% the remaining fouling for 50°C, 70°C and 90°C are 94.9%, 93.5 and 85.3%. The remaining fouling for 50°C and 70°C are really close to each other and the error bars are shows that it is not possible to separate the cleaning effect of these two temperatures. At 90°C the error bar is quiet big but it clear that at this temperature the dissolved fouling is higher than for 50°C and 70°C. From the result it is suggested that the dissolution of fouling starts to a higher degree at temperatures above 70°C for the 0.5 wt% NaOH.

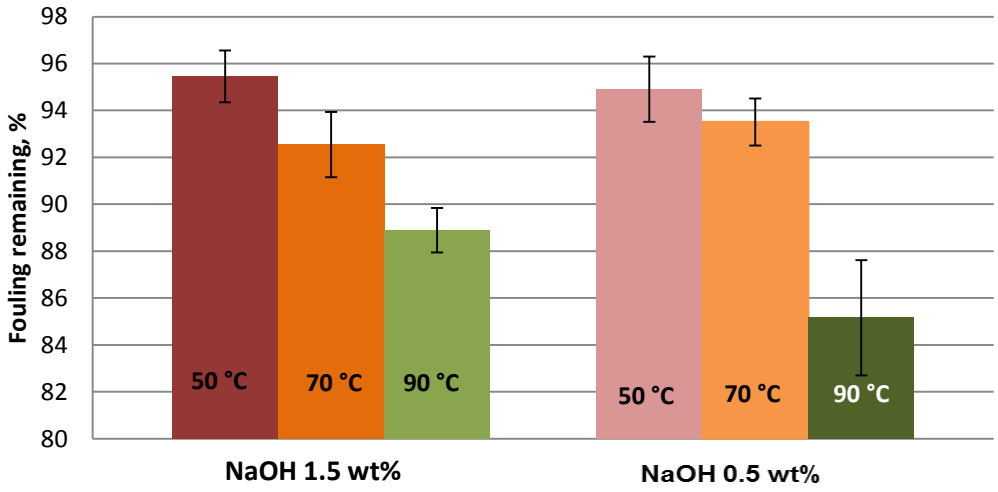

Figure 16 Fouling remaining after cleaning step 1 using sodium hydroxide as cleaning liquid.

In cleaning with 1.5 wt% NaOH the result for the 50°C, 70°C and 90°C are 95.5%, 92.5% and 88.9% fouling remaining. The least fouling has been dissolved at 50°C and most at 90°C, for 70°C the dissolved amount lies between the high and low temperature. The trend is that with increasing temperature more fouling are dissolved.

Comparing the 1.5 wt% and 0.5 wt% NaOH there does not occur to be any difference in dissolved fouling for 50°C and 70°C. At 90°C the 0.5 wt% dissolves some more fouling, but the error bar is quit big which could mean that the difference between the both 90°C is not as large as it appear in Figure 16. The trend that are shown are still viable tough. For the cleaning with NaOH the dissolved fouling are around 5-15% which would indicate that fouling are being dissolved since according to the chemical analysis performed on the fouling the protein content is around 12-13%. The sodium hydroxide is used for its ability to break down and cause swelling of the protein. Overall the temperature of 90°C for both cleaning liquids achieve better cleaning compared to 50°C and 70°C for the both. The most fouling dissolved is achieved when 0.5 wt% NaOH at 90°C is used.

Cleaning with nitric acid

Figure 17 shows the result after cleaning with 1 wt% HNO_3 at 80°C. The results were calculated from weight remaining after cleaning with NaOH and the weight after cleaning with HNO_3. For the fouling that has been treated with 1.5 wt% NaOH prior the HNO_3 cleaning the remaining fouling are similar for all temperatures. There is a small detectable difference between the 50°C and 90°C where at the higher temperature more fouling are dissolved. In the fouling cleaned with 0.5 wt% NaOH before the HNO_3 step the 50°C and 70°C shows to have smaller effect than the one at 90 degree. The effect of 50°C and 70°C cannot be distinguished from each other.

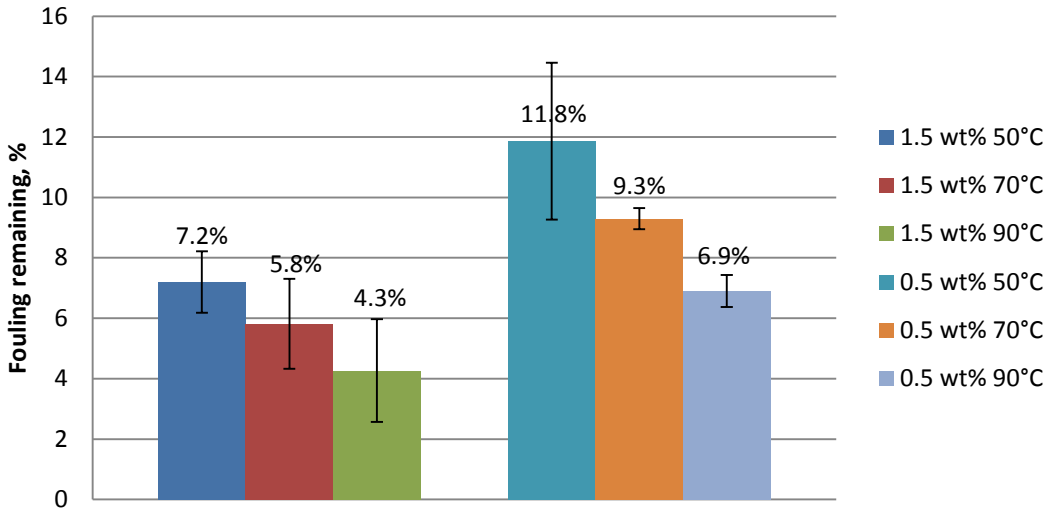

Figure 17 Fouling remaining after cleaning with NaOH and HNO_3. Presented by which condition that was used in the NaOH cleaning step.

When comparing the concentration effect of the used it is clear that the 1.5 wt% NaOH treatment is better. Looking for both concentrations the same trend as previously is displayed that higher temperature dissolved fouling better. That is probably due to that the fouling may have swollen and protein further decomposed which would allow for the nitric acid to access and diffuse further in the fouling matrix. In total between 88-95% of the remaining fouling from the first cleaning step is dissolved. It is reasonable since the fouling would mainly be composed by minerals and the sodium hydroxide would have dissolved much of the protein fraction. To conclude the nitric acid cleaning occurs to be more efficient if 1.5 wt% NaOH was used in the first cleaning step.

5. Conclusions

Cleaning of fouling is important for the industry and an improvement in cleaning efficiency can save both times, cost and reduce environmental impact. A laboratory method that could screen for which cleaning liquids that would be suitable for trials in pilot and industrial scale would be of help since the more time consuming and energy demanding screening in pilot plants can be reduced.

For the method development the fouling showed that the weight changed according to storage condition, and it is important to have a stable weight for each sample to be able to compare results. That gave valuable information that a drying step is needed to achieve comparable weights of fouling. From the comparison of small and large fouling pieces no clear difference in cleaning effects could be distinguished for processes performed at 10 minutes, but it could be that for shorter run times there could be a measurable difference. For fouling that already has been dried in room temperature no difference in cleaning effect of wet and dried pieces could be proven. The generation of fouling showed that fouling taken from UHT units of the fouling production pilot plant had a chemical composition that could be regarded as mineral fouling.

The repeatability of the method could not be statistically proven without any further experiments, but the trend showed that it was similar spreading for all the cleaning conditions that were around 4-6%. That resolution was enough to distinguish some trends in cleaning effects by using cleaning liquids under different concentration and temperature. The effect of detaching of larger pieces cannot be measured with this method only dissolution of fouling is able to be determined. The important CIP parameters flow and time were not investigated, the effect of time is able to be measured but the flow effect is not as easily performed. The complexometric titration showed that it must be performed precise and carefully but could be used for measure and compare the dissolved calcium and magnesium between alkaline and acidic cleaning liquids.

The cleaning method showed that using 1.5 wt% NaOH in the alkaline cleaning step resulted in that more fouling has been dissolved in the end. The temperature increases the cleaning efficiency with increasing temperature but that there is no large difference between 50°C and 70°C. It would suggest that increased cleaning efficiency is occurring between 70°C and 90°C. The fouling changed color and shape during the cleaning, from being gritty and whole flakes into being smaller pieces with gritty surface in the alkaline step to being a pellet of dark brown color after the acidic cleaning step. The best cleaning was achieved using 1.5 wt% NaOH at 90°C followed by 1 wt% HNO_3 at 80°C.

6. Future work

From the result and conclusions there are possibilities for future research in the laboratory method development for dissolving mineral fouling

- Perform more laboratory experiments to validate the repeatability of the method
- Compare if there is any significant different in dissolving fresh fouling to the room dried fouling.
- Change separation method after cleaning steps to improve the timings in which the cleaning stops.
- Improve the complexometric titration to be more precise for evaluate the calcium concentration in the cleaning liquids after cleaning.
- Analyze the chemical composition of the remaining fouling after the whole cleaning process. To see what the non-dissolved fouling is composed of.
- Do investigation on how the cleaning order and effect of different concentration and temperature of nitric acid influence the process.

References

Awad, M. M. (2011). *Heat Transfer - Theoretical Analysis, Experimental Investigations and Industrial Systems* A. Belmiloudi (Ed.) *Fouling of Heat Transfer Surfaces* (pp. 40). doi:10.5775/13696

Bansal, B., & Chen, X. D. (2006). A Critical Review of Milk Fouling in Heat Exchangers. *Comprehensive Reviews in Food Science and Food Safety, 5*(2), 27-33. doi: 10.1111/j.1541-4337.2006.tb00080.x

Bird, M. R., & Fryer, P. J. (1991). An experimental study of the cleaning of surfaces fouled by whey proteins. *Trans. ICemE, 69*, 13-21.

Burton, H. (1968). Deposits from whole milk in heat treatment plant - A review and discussion. *Journal of Dairy Research, 35*, 317-330.

Bylund, G. (1995). *Dairy processing handbook*. Lund: Tetra Pak Processing Systems AB.

Changani, S. D., Belmar-Beiny, M. T., & Fryer, P. J. (1997). Engineering and chemical factors associated with fouling and cleaning in milk processing. *Experimental Thermal and Fluid Science, 14*(4), 392-406. doi: http://dx.doi.org/10.1016/S0894-1777(96)00141-0

Coultate, T. (2009). Food protein systems *Food: The Chemistry of its Components* (5 ed., pp. 177-182): The Royal Society of Chemistry.

Foster, C. L., & Green, M. L. (1990). A model heat exchange apparatus for the investigation of fouling of stainless steel surfaces by milk II. Deposition of fouling material at 140 °C, its adhesion and depth profiling. *Journal of Dairy Research, 57*(03), 339-348. doi: doi:10.1017/S0022029900026996

Hagsten, C., Loren, N., Hamberg, L., Wiklund, J., Innings, F., Nilsson, L., . . . Nylander, T. (2013). *A novel fouling and cleaning set-up for studying the removal of milk deposits produced during UHT-treatment.* Paper presented at the Proceedings of International Conference on Heat Exchanger Fouling and Cleaning, Budapest, Hungary.

Jeurnink, T. J. M. (1992). Changes in milk on mild heating: turbidity measurements. *Netherlands Institute for Dairy Research*(46), 183-196.

Jeurnink, T. J. M., & Brinkman, D. W. (1994). The cleaning of heat exchangers and evaporators after processing milk or whey. *International Dairy Journal, 4*(4), 347-368. doi: http://dx.doi.org/10.1016/0958-6946(94)90031-0

Jeurnink, T. J. M., Walstra, P., & de Kruif, C. G. (1996). Mechanisms of Fouling in Dairy Processing. *Netherlands Milk and Dairy Journal, 50*, 407-426.

Kelly, A. L., & Bach Larsen, L. (2010). 1 - Milk biochemistry. In M. W. Griffiths (Ed.), *Improving the Safety and Quality of Milk* (pp. 3-26): Woodhead Publishing.

Lalande, M., Tissier, J.-P., & Corrieu, G. (1984). Fouling of a plate heat exchanger used in ultra-high-temperature sterilization of milk. *Journal of Dairy Research, 54*, 557-568.

Lorenzen, K. (2005). 27 - Improving cleaning-in-place (CIP). In H. L. M. Lelieveld, M. A. Mostert & J. Holah (Eds.), *Handbook of Hygiene Control in the Food Industry* (pp. 425-444): Woodhead Publishing.

Plett, E., & Grashoff, A. (2006). Cleaning and Sanitation *Handbook of Food Engineering, Second Edition* (pp. 929-975): CRC Press.

Science, C. o. (2014). Determination of Total Calcium and Magnesium Ion Concentration.

Singh, P., & Heldman, D. R. (2009). Heat transfer in Food Processing. In S. L. Taylor (Ed.), *Introduction to Food Engineering* (4 ed., pp. 247-269): Elsevier.

Walstra, P., Wouters, J. T. M., & Geurts, T. J. (2005a). Colloidal Particles of Milk *Dairy Science and Technology, Second Edition* (pp. 109-158): CRC Press.

Walstra, P., Wouters, J. T. M., & Geurts, T. J. (2005b). Fouling and Sanitizing *Dairy Science and Technology, Second Edition* (pp. 399-410): CRC Press.

Walstra, P., Wouters, J. T. M., & Geurts, T. J. (2005c). Milk Components *Dairy Science and Technology, Second Edition* (pp. 17-108): CRC Press.

Visser, J., & Jeurnink, T. J. M. (1997). Fouling of heat exchangers in the dairy industry. *Experimental Thermal and Fluid Science, 14*(4), 407-424. doi: http://dx.doi.org/10.1016/S0894-1777(96)00142-2

Xin, H., Chen, X. D., & Özkan, N. (2002). Whey Protein-Based Gel as a Model Material for Studying Initial Cleaning Mechanisms of Milk Fouling. *Journal of Food Science, 67*(7), 2702-2711. doi: 10.1111/j.1365-2621.2002.tb08802.x

Appendix

A. Determine cleaning liquid concentrations

Method to determine cleaning liquid concentrations for nitric acid and sodium hydroxide solutions.

Cleaning concentration of nitric acid:

1. Add 50 mL HNO_3 solution into a 100 mL glass beaker.
2. Add phenolphthalein, 2-3 drops.
3. Titrate the cleaning solution with 0.5 M NaOH using a burette.
4. Note the volume added of 0.5 M NaOH when the color shift occurs.

Calculation

$$C_{HNO_3} = \frac{C_{NaOH} * V_{NaOH}}{V_{HNO_3}} * \frac{M_{HNO_3}}{\rho_{HNO_3}} * 100\ [wt\%]$$

C_{HNO3}	Concentration of cleaning nitric acid liquid in weight %
V_{HNO3}	Volume of cleaning nitric acid liquid in liter
C_{NaOH}	Concentration of sodium hydroxide, mol/ dm^3
V_{NaOH}	Volume of added sodium hydroxide in liter
M_{HNO3}	Molecular mass of nitric acid, 63 g/dm^3
ρ_{HNO3}	Density of nitric acid solution, g/ dm^3

Cleaning concentration of sodium hydroxide

1. Add 30 mL NaOH solution into a 100 mL glass beaker..
2. Add phenolphthalein, 2-3 drops.
3. Titrate the cleaning solution with 2 M HCL using a burette.
4. Note the volume added of 2 M HCL when the color shift occurs.

Calculation:

$$C_{NaOH} = \frac{C_{HCl} * V_{HCl}}{V_{NaOH}} * \frac{M_{NaOH}}{\rho_{NaOH}} * 100\ [wt\%]$$

C_{NaOH}	Concentration of cleaning sodium hydroxide liquid in weight %
V_{NaOH}	Volume of cleaning sodium hydroxide in liter
C_{HCl}	Concentration of hydrochloric acid, mol/ dm^3
V_{HCl}	Volume of added hydrochloric acid, dm^3
M_{NaOH}	Molecular mass of sodium hydroxide, 40 g/mol
ρ_{NaOH}	Density of sodium hydroxide solution, g/ dm^3

B. Chemical analysis

Table 14 Concentration of Calcium, Magnesium and phosphor present in UHT and preheat fouling.

Sample	Ca, mg/L	Mg, mg/L	P, mg/L	Weight, g
UHT 1	2683	159,2	1450	0,5017
UHT 2	2609	151,8	1408	0,4955
UHT 3	2596	153,6	1379	0,4935
Preheat	1837	54,22	860,2	0,4979