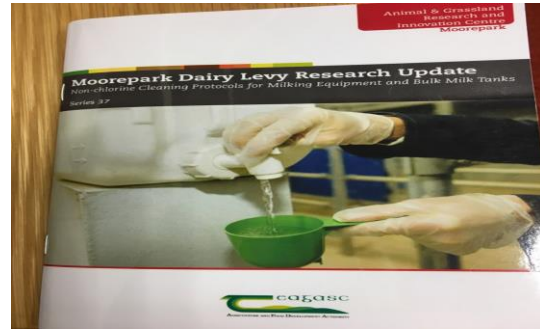


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Examining the interrelationship between milk storage and processing conditions on the quality of milk and dairy products



Key external stakeholders:

Milk processors, advisors, farmers, scientific, chemical manufacturers

Practical implications for stakeholders:

- Milk may be stored on farm at 3 °C for 72 h, and it will have minimal impact on bacterial counts and composition of milk, whether the milk is pre-cooled or not.
- Tracking milk from farm to final product, can aid industry in targeting sources of contamination throughout processing stages and practices to control bacterial numbers.
- Chlorates levels increased 50 fold from raw milk to final product indicating an impact of CIP cleaning and processing conditions on chlorate levels.
- Effective chlorine-free cleaning protocols are now available to industry
- The levels of psychrotrophic bacterial protease(s) tested in milk during storage may not contribute to increase proteolysis in cheese

Main results:

- a. Milk microbiological quality and composition was not affected whether the milk was pre-cooled or not prior to entering the bulk milk tank, however, milk entering the tank should have good initial microbiological quality.
- b. The microbiological quality of SMP produced was better when using mid-lactation than late-lactation milk. The bacterial counts of some CTs and of the WMS samples were higher than predicted using the bacterial counts measured in the farm milk samples, indicating that the transport conditions or cleaning protocols could have influenced the microbiological load.
- c. Iodine levels in mid-lactation reconstituted SMP were higher than that required by manufacturers (100 µg/ L), indicating that the levels in bulk tank milk should be lower than 142 µg/ L. Chlorate levels in late-lactation bulk tank milk were higher than mid-lactation levels and specifications. In both test periods chlorates levels increased 50 fold from raw milk to final product indicating an impact of CIP cleaning and processing conditions on chlorate levels.
- d. Chlorine-free cleaning protocols will maintain the microbiological quality of milk as well as minimizing residues, if used as recommended.
- e. The thermoresistant psychrotrophic bacterial protease(s) may affect the manufacture or quality of Cheddar cheese during ripening to a relatively limited extent.

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1. Project background:

Manipulation of cold storage conditions based on the intended use of the milk would be advantageous for farmers and processors alike to achieve premium quality alongside economic efficiency. From an industry

point of view there are economic savings if milk collection time on farm could be extended to 3 days. However a balance must be achieved between the economic savings and milk quality. A definitive study on milk storage requirements to meet new product specifications and cooling efficiency would provide guidelines for industry on optimum storage temperature and time. In the milk powder manufacturing process, the main concern is the contamination of milk with *Bacillus cereus* and sulphite-reducing *Clostridia* strains, due to their capacity of producing toxins that can cause severe food-borne diseases. Identifying the production stage where milk may be contaminated with such microorganisms needs investigation. No previous study has tracked milk quality from individual farms right through the processing stages to final product. A number of milk residues (TCM & Iodine) have been detected in products manufactured in Ireland in the last number of years. Even if the levels of each residue detected remain within the regulations for product content the presence of such residues can create marketing issues for the export of products in the future. It is critically important that Irish dairy processors can produce lactic butter suitable for the German market. Competition exists between the different exporting countries. Thus, low TCM levels in Irish milk and product are required in order to retain the Irish position within the marketplace. Sodium hypochlorite is used as an effective cleaning agent for the cleaning of milking equipment and bulk milk tanks. However, it is associated with the occurrence of the TCM. An alternative cleaning regime formulated without this cleaning agent is required for cleaning both milking machines and bulk milk tanks and minimizing milk residues. The quality of milk supplied for cheese manufacture is one of the main factors that can affect the quality of the final product. The microbiota of raw milk during cold storage is mainly composed of Gram-negative and Gram-positive psychrotrophic bacteria. The most common psychrotrophs identified in raw milk belong to the genera *Pseudomonas spp.* Most of the lipases and proteases produced by this bacterial species are thermoresistant, are not eliminated after heat treatment, affecting the quality of cheese products.

2. Questions addressed by the project:

- The impact of pre-milk cooling method on the microbiological quality of bulk tank milk
- Tracking the microbiological load of milk throughout low-heat skim milk powder (SMP) manufacturing process, from farm bulk tanks to final powder, during mid- and late-lactation.
- Seasonal differences in the residue levels of chlorate (CHLO), perchlorate (PCHLO), trichloromethane (TCM) and iodine residues in bulk tank (BT) milk, and to monitor those levels throughout a skim milk powder (SMP) production chain.
- Will chlorine-free cleaning protocols maintain good milk microbiological quality?
- The effect of adding to milk different levels of a thermoresistant protease, produced by a *Pseudomonas fluorescens* strain, on the manufacture and quality of Cheddar cheese.

3. The experimental studies:

- **Pre-cooling of milk and storage time:** Three milk pre-cooling treatments were applied prior to milk entering three identical bulk milk tanks: no plate cooler (NP), single stage (SP) and double stage (DP) plate cooler. These pre-cooling treatments cooled the milk to 32.0 ± 1.4 °C, 17.0 ± 2.8 °C and 6.0 ± 1.1 °C, respectively. Milk was added to the bulk tank twice daily for 72 h, and the tank refrigeration temperature was set at 3 °C. The blend temperature within each bulk tank was reduced after each milking event as the volume of milk at 3 °C increased simultaneously. The bacterial counts of the milk volumes pre-cooled at different rates did not differ significantly at 0 h or at 24-h intervals.
- **Tracking of milk from farm to final product:** This study was performed in a milk powder processing plant, where skimmed milk powder was produced using only the milk supplied by the farms involved in this study. Samples of milk were collected from farm bulk tanks (mid-lactation: 67 farms; late-lactation: 150 farms), collection tankers (CTs), whole milk silo (WMS), skim milk silo (SMS), cream silo (CS) and final SMP. In mid-lactation, samples were collected at various points of the manufacturing process between the farm BTs and the SMP [BTs, collection tankers (CTs), whole milk silo (WMS), skim milk silo (SMS) and final SMP] and were tested for a range of bacteria and residues including CHLO, PCHLO and iodine. In late-lactation, samples were collected at various points between the CTs and the SMP [CTs, WMS, cream silo (CS), SMS and final SMP] and were tested for CHLO, PCHLO and iodine.
- **Chlorine-free cleaning protocols:** Newly designed chlorine-free cleaning protocols were designed and successfully tested on research farms. These protocols were subsequently tested on 65 commercial farms in cooperation with 3 milk processors (Dairygold, Glanbia, Carbery, Tipperary).
- ***Pseudomonas fluorescens* strain and cheese manufacture:** Fresh raw milk was collected, standardized and pasteurized at 72 °C for 15 s and the enzyme was added to give a protease

activity of 0.15 or 0.60 U/ L (treatments P1 and P4, respectively), while one sample had no enzyme added (control, C). Milk was stored at 4 °C for 48 h and Cheddar cheese was manufactured after 0 and 48 h of storage.

4. Main results:

- The bacterial counts of the milk volumes pre-cooled at different rates did not differ significantly at 0 h storage time or at 24-h intervals. After 72 h of storage, the total bacterial counts (TBC) of the milk that was not pre-cooled (NP) was $3.90 \pm 0.09 \log_{10} \text{ cfu/ mL}$, while the TBC of the milk volumes that were pre-cooled were $3.77 \pm 0.09 \log_{10} \text{ cfu/ mL}$ (SP) and $3.71 \pm 0.09 \log_{10} \text{ cfu/ mL}$ (DP). The constant storage temperature (3 °C) over 72 h aided in reducing bacterial growth rates in milk, and consequently milk composition was not affected whether the milk is pre-cooled or not; however, milk entering the tank should have good initial microbiological quality.
- During mid-lactation, the raw milk produced on-farm and transported by the CTs had better microbiological quality than the late-lactation raw milk (e.g., total bacterial count (TBC): 3.60 ± 0.55 and $4.37 \pm 0.62 \log_{10} \text{ cfu/ mL}$, respectively). After pasteurisation, reductions in TBC, psychrotrophic (PBC) and proteolytic (PROT) bacterial counts were of lower magnitude in late-lactation than in mid-lactation milk, while thermoduric (LPC – laboratory pasteurisation count) and thermophilic (THERM) bacterial counts were not reduced in both periods. The CHLO, TCM and iodine levels in the mid-lactation milk stored in the WMS ($0.0010 \pm 0.0000 \text{ mg/ kg}$, $0.0009 \pm 0.0000 \text{ mg/ kg}$ and $135.5 \pm 7.6 \mu\text{g/ L}$, respectively) were lower than legislative and industrial specifications (0.0100 mg/ kg , 0.0015 mg/ kg and $150 \mu\text{g/ L}$, respectively); however, in late-lactation, those levels were numerically higher than the mid-lactation levels and specifications ($0.0410 \pm 0.0554 \text{ mg/ kg}$, $0.0020 \pm 0.0007 \text{ mg/ kg}$ and $437.6 \pm 155.2 \mu\text{g/ L}$, respectively). Consequently, CHLO and iodine levels in SMP were numerically higher in late-lactation ($0.1263 \pm 0.0071 \text{ mg/ kg}$ and $398.2 \pm 22.8 \mu\text{g/ L}$, respectively) than in mid-lactation ($0.0570 \pm 0.0090 \text{ mg/ kg}$ and $142.2 \pm 10.0 \mu\text{g/ L}$, respectively). Trichloromethane accumulated in the cream portion after separation.
- No negative effects on bacterial levels in bulk tank milk were observed when chlorine-free cleaning protocols were compared to traditional cleaning systems using chlorine based products used on commercial farms, when cleaning protocols were used as recommended.
- Results indicated that the protease was active in milk during 48 h of storage; however, its effect on the milk composition was minimal. The mean cheese yield (Y%) and recovery of fat and protein (%REC_{FAT} and %REC_{PROT}, respectively) obtained for all cheeses were not affected by protease activity ($P > 0.05$). However, slight increases in proteolysis were observed in cheeses from treatment P4 produced using milk stored for 48 h. Both cheeses had higher concentrations of free amino acids (FAA) compared to the control, while urea-PAGE electrophoretograms indicated a higher breakdown of caseins in the P4 cheese samples, which may be related to possible increases in proteolytic bacteria numbers in milk during storage.

5. Opportunity/Benefit:

- a. Opportunity to store milk on farm for 3 days at 30C without any negative impact on milk quality. The use of the SP pre-cooling systems is recommended to maintain low levels of bacterial counts and reduce energy consumption.
- b. Tracking of milk from farm to final product highlighted issues such as increased chlorate levels in milk powder and microbial load at different stages of processing indicating issues with CIP cleaning.
- c. Reducing iodine intake at farm level will have positive impact on the levels of Iodine in the raw milk supplied for Infant Milk Formula manufacture.
- d. Removing chlorine products for the cleaning of milking equipment and replacing with the new chlorine free cleaning protocols will have a positive impact on TCM and chlorate levels in Irish milk.

6. Dissemination

Information from this project were shared with industry on the Teagasc milk quality webpage, at open days, milk quality workshops, residue industry group meetings and popular articles in the Farmers Journal, Dairy Farm Business. Six peer reviewed papers from this study have been published. Poster and oral presentations were made at the EGF, BSAS and IDF Conferences.

7. Publications:

Paludetti, L.F., Jordan, K.N., Kelly, A.L. and Gleeson, D. (2018). Evaluating the effect of storage conditions on milk microbiological quality and composition. Irish Journal of Agricultural & Food Research IJA FR - 57 - 2018 - 52-62

Paludetti, L.F., Kelly, A.L., O'Brien, B., Jordan, K.N. and Gleeson, D. (2019). Microbiological quality of milk

from farms to milk powder manufacture: an industrial case study. *Journal of Dairy Research* 10.1017/S0022029919000347

Paludetti, L.F., Kelly, A.L., O'Brien, B., Jordan, K.N. and Gleeson, D. (2018). The effect of different precooling rates and cold storage on milk microbiological quality and composition. *Journal of Dairy Science* 101: 1921-1929

Popular publications:

Paludetti, L. P., Kelly, A.L., O'Brien, B. and Gleeson, D. (2018). Iodine levels in bulk tank milk produced by dairy cows during the mid- and late-lactation periods. In: *Advances in Animal Biosciences 2018* (British Society of Animal Science), Dublin, 09-Apr-2018, pg 218

Gleeson, D. (2018). Non chlorine cleaning protocols for milking equipment and bulk milk tanks. *Teagasc IE Series 37 2018 Pg 20 ISSN Teagasc MPK D*

Paludetti, L., Kelly, A.L. and Gleeson, D. (2016). Influence of storage time on the microbiological load of raw milk destined for UHT processing. In: *18th World Congress of Food Science and Technology Proceedings - Greening the Global Food Supply Chain*, Dublin, Ireland, 21-Aug-2016,

Paludetti, L.F., Kelly, A.L., O'Brien, B. and Gleeson, D. (2018). Control of chlorate and trichlormethane residue levels in bulk tank milk. In: *Annual Meeting of the Federation of Animal Science*, Dubrovnik, 27-Aug-2018, P192

Gleeson, D. and O'Brien, B. (2016). Minimizing chlorate levels in the dairy chain. *Irish Farm Business-Dairying* 3 (2) Summer p. 20-22

8. Compiled by: David Gleeson
