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Development of a Process-based milk processing sector model for the Irish Dairy Industry

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MBA

This thesis is submitted in partial fulfilment of the requirements for the degree of

Doctor of Philosophy in Food Science and Technology

UCC Supervisors: Prof. Alan L. Kelly,

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Puneet Parmar, James A. O'Mahony, John T. Tobin, Laurence Shalloo. (2018). Evaluation of butter manufacture process-Mass balance approach to assess fat conversion process in a dairy processing unit. In: Internet of Things 4 Food, UCD, Ireland.

Peer reviewed research articles

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Declaration

I hereby certify that this material, which I now submit for assessment on the programme of study leading to the award of Doctor of Philosophy is entirely my own work. I have exercised care to ensure that the work is original, and does not, to the best of my knowledge, breach any law of copyright, and has not been taken from the work of others, save and to the extent that such work has been cited and acknowledged within the text of my work.

_____

Puneet Parmar Date:

For my father, Amrik,

My wife, Shivani

And My daughter,

Maya

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Summary

In recent times, the EU dairy industry has been hampered by volatility and uncertainty due to changes in the Common Agricultural Policy (CAP) introduced in 2015. Milk quotas, first introduced in 1984, have since been abolished, which has led to unhindered growth in milk production. The unhindered growth in milk production, combined with volatile supply and demand scenarios, has posed challenges to dairy processing sector. The dairy industry continues to face numerous opportunities and challenges, like seasonality, variation in milk composition, low profitability and idle processing capacity. There are several factors that impact the quality of milk and how it is processed in a dairy environment. The studies presented in this thesis provided information that can aid dairy suppliers and processors on making well-informed, business-critical decisions using information generated from the models and density parameters described in the studies.

Firstly, the impact of seasonal variation on milk composition was determined and an algorithm was designed to predict the season-based density of milk based on milk composition. Three separate cow genetic groups were selected, and composite samples (morning+evening) were collected for a period of 9 months. Milk composition parameters like milk fat, protein, and lactose content were determined and the impact of the variation in milk composition on milk density was analyzed using three analytical techniques. The mean density values and standard error of mean estimated for milk samples in each season, i.e., spring, summer and autumn were 1.0304 ±

0.00008 g/cm3, 1.0314 ± 0.00005 g/cm3 and 1.0309 ± 0.00007 g/cm3, respectively. As a result of this study, The estimation of new density factors may enable improvements in the milk payment systems for the production and processing industry.

The density of milk is dependent upon various factors including temperature, processing conditions, and animal breed. Second study carried out as a part of this thesis evaluated the effect of different cow genetic groups, Jersey, elite Holstein Friesians (EHF), and national average Holstein Friesians (NAHF) on the compositional and physicochemical properties of milk. Dataset collected as a part of the first study was analyzed to assess the impact of genetic merit of the animal/herd on milk density. As an outcome of this study, milk density was found to be significantly higher (1.0313 g/cm³ ± 0.00026, $p<0.05$) for the milk of Jersey breed when compared to the EHF (1.0304± 0.00026 g/cm³) and NAHF (1.0303± 0.00024 g/cm³) genetic groups.

The impact of temperature on whole milk density for the milk production and processing sector has not been revisited since the early 1950s. The objective of this study was to determine the effect of temperature on whole milk density measured at four different temperatures :5, 10, 15, and 20°C. The temperatures identified to conduct density trials are important during milk processing within a dairy plant and, therefore, can be used to establish weight-volume relationships and to estimate the variations in yield of products and profitability of the milk conversion processes. It is also worthy to note that, in practice, most density measurements are completed at 20°C at the dairy plant sites, while milk is collected from farms at 4-5 °C. This difference in

temperature (between collection and processing) leads to variance in milk density estimation. The main aim of this study was to help address any bias in weight-volume calculations and thus may also improve the financial and operational control for the dairy processors. The density of milk was assessed using two analytical methods and regression equations describing the inverse relationship between density and temperature were shown. Density values determined at 5 °C was 1.0334 g/cm3, with corresponding figures of 1.0330, 1.0320 and 1.0305 g/cm3 at 10, 15 and 20 °C, respectively.

Finally, the last section of the thesis was aimed at developing and evaluating processing sector model for the butter manufacture process and used the new density factors developed in the previous studies. Two dairy processing sites were selected and butter manufacture process was analyzed using mass balance technique to determine the fat utilization in various subprocesses of butter manufacture. The butter manufacture was studied as a batch process in a closed loop with fat content in each substream recorded and forming an input of the mass balance. Losses at the end of butter production ranged between 1.90% and 2.25% of the total fat input for both sites.

The new density factors also formed as an input to assess three different scenarios deployed for the evaluation of the mass balance model and to estimate best product portfolio/mix and net value of milk was estimated. The three scenarios were: S1 (Animal Breed) high genetic merit (Elite) and national average (NA) Holstein Friesian (HF) cows were evaluated, for their effect on the net value of milk; S2 (Product Portfolio) a mixed product portfolio of

cheese, butter and skim milk powder (SMP) was compared to a product portfolio comprised of butter alone; and S3 (Process Efficiency) the impact of varying process losses on net values of milk and the quantities of products produced was simulated. The value per 1000 L of milk for S1 was €410.69 and €393.20 for Elite and NA cow's milk, respectively. For S2, the butter-only product portfolio returned €355.10, whereas the mixed-products portfolio returned €369.60. Lastly, S3 corresponding returns for 1%, 2.2% and 5% losses was €365.90, €361.47 and €351.12, respectively.

Chapter 1

Literature Review

Declaration: This chapter was written by Puneet Parmar, with corrections and comments from Prof. Alan L. Kelly, Dr. James A O'Mahony, Dr. Shane V. Crowley from University College Cork, Prof. Laurence Shalloo, Dr. John T. Tobin, Dr. Eoin Murphy from Teagasc Moorepark and Dr. Nicolas Lopez-Villalobos from Massey University.

Abstract

In recent times, the EU dairy industry has been hampered by volatility and uncertainty due to changes in the Common Agricultural Policy (CAP) introduced in 2015. Milk quotas, first introduced in 1984, have since been abolished, which has led to unhindered growth in milk production. The unhindered growth in milk production, combined with volatile supply and demand scenarios, has posed challenges to dairy processing sector. The dairy industry continues to face numerous opportunities and challenges, like seasonality, variation in milk composition, low profitability and idle processing capacity. The study attempts to evaluate the applicability and constraints of these models, thus opening up scope for further research. Dairy industry processes require development of mathematical, mass balance and process-based simulation models critical for decision-making and optimization without putting actual processes at risk. The review recommends future research around developing mass balance models for individual constituents of milk.

Key words: dairy processing, modeling, seasonality, mass balance

Abbreviations: MDSM = Moorepark dairy sector model, MPSM = Moorepark processing sector model, GIS= geographical information systems, NMV = Net milk value, MRTS = marginal rate of technical substitution, MCP = Multiple component pricing, GAMS = General Algebraic Modeling system, SMP = skim milk powder, WMP = whole milk powder, BMP = buttermilk powder, NZ= New Zealand, EU= European Union, CAP = Common Agricultural Policy

1. Introduction

The dairy industry is one of Ireland's most important industries and comprises a vital part of the agri-food sector. The Irish dairy industry accounts for approx. 40% of exports in Irish Agriculture (2021) with an approximate annual production of 8,300 million liters of milk in 2020 (CSO 2020). Dairy products accounted for more than 5.4 billion euros of exports in the year 2020 (CSO 2020). The Irish dairy industry is characterized by its dependency on temperate pastures (Shalloo et al., 2014) and, therefore, is affected by seasonal changes having a peak to trough ratio of 6.1:1 (May vs January,2019) with a milk processing capacity utilization of 62.1%. This ratio has improved since 2014 when milk quotas were in place.

For the period from 1984 to 2015, the European Union dairy output was controlled by a milk quota system to stabilize prices and maintain dairy activities in less competitive regions (Witzke, 2009). However, policy reforms around market support had created a situation where there was milk price volatility and farmers were restricted from expansion and thus could do little to protect their business (Geary, Lopez-Villalobos, Garrick, & Shalloo, 2010). Common Agricultural Policy (CAP) reforms abolished market support mechanisms, along with milk quotas in 2015, and milk production in Ireland was expected to increase by 40-50% thereafter (Gleeson, 2017). Between 2014 and 2019 Irish milk production increased by 41.4%.

Various models have been used in the past to identify and evaluate the profitability of dairy processing sector. These models have also helped processors in making strategic decisions regarding product mix and composition. Models are tools which may be used to define a problem, to analyze process-related data, identify the main causes of the problem, and, finally, identify possible solutions to the problem (Heinschink, Shalloo, & Wallace, 2012). Models can be divided into two types: positive models and normative models. Positive models are generally descriptive and are used to describe an actual process and analyze cause-effect relationships. They can be further classified as statistical and econometric models. On the other hand, normative models are prescriptive in nature, giving ideal outcomes for a process

under controlled/theoretical conditions. They are further classified as simulation and mathematical programming models.

Statistical models are generally used to describe variances in data or samples collected. Statistical models use mathematical equations to translate information from the data (Eaton, 2001), whereas econometric models are developed using economic data and statistical inference. Econometric models are based on economic theories and utilized to optimize behavior or a process using economic parameters (Slade, Kolstad, & Weiner, 1993). Simulation models are useful to predict process behavior and may be used in decision-making processes. Simulation modeling is the process of generating and examining a virtual prototype of the actual process to simulate/predict the performance of the process (Snow, 2001). Mathematical programming models may fall into any of the following: linear programming, network optimization, nonlinear programming, dynamic programming, multiple criteria optimization, and stochastic programming (Shapiro, 1993). For example, dairy processing models like the milk transport model (Quinlan et al., 2010), Moorepark dairy systems model (MDSM) (Shalloo, 2004), Moorepark processing sector model (MPSM) (Geary et al., 2010) and the combined farm systems and processing sector model (Geary, Lopez-Villalobos, Garrick, & Shalloo, 2014) have been used to simulate profitability and product mix specific to the Irish dairy industry.

Other than these, models have also been classified as (a) component research, (b) systems research and (c) management models. Examples of component research models include those of McNamara, Huber, and Kenéz (2016) and Turino et al. (2010), who studied metabolism in dairy cattle. McNamara and Shields (2013) studied the reproductive control using a systems research approach while Crosson et al. (2011) reviewed different models studying the greenhouse gas (GHG) emissions from beef and dairy cattle production farms. Farm management models have studied changes in output, including profitability and risks associated, affected by changes in short-term and long-term management approaches.

Examples of management models include those analyzing grazing management (Cros, Duru, Garcia, & Martin-Clouaire, 2003) and management of resources (Castelán-Ortega et al., 2016).

Research to date on milk processing around the world, including, in Ireland, has mainly focused on product mix, seasonality effects of supply profile for processors (Heinschink et al., 2012), and profitability analysis for processors (Geary et al., 2010) and for dairy farmers (Shalloo, 2004). Another model evaluated interaction of farm and processing sector models in conjunction with seasonality of milk production and prices (Geary et al., 2014). Similar models reviewed in this study include a mathematical model assessing returns in cheese manufacture (Burke, 2006; Papadatos, Berger, Pratt, & Barbano, 2002) and models have also been developed to estimate the value of milk based on product mix of fluid milk, cheese, butter and nonfat dried milk (skim milk powder) for the American dairy industry (Bangstra, Berger, Freeman, Deiter, & La Grange, 1988). Another comprehensive model studying the impact of seasonal composition on profitability of New Zealand dairy processors' product mix, i.e., fluid milk, butter, cheese, casein, whole milk powder (WMP), skim milk powder (SMP), whey powder and Buttermilk powder (BMP) was developed by Garrick and Lopez-Villalobos (2000). An optimization model was developed to minimize nitrogen leaching while increasing farm profit in Florida (Cabrera, Breuer, Hildebrand, & Letson, 2005). These models have been instrumental in analyzing the profitability of dairy industry from both farmer and processor perspectives.

This review identifies various factors and models developed across the world and how these models have helped to resolve or respond to issues challenging the dairy processing sector and for the dairy industry in general. These factors include but are not limited to seasonality, profitability analysis, product portfolio and product mix, and milk density. Considering these major factors, different processing sector models were evaluated and are discussed in terms of their application to dairy industry.

1.1 Product portfolio and profitability analysis

Seasonality of milk supply is one of the main talking points in the dairy industry and it has significant impact on the type of products produced, quality of products, acceptability by consumers and also impacts the profitability of the dairy industry significantly. The impact of seasonal variation on milk composition has been studied widely (Bansal et al., 2009; Bernabucci et al., 2015; Festila et al., 2006; Grimley et al., 2009). Seasonal variation in milk composition significantly impacts the milk constituents' content, i.e., milk fat, protein, lactose etc. (Auldist et al., 2016; O'Callaghan et al., 2016; O'Callaghan et al., 2017). The variation in milk constituents, subsequently impacts the choice of products produced. The impact of product mix/portfolio on the profitability and economic analysis has been studied in various processing sector models such as Moorepark Processing Sector Model (MPSM) (Geary et al, 2010), Milk optimisation model (Heinschink et al., 2012), and the dairy production and lactose model (Sneddon et al., 2016) etc. The MPSM is a tool that can be used to quantify the quantity of the products produced from milk and, when combined with their value while subtracting the cost of processing, can used to put a value on milk. This model of the value of milk can be used to help guide the milk pricing systems. Multiple components pricing (MCP) is a method of milk pricing which is used in this model to put a value on fat and protein and determine the price per kg paid to farmers. The MCP was defined as the pricing of milk on the basis of more than one constituent, i.e., fat and protein, fat and lactose, fat and SNF etc. (Emmons, Tulloch, & Ernstrom, 1990). Over the years, various MCP systems (Bailey, Jones, & Heinrichs, 2005; Garrick & Lopez-Villalobos, 2000; Moon, 2015) have been developed to determine component pricing and devise payment methodologies.

The MPSM is a mass balance model which accounts for all inputs, outputs and losses observed during dairy processing. It represents the conversion of milk from the start, i.e., intake into milk plant/silos, conversion process, separation to cream and skim and manufacture process into a relevant product mix. Product portfolio is one of the key

parameters affecting the net value of milk and the returns generated from milk processing. In previous models, simulations indicated the best possible combination of products such as fluid milk and cheese from a product mix that generated maximum revenue (Garrick & Lopez-Villalobos, 2000). Similarly, Papadatos et al. (2002) suggested that any changes in milk and product composition, along with market prices, could alter the ideal product portfolio and needed to be transformed depending upon the demand change. Milk composition changes affects the relative values of fat and protein, so in the model the Marginal Rate of Technical Substitution (MRTS) is used to quantify the relative values of fat and protein. The MPSM was able to address these questions for the Irish dairy industry by simulating different product portfolios and composition.

Another model assessed product portfolio and profitability analysis for New Zealand dairy industry. The processing model for dairy production and lactose (Sneddon et al., 2016) was developed to analyze different production scenarios affected by varying costs, prices and constituents' availability, i.e., fat, lactose and, protein. This model was based on the work done under MPSM (Geary et al., 2010) and studied the same portfolio consisting of WMP, SMP, butter, cheese, and fluid milk. Calculations similar to MPSM were completed to estimate net value of milk, fat, protein, gross income and net revenue. Mass balance models for fat and protein were used to assess the lactose content (surplus or deficit).

Production yields for various products affect the profitability of any dairy company and optimization of yields is a key challenge faced. Predictability of yields could be beneficial for dairy companies to anticipate areas like labour, equipment, and raw material requirements. Controlled monitoring of production processes, along with standardization, may enable accurate prediction of yields for different dairy products. Brito, Niklitschek, Molina, and Molina (2002) developed a mathematical model to predict yields of Gouda cheese for the Chilean dairy industry and compared the predicted yields against the theoretical yields obtained from using different yield equations. Eleven different equations were used by Brito et al. (2002) to

predict the theoretical yield of Gouda cheese using actual values of milk components or whey and cheese. The actual yields of cheese were studied from 8 different vats. Control measures were in place for acidity and pH monitoring, along with quality of milk, whey and cheese. The actual yields of cheese were calculated on the third day after processing and also, at the end of the ripening period (30 days). Of the equations used in the analysis, equation 4 and 7 showed the least difference between theoretical and actual yield. These two equations allowed for prediction of cheese yield even before ripening, which is a very useful application in the dairy industry.

1.2 Milk Seasonality

Several studies in the past have also shown the implications faced by processors due to seasonality. The implications and challenges to processors can include a seasonal milk supply involving unequal distribution over the year with peak and trough supply periods. The composition of milk also changes with transition from mid to late stage of lactation. Milk mineral concentrations change coinciding with the transition from mid to late stage of lactation and poses challenges to processors in terms of varied milk intake volumes, formulation and production of seasonal products and changes in milk functionality (Downey and Doyle, 2007; Quinlan, Keane, O'Connor, and Shalloo, 2012; Hennessy and Roosen, 2003; Guinee, O'Brien, and Mulholland, 2007; Gulati et.al, 2018). Models in the past, Bangstra et al. (1988), Papadatos et al. (2002), and Burke (2006) have been developed to study the correlation between pricing systems, seasonality, and overall profitability for dairy processors.

One such model developed for assessing the impact of seasonal variation in milk production and processing was developed by Heinschink et al. (2012). The Irish dairy industry is characterized by seasonal variations in milk production and processing, which arises due to calving and grass growth patterns, and induces capacity-related constraints on milk processors throughout the country. Constraints also include labor requirements, storage space for produced goods, poor capacity planning and resource utilization. These constraints

add additional stress on the profitability of processors, through increased costs and capacity loss. The model developed by Heinschink et al. (2012) incorporated key factors like product mix, costs, and capacity planning impacting the processors and also estimated the price to be paid to producers (Marginal Producer Milk Price; MPMP). Heinschink et al. (2012) analyzed the MPSM model and its three different scenarios for profitability optimization. Similar plant and labor availability conditions were applied to evaluate the results obtained. The smooth scenario applied a flatter supply profile for calculations, whereas the seasonal scenario studied a more seasonal supply base. The outcomes from the model showed that liquid milk was the most profitable product from the standard product mix simulated. Casein and cheese ranked 2nd and 3rd, respectively, in the margin calculations. Powders were the dominant product from the seasonal scenario, whereas casein was the most profitable product in the smooth scenario simulation. In terms of sales revenue, the smooth profile showed a higher return and highest gross margin for processors with lowest fixed, variable, collection and handling costs.

Milk production seasonality also impacts other parameters like milk transport and handling cost which was assessed in a model by Quinlan et al. (2010). The model incorporated the seasonality changes in Ireland, i.e., highest milk availability from mid-April to August and a lean period in December and January. This model studied the spatial presence of the largest dairy processors of Ireland and also considered the consolidation of dairy production farming. Different scenarios were simulated in the model, such as benchmark scenario (S1) based on 2008 production data, scenario 2 (S2) with 30% higher production than scenario 1 (S1) and same tanker size, and scenario 3 (S3) with the same production as scenario 2 but higher tanker capacity. The resulting costs based on different production data were evaluated. The major costs included, i.e., capital, labor and running costs and were the highest for scenario 2, and total costs were approx. 20% higher in S2 than S1. However, per-liter costs were lower compared to S1 owing to higher volume of production and were distributed over the 12-month period (Quinlan et al., 2006). For S3, the peak supply month accounted for 10% of the total

annual cost, while accounting for 15% of milk supply, and the trough month accounted for 6% of total annual costs while accounting for only 2% annual milk supply.

Geary et al. (2014) developed a model which analyzed the impact of supply seasonality on dairy production and processing sector. The supply pattern of milk was changed from seasonal to less-seasonal. Cost and profit comparison were completed for both scenarios, and it was shown that a less seasonal profile allowed better capacity utilization and provided higher net returns to processors but, on the contrary, the costs related to operations on a farm increased significantly for such a production scenario. The model of Geary et al. (2014) analyzed the impact of changes within the confines of a dairy farm, and also analyzed the impact of changes introduced from the supply pattern on the processing sector. From the farming perspective, the changes in calving pattern were studied and generated supply profiles using the national pool; while from a processing perspective, the model used the supply patterns, i.e., volume and composition, to estimate the production of chosen product portfolio. The model also evaluated returns for processors identifying the ideal product mix, which generated maximum profits.

1.3 Milk Density

Milk composition and its impact on dairy processing and the portfolio of products produced has been widely studied in the past but not in the resent literature. Several factors, significantly impact the composition of milk, such as breed and genetic groups, feeding pattern, the impact of seasonal changes and climatic conditions, animal health and management practices including feed and farm management (Fox et al., 1998; Grimley et al., 2009; O'Callaghan et al., 2016; Parmar et al., 2020). Milk composition in Ireland has been observed to vary significantly, with fat content decreasing from January to July period of the year and increasing in the August and September periods. Similarly, protein content was noted to decline from November to the April period and increased from July to November period (Dairyco 2018; Parmar et.al, 2020). Composition of milk is one of the most significant factors

that affects physical attributes of milk like density (Walstra, 1999). Variations in milk density are observed as a result of variation in the content of solids-non-fat and fat content, with higher fat content giving lower milk density and vice versa (Short, 1955). Milk density is directly correlated with the fat globule size of milk, which usually varies between 0.1 to 15 µm (Wiking, 2004). Fat globule size of milk is dependent upon certain factors like feed, physiology and genetics of the animal, lactation stage and seasonal and climatic changes (Heck, Van Valenberg, Dijkstra, & Van Hooijdonk, 2009; Mulder & Walstra, 1974; Parmar et al., 2020). Breed or genetic merit of the animal has been shown to impact the fatty acid content of milk, which affects milk fat content and, thus, density. Milk from different breeds of cows has varying fatty acid content, which affects the overall fat concentration and also affects the fat globule size (Marin et al., 2018). Past research has shown that certain breeds of cow produce higher fat and protein yield compared to other breeds; Jersey milk have higher yield of protein and fat compared to Holstein cow milk (Auldist, Johnston, White, Fitzsimons, & Boland, 2004). A comparison of Danish Jersey, Swedish red and Danish Holstein cows also showed that there was a significant difference in concentration of protein in milk (Gustavsson et al., 2014); Danish Jersey milk had the highest protein percentage (4.30%) compared to Red (3.70%) and Holstein milk (3.40%).

Milk density fluctuates between the range of 1.025 to 1.035 g/cm^3 (Scott et al., 1998). Seasonal changes in milk density values are observed, with higher milk density observed in summer and lower density in winter. Other factors that impact milk density include milk temperature (Short, 1955, Parmar et al., 2020), processing conditions, and processes like agitation and homogenization. The effect of parameters like temperature and pressure and their impact on milk density have been assessed in the past. Thermal treatment of milk impacts the fat globule size by affecting the crystallization of fat globules, which directly affects density (Huppertz and Kelly, 2006; Mulder and Walstra, 1974). Other studies also showed that the density of milk decreased as the temperature of milk increased from 0 to 40 °C. Pasteurization

process negligibly impacted the density, while heating the milk sample to a temperature of 95°C decreased whole milk density (Short, 1955, 1956). The density of milk changes within a temperature range of 0–60 °C (Guignon, et al., 2014) with the density reducing from 1.0338 g/cm^3 at 0.5 °C to 1.0296 g/cm^3 at 20 °C, and further decreases with increasing temperature (1.0220 g/cm^3 at 40 °C and 1.0132 g/cm^3 at 60 °C).

The density of milk is an important physical characteristic that is widely used for weight-volume calculations, product mix management and profitability calculations. The density of milk is used to convert the volume of milk entering a processing environment to weight/mass of milk. The weight of individual constitutents in milk can then be determined by multiplying the mass of milk entering the processing system by the constituents' percentage. This forms an important data point in developing processing models and simulating milk processing. Seasonal variations in milk composition, along with inaccurate milk density conversion factors, pose a significant challenge to the processing industry. Mass balance models developed around the world have formed the basis of determining the available mass in a system. In general, the principle of a mass balance is based on the law of conservation of mass. A mass balance equation (Warn and Brew, 1980) is represented as :

Mass in = Mass out + Mass stored

Or

Raw Materials = Products + Wastes+ Stored Materials

Milk density plays a crucial role in determining available fat mass in a process with accuracy. Volume of milk multiplied by density of milk gives the mass (weight) of milk.The density factor can also be used to calculate the individual milk constituents present in milk. Several processing sector models (Bangstra et al., 1988; Burke, 2006; Geary et al., 2014; Papadatos et al., 2002) have been developed in the past using mass balance approaches and this technique has found significant use in industries such as climate studies (Medwedeff and Roe, 2017), environmental monitoring (Ashfaq et al. 2017; Irvine et al. 2017), and chemical

analysis (Little et al. 2014; Chen et al. 2015). In dairy processing, mass balances have been used in estimation of milk constituents like fat protein and lactose (Bangstra et al. 1988; Garrick and Lopez-Villalobos, 2000; Bailey et al. 2005; Geary et al. 2010; Sneddon et al. 2016). A mass balance may be described using mass balance equation as mass in = mass out + mass stored. This works on the principle of law of conservation of mass. Mass balance was used in the butter manufacture process at dairy processing sites and may be shown as:

Fat Intake = Fat in products + Fat losses + Recycled Fat + Excess fat sold

where, *Fat intake* = fat content of the total milk volume processed (kg); *Fat in products* = fat in each of the products produced (kg); *Fat losses* = fat lost during processing (kg); *Recycled fat* = fat collected from cleaning-in-place (CIP) activities such as cream silo flush and churn residue flush and sent into separation again (kg); *Excess fat sold* = any fat not used in the production of products sold to internal/external customers (kg).

This literature review of some of the major dairy processing sector models from the global dairy processing industry highlighted the importance of one critical factor, milk density, which was not accounted for while developing and simulating milk processing scenarios. Milk density's relationship with factors such as animal breed, seasonal and compositional variations in milk and milk temperature have not been analyzed for the dairy industry in the past, and previous research on a temperature-density relationship was completed many years ago (Short, 1955;1956). The compositional profile of milk has altered considerably since then, due to improvements in animal genetics, health and physiology, management practices, feeding regimes and other factors, thus requiring current density factors to be evaluated.

1.4 Conclusions

An assessment of the previous models gave insights into the model-based solutions catering to the issues affecting the dairy industry. These models were effective tools of their time, capable of producing real-time analysis and facilitated managers and stakeholders in decision-making. These models covered the gap in knowledge and practice applicable then

but, with recent changes in dairy policies across the EU, a high level of volatility and uncertainty has crept into the dairy value chain. Producers have an unlimited scope of expansion but there is only limited intake capacity at the processors' end. Milk and commodity prices are in turmoil and focus is aligning towards value-added products and processes.

The dairy value chain needs to be tailored to varying nutritional needs of today's consumers. Due to specific nutritional demands, milk constituents like fat, protein, and casein have become even more important and dairy processors need to be more focused and agile to service these demands. Models developed to date have attempted to counter seasonal variation, but the emphasis had been on the entire product portfolio or a few selected products only. By targeting milk constituents, processors can align themselves as per product demands and also monitor any wastages or losses in the process. Therefore, the emphasis of future research should be towards developing mass balance models for individual constituents of milk. This will enable processors to keep a track of their manufacturing efficiency and can focus on key areas in processing. The industrial dairy manufacturing process requires development of a mathematical, process-based simulation model critical for decision making and optimization without putting the actual commercial practice at risk. Seasonal variation in milk composition is also one of the factors that affects processors, and future research assessing this problem will address an important parameter in profitability evaluation. Specific factors like compositional changes and density variations need to be studied to address the issue of seasonality impacting the processors. A model analyzing the inter-relationship of compositional changes (individual constituents), product portfolio, temperature and density variations, and processing capacities could be a useful tool in countering the critical issue of seasonality.

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Study Objectives

Considering the gaps in knowledge and current challenges facing dairy suppliers and processors, the major objective of this study was to investigate how factors like seasonal and compositional changes, temperature and cow genetic group affect milk density. Milk density is useful in estimating the mass of constituents in milk supplied to dairy processors and impacts the subsequent processing of milk to milk products in a dairy environment. The studies were conducted in research dairy farms and in two commercial milk processing plants, which ensured that milk was produced and processed according to typical farm and industrial conditions.

The specific objectives of this thesis were to:

- Determine the effect of seasonal and compositional changes on milk and its impact on milk density, and determine season-based density factors for use in dairy industry;
- Investigate the impact of milk temperature and animal genetic group on milk density and determine density factors for four different temperatures and three genetic groups;
- Determine the losses within a dairy processing environment using a mass balance technique, develop, evaluate and validate a mass balance model for the milk fat conversion process, and apply the model across two dairy processing sites in Ireland.

Chapter 2

The Effect of Compositional Changes Due to Seasonal Variation on Milk Density and the Determination of Season-Based Density Conversion Factors for Use in the Dairy Industry

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Declaration: Milk collection (Puneet Parmar and Arleen McDonagh), analysis and determination of milk composition and density were conducted by Puneet Parmar at Teagasc, Moorepark Food Research Centre. Dr. Laurence Shalloo gave guidance regarding the experimental design and analysis. All experimental data were analysed and the chapter written by Puneet Parmar, with corrections and comments from Prof. Alan L. Kelly and Dr. Shane V. Crowley from University College Cork, Dr. Laurence Shalloo, Dr. John T. Tobin and Dr. Eoin Murphy from Teagasc Moorepark and Dr. Nicolas Lopez-Villalobos from Massey University.

Abstract

The objective of this study was to determine the effect of seasonal variation on milk composition and establish an algorithm to predict density based on milk composition to enable the calculation of season-based density conversion calculations. A total of 1035 raw whole milk samples were collected from morning and evening milking of 60 spring-calving individual cows of different genetic groups, namely Jersey, Elite HF (Holstein–Friesian) and National Average HF, once every two weeks for a period of 9 months (March–November, 2018). The average mean and standard deviation for milk compositional traits were 4.72 ± 1.30% fat, 3.85 ± 0.61% protein and 4.69 ± 0.30% lactose and density was estimated at 1.0308 ± 0.002 g/cm3. The density of the milk samples was evaluated using three methods: a portable density meter, DMA 35; a standard desktop version, DMA 4500M; and an AOAC method using 100-mL glass pycnometers. Statistical analysis using a linear mixed model showed a significant difference in density of milk samples ($p < 0.05$) across seasonal and compositional variations adjusted for the effects of days in milk, parity, the feeding treatment, the genetic group and the measurement technique. The mean density values and standard error of mean estimated for milk samples in each season, i.e., spring, summer and autumn were 1.0304 ± 0.00008 g/cm^3, 1.0314 ± 0.00005 g/cm^3 and 1.0309 ± 0.00007 g/cm3, respectively.

2. Introduction

Milk and dairy products are important components in the majority of western diets. The composition of milk significantly impacts the quality of final products, acceptability by consumers, and profitability of the dairy industry (Amenu and Deeth, 2009) . Over the past years, multiple studies have been performed to assess variations in the composition of milk. Several factors have been found to be directly or indirectly linked to the changes in milk composition (Lindmark et al., 2003; Botaro et al., 2008; Bansal et al., 2009; Heck et al.,2009) . Some of these factors include breed and genotype effects, changes in feeding systems, and the impact of seasonal changes and climatic conditions (Fox and McSweeney, 1998; Grimley, Grandison and Lewis, 2009; O'Callaghan et al., 2016; O'Callaghan et al., 2017; Kljajevic et al.,2018). Climatic conditions may include high temperature variations, microclimate and cold weather conditions. High temperatures may induce heat stress in animals and heat stress has been observed for milk characteristics in Italy (Bernabucci et al.,2015) and fatty acid composition in Swiss (Collomb et al., 2008), Swedish (Lindmark et al., 2003) and Dutch milk (Heck et al.,2009).

Other factors linked to milk composition include lactation stage (Stoop et al.,2009), animal health (Moran et al.,2018), herd management and farm and feed management practices (Adler et al.,2013; Soberon et al.,2011). The effect of processing on milk composition such as chemical composition, amino acids and fatty acid profile were studied in Ireland (Lin et al., 2017; Mehra et al., 1999; O'Brien (3) et al., 1999; O'Brien (1) et al., 1999; O'Brien (4) et al., 1999; O'Callaghan et al., 2017) and other parts of the world (Smit et al., 2000; Chion et al., 2010; Chen et al., 2014). It has been reported that the availability and concentrations of different constituents of milk, such as fat and protein along with other physico-chemical properties, vary throughout a year (Dairyco, 2013; Chen et al., 2014). This has been mainly attributed to the changes in feeding pattern and the stage of lactation (Bansal et al., 2009). When cows are grazed outdoors, changes in the feed are induced due to variable climatic

conditions and growth stages of the grass that can introduce changes into the milk composition on a frequent basis. Change in the feed type and its effect on milk composition was studied (Kelly et al., 1998) while significant compositional variations were observed when the diet was switched from silage-based to pasture-based and vice versa (Elgersma et al., 2004).

Significant variations in fat concentration, fatty acid profile and cheese yield in relation to feed patterns were reported in the past (Auldist et al., 2016; O'Callaghan et al., 2016; O'Callaghan et al., 2017). Similarly, alterations in feed leading to changes in milk composition have a significant effect on product quality (O'Callaghan et al., 2017; Gulati et al., 2018). Milk fat and protein content are the two main components that vary significantly due to seasonal variability in feed (Larsen et al., 2010). A study in the UK showed that the fat content in bovine milk collected between 2009–2013 decreased from January to July, followed by a sharp increase in August and September, remaining constant thereafter (Dairyco, 2013), while protein content declined steadily from November to April (3.35% to 3.23%), remained constant (April to July), and increased marginally thereafter (Dairyco, 2013).

Milk composition affects physical attributes like density (Walstra, 1999) and, thus, the basis of weight–volume calculations in the dairy processing industry. Changes in density are closely related to solids-non-fat content and fat content of milk (Short, 1955), higher milk fat represents lower density and vice versa. The density of milk fluctuates between 1.025 to 1.035 g/cm3 (Scott et al., 1998) with seasonal changes throughout the year.. Density has also been noted to be dependent upon other factors such as temperature and processing conditions like agitation and homogenization (Rutz et al., 1955; Sodini et al., 2004).

The density of milk within a temperature range of 0–60 °C has been studied (Guignon et al., 2014); the density reduced from 1.0338 g/cm^3 at 0.5 °C and to 1.0296 g/cm^3 at 20 °C, while further decreasing with increasing temperature (1.0220 g/cm^3 at 40 °C and 1.0132 g/cm^3 at 60 °C). The physical state of fat globules becomes important at different temperatures, with crystallisation at lower temperatures (higher density) and melting of fat at higher temperatures

(lower density) (Murthy et al., 2016). The impact of seasonal variation in milk composition profile has been assessed by various studies in the past, but its impact on milk density has not been studied extensively. Milk density is an important parameter in the dairy industry for estimating weight–volume relationships. In dairy processing, milk is supplied in volume (litres) while the final product mix is usually measured as mass/weight (kg), which may introduce variations in measurement. Current practice includes using an average single annual density factor to convert weight to volume; however, milk composition profile varies with different parameters, as stated earlier. Therefore, the use of a single density conversion factor for the weight–volume relationship in a processing environment is not representative of the seasonal changes in milk composition and may cause incorrect estimation of milk constituents (as it does not account for variations in composition observed over different seasons) highlighted in later sections.

The current study was designed to assess seasonal changes observed in raw milk composition by monitoring variations in individual milk constituents over a period of 9 months, covering spring, summer and autumn periods in Ireland. These seasonal changes in raw milk profile were then correlated with milk density to establish a density–composition relationship. The density–composition relationship helped to evaluate patterns of variation in density across different seasons and determine season-based density conversion factors which can be used by dairy processors to accurately estimate the yield of products and profitability of individual processors and the dairy industry as a whole.

2.1 Materials and Methods

2.1.1 Experimental Design and Sample Collection

The experiment was carried out over a period of approximately 9 months from March 2018 to November 2018, divided into spring (March, April and May), summer (June, July, August) and autumn (September, October and November) seasons. Raw whole milk samples from spring-calved cows was collected from evening and morning milking from the Teagasc Research

farm, Kilworth, Co. Cork (Latitude 50°07′ N, Longitude 08°16′ W). In a spring calving system, cows are calved close to the time when grass grows rapidly, allowing farmers to maximise production from grazed grass, subsequently positively impacting the profitability of their farm. Cows were selected based on their economic breeding index (EBI) (genetic merit) and the individual animal performance. The genetic groups assessed in this study included Jersey and Elite and National Average genetic merit Holstein–Friesian cows. All the cows (n = 60 total, 20 of each genetic group) included in the study were healthy and milked twice a day at 0700 and 1500 h.

Days in milk (DIM) was used as a parameter in the analysis for variation in milk density with season and stage of lactation. The spring calving period for the cows used in this study started at the end of January and continued until the third week of March. Spring season was classified for samples collected between March to May (DIM = 1–123), summer season for samples collected between June to August (DIM = 79–210) and autumn season for samples collected between September to November (DIM = 173–299), respectively.

The cows were also segregated into three groups, for each breed, based on feed. Between six and seven cows from each genetic group were selected based on EBI to be included for each diet pattern and were classified as control, high concentrate and low grass allowance groups [50]. The description of feed allowance is given below.

(a) High grass allowance: Stocking rate of 2.75 cows/ha, 250 kg N/year. Three kg of concentrate was offered per cow per day immediately post calving to supplement pasture availability in the spring for 12 weeks. Pasture was allocated in accordance with best management practice (approx. 4.5 cm post grazing residual). A grass only diet was offered in the autumn period for 12 weeks.

(b) High concentrate system: Stocking rate of 2.75 cows/ha. Concentrate (7 kg) was offered per cow per day immediately post-calving to supplement pasture availability in the spring for

12-weeks. Supplementation of 4 kg/day per cow was offered in the autumn period for 12 weeks.

(c) Low grass allowance: Similar to control with a lower post-grazing residual of 3.5–4.0 cm in spring and autumn.

A total of 1035 milk samples (combined morning + evening milk), approx. 150 mL each, were collected during this period and each of the samples were tested for compositional profile and whole milk density. The evening samples were collected once every two weeks and stored in a standard refrigerator at 4–5 °C overnight to prevent spoilage, while morning samples collected the next morning were then mixed with these to create a representative sample for analysis. The samples were proportionately mixed based on milk yield for the morning and evening milking to ensure that a representative sample was prepared, which was then properly agitated to ensure thorough mixing of constituents and to remove errors due to settling. Sampling requirements were in accordance with ISO 707:2008 (Milk and Milk Products: Guidance on sampling).

2.1.2 Sample Analysis

The following parameters were tested during the process: milk fat, protein and lactose content and raw milk density. A sample of approximately 30 mL was required for testing on the Dairyspec infrared manual FT model (Make-Bentley systems, Chaska, MN, USA) calibrated for raw whole milk compositional analysis. Milk density (measured at 20 °C, for all three equipment) was determined using three different pieces of equipment, i.e., DMA 35 portable density meter, DMA 4500 desktop density meter (Make-Anton Paar GmbH, City, UK) and 100-mL calibrated glass pycnometers (Make-BRAND GMBH + CO KG, City, Germany), following the procedure described by AOAC standard 925.22.

Before analysis, the density meters were calibrated using distilled water. The measured density of water on DMA 35 was 0.9974 g/cm3 and, for DMA 4500, it was 0.99826 g/cm3. The values fall under permissible limits of the theoretical value of 0.9982 g/cm3 for water at 20 °C.

DMA 35 is commonly used for density measurement across industry due to its easier handling and manoeuvrability. DMA 35 works on the FTIR (Fourier transform infrared spectroscopy) principle of a hollow oscillating U-tube technology; the principle of operation is based on changing frequency of a hydrogen-filled hollow oscillator when filled with different liquids. The mass and density of the liquid changes the natural frequency of the oscillator due to overall change in mass of the oscillator when a liquid is added into the tube. The DMA 4500 also works on the similar principle of FTIR as described above. DMA 4500 has an operational range of temperature 0–100 °C and takes only 1–2 mL of sample for density measurement. The equipment is capable of automated cleansing and introduces immediate temperature equilibrium. The measurement principle and method of operation makes it robust and independent of manual interference, thus, reducing risk of errors in measurement. The sample was tested on the DMA 35 with approx. 1–2 mL sample drawn directly from the sample container, and density was noted from the display screen of the equipment. Syringes (2 mL) were used to inject the samples into the oscillating tubes of the DMA 4500 equipment, preventing the flow of air into the sample. Additional sample could be injected into the equipment if air bubbles were noticed on the display, which enabled optimization of the sample measurement to eliminate any errors.

The third method of measuring density was the AOAC 925.22 official method for determining the specific gravity of a liquid using pycnometer. The densities of liquids attained from the pycnometer method are obtained against water. In this method, firstly, an empty glass pycnometer was weighed and noted. The glass pycnometer was then filled with distilled water and wiped dry to remove any water molecules on the outer surface of the pycnometer. This filled weight was then measured and noted, after which the pycnometer was emptied completely. The pycnometer was then filled with liquid (milk) and the outer surface was wiped dry and weighed again. Excess liquid or water from the pycnometer was removed from the

pycnometer through a capillary action of the pycnometer lid. The density of the liquid against water was measured using the formula

$$Density = \frac{WS - WE}{WW - WE}$$

where WS is the weight of the sample-filled pycnometer, WE is the weight of the empty pycnometer, and, WW is the weight of the water-filled pycnometer.

2.1.3. Statistical Analysis

The data for each sampling run were collected and collated for profile and density values for each season. The collected data were firstly analyzed to estimate the distribution of composition throughout the monitored period. Descriptive statistics (mean, standard deviation, minimum and maximum values) for density and milk compositional profile were determined using the MEANS procedure of SAS 9.4 (SAS Institute, Cary, NC, USA). Analyses of variance of the dependent variables (contents of fat, protein and lactose and density) were performed with a linear mixed model using the MIXED procedure of SAS 9.4 (SAS Institute, Cary, NC, USA). The model included the fixed effects of the genetic group, the feeding treatment, parity, the analytical approach for density measurement, days in milk with the linear and quadratic effect as the covariate and random effects of the cow and residual error.

A prediction model was developed using the linear mixed model for estimating density values considering the feeding treatment, the season, the measurement instrument, the genetic group, parity, the interaction between genetic group and the season, the linear effects of percentages of fat, protein and lactose, the linear and quadratic effects of days in milk, and random effects of the cow.

2.2. Results

A total of 1035 samples (combined morning + evening) were collected and analyzed to obtain the descriptive statistics results shown in Table 2.1. The average fat content in milk samples was 4.72 ± 1.30%, and protein, casein, total solids and lactose contents were 3.85 ± 0.61%, 2.88 ± 0.58%, 14.02 ± 2.65% and 4.69 ± 0.30%, respectively, while average density for the

study period was estimated at 1.0308 ± 0.0021 g/cm3. Table 2.1 also shows the somatic cell count (SCC), calculated as somatic cell score (SCS = log10 (SCC)), which is a marker for hygienic quality of milk samples. The somatic cell score (SCS) average was estimated at 4.66 ± 0.48, while the average somatic cell count was estimated at ~93,300 cells/mL. The somatic cell score calculated for the period of study had no significant impact on milk density found during analysis ($p > 0.05$). Table 2.2 shows the variations in the composition of milk constituents along with the standard error of the mean with fat contents; there was no significant difference between the seasons of spring (5.00 ± 0.14%) and autumn (5.13 ± 0.14%), while a significantly lower fat content ($p < 0.05$) was obtained in summer (4.71 ± 0.11%). On the other hand, protein content for each season was not significantly different ($p > 0.05$) (3.93 ± 0.05% protein in spring, 3.86 ± 0.04% protein in summer and 3.92 ± 0.05% protein in autumn) and lactose content varied significantly in autumn ($p < 0.05$) compared to the seasons of summer and spring (4.59 ± 0.26% in spring, 4.62 ± 0.17% in summer and 4.68 ± 0.31% in autumn). There was a significant difference in casein content in summer and spring season ($p < 0.05$), while no significant difference was found in casein content for autumn compared to spring and summer (3.00 ± 0.06% in spring, 2.91 ± 0.04% in summer, and 2.93 ± 0.05% in autumn). The total solids content with standard error of mean was significantly different ($p < 0.05$) for autumn when compared to spring and summer (13.95 ± 0.37% in spring, 13.68 ± 0.32% in summer, and 14.72 ± 0.37% in autumn). Descriptive statistics for the complete dataset showed that the minimum density was observed in April, at 1.0298 ± 0.0016 g/cm3, while maximum density was observed in the autumn period (November at 1.0316 ± 0.0022 g/cm3).

Table 2.1. Descriptive statistics of milk composition, somatic cell score and density in milk in samples (n = 1035) collected from Jersey (n = 20) and Elite (n = 20) and National Average (n = 20) Holstein–Friesian cows over a period of 9 months (Mar–Nov 2018).

Trait	Mean	SD	Minimum	Maximum

Fat, %	4.72	1.30	2.14	14.86
Protein, %	3.85	0.61	1.76	5.95
Lactose, %	4.69	0.30	2.45	5.61
Casein, %	2.88	0.58	0.61	5.00
Total Solids, %	14.02	2.65	8.66	22.48
SCS (SCC × '000) [1]	4.66 (93.3)	0.48 (3.35)	3.00 (1)	6.39 (2452)
Density, g/cm3	1.0308	0.0021	1.0153	1.0378

[1]Somatic cell score (SCS) calculated as = $\log_{10}$(SCC), SCC = somatic cell count measured in '000 cells/mL.

Table 2.2. Least squares means and standard error of the mean (SEM) of milk composition in samples (n = 1035) collected from Jersey (n = 20) and Elite (n = 20) and National Average (n = 20) Holstein–Friesian cows over a period of 9 months (Mar–Nov 2018).

Trait	Season	Mean	SEM
Fat, %	Spring	5.00 [a]	0.14
	Summer	4.71 [b]	0.11
	Autumn	5.13 [a]	0.14
Protein, %	Spring	3.93 [a]	0.05
	Summer	3.86 [a]	0.04
	Autumn	3.92 [a]	0.05
Lactose, %	Spring	4.59 [a]	0.26
	Summer	4.62 [a]	0.17
	Autumn	4.68 [b]	0.31
Total Solids, %	Spring	13.95 [a]	0.37
	Summer	13.68 [a]	0.32

	Autumn	14.72 [b]	0.37
Casein, %	Spring	3.00 [a]	0.06
	Summer	2.91 [b]	0.04
	Autumn	2.93 [a]	0.05

[a,b,c] Means with different superscript within each milk component are significantly different (p-value < 0.05).

As shown in Table 2.3, the highest density value was obtained for the summer season (1.0314 ± 0.00005 g/cm^3) while the lowest density value was estimated for the spring season (1.0304 ± 0.00008 g/cm^3) and autumn had an intermediate density value of 1.0309 ± 0.00007 g/cm^3. There were significant differences in density values for all the seasons (p < 0.05), with greatest difference being between spring and summer season (0.001 g/cm^3). All the parameters, i.e., the season, the feeding treatment, the instrument, the genetic group of the animal, parity, the days in milk, and the days in milk squared as well as milk constituents, i.e., fat, lactose and protein, had a significant effect on the variation in milk density (p < 0.05), as also shown by the probability values estimated for the factors during analysis (Table 2.4). The interactive effect of genetic group and season was the only factor which was not significant (p > 0.05), while parity of the animal was also a significant factor and could be included as a parameter in the model. Further analysis of results from the linear mixed model procedure showed significant differences (p < 0.05) between measurement techniques (pycnometers and DMA4500, pycnometers and DMA35) but no significant difference between the results for DMA35 and DMA4500. Table 4 also shows the parameters of a linear model to predict milk density, including the season, the feeding treatment, the measurement instrument, the genetic group, parity, the interaction between the genetic group and the season, the linear effects of percentages of fat, protein, lactose, the linear and quadratic effects of days in milk, and random effects of the cow.

Table 2.3. Least squares means and standard error of the mean (SEM) of milk density in samples (n = 1035) collected from Jersey (n = 20) and Elite (n = 20) and National Average (n = 20) Holstein–Friesian cows over a period of 9 months (Mar–Nov 2018).

Season	Mean	SEM
Autumn	1.0309 [b]	0.00007
Spring	1.0304 [a]	0.00008
Summer	1.0314 [c]	0.00005

[a,b,c] Means with different superscript are significantly different (p-value < 0.05).

Table 2.4. Estimates of parameters and p-values of a linear model to predicted milk density, including the season, the feeding treatment, the measurement instrument, the genetic group, parity, the interaction between the genetic group and the season, the linear effects of percentages of fat, protein, lactose, the linear and quadratic effects of days in milk, and random effects of the cow, in Jersey (n = 20) and Elite (n = 20) and National Average (n = 20) Holstein–Friesian cows.

Effect	Genetic Group	FT	Season	Instrument	Parity	Estimate	p-Value
Intercept						1.00700	
FT							0.024
		HC				0.00012	
		HGA				9.26×10^{-6}	
		LGA				0.00000	
Season							<0.0001
			Autumn			−0.00054	
			Spring			−0.00097	
			Summer			0.00000	
Instrument							<0.0001
				Pycnometer		0.00205	
				DMA35		−0.00006	

Effect	Genetic Group	FT	Season	Instrument	Parity	Estimate	*p*-Value
				DMA4500		0.00000	
Genetic Group							<0.0001
	Elite HF					0.00009	
	Jersey					0.00036	
	NA HF					0.00000	
Parity							0.0037
					1	0.00035	
					2	0.00032	
					3	0.00044	
					4	0.00041	
					5	0.00023	
					6	0.00053	
					8	0.00000	
Genetic group × season							0.5545
	Elite HF		Autumn			−0.00002	
	Elite HF		Spring			−0.00015	
	Elite HF		Summer			0.00000	
	Jersey		Autumn			−0.00003	
	Jersey		Spring			−0.00016	
	Jersey		Summer			0.00000	
	NA HF		Autumn			0.00000	
	NA HF		Spring			0.00000	
	NA HF		Summer			0.00000	
dim						−0.00002	<0.0001
dim * dim						6.713×10^{-8}	<0.0001
Fat						−0.00066	<0.0001
Protein						0.00305	<0.0001
Lactose						0.00342	<0.0001

(Elite HF = Elite Holstein–Friesian, NA HF = National Average Holstein–Friesian; FT = feeding treatment, HC = high concentrate feeding, HGA = high grass allowance, LGA = low grass allowance; dim = days in milk).

The expression from the model developed incorporating all relevant factors may be presented as below:

$$\rho = 1.007 - 0.00054 * autumn - 0.00097 * spring + 0.00009 * Elite + 0.00036 * Jersey + 0.00035 * Parity - 0.00002 * dim + 0.00000006713 * \mathrm{dim} * \mathrm{dim} - 0.00066 * \mathrm{Fat} + 0.00305 * \mathrm{Protein} + 0.00342 * \mathrm{Lactose}$$

2.3. Discussion

2.3.1. The Effect of Seasonal Variation and Photoperiod on Milk Composition

The effect of seasonal variation and other factors on milk compositional profile has been extensively studied in the literature in the past (Lindmark-Månsson et al., 2003; Botaro et al., 2008; Ozrenk and Inci, 2008; Bansal et al., 2009; Festila et al., 2012; Bernabucci et al., 2015) . However, the most important parameters that affect milk composition are diet/feed and the stage of lactation (Bansal et al., 2009; Gulati et al., 2018). The lactation period significantly affected the milk composition, with late-lactation milk having higher fat and protein content as compared to mid-lactation (Gulati et al., 2018). The results of this study also align with (Gulati et al., 2018), wherein the fat and protein contents were higher during the later phase of lactation, lowest in the spring period and highest in the autumn period. The density of milk has previously been shown to be dependent on fat and solids-non-fat (SNF) content in milk, and is normally measured at 20 °C (Scott et al., 1998). The results from our study show the variation in milk density with season and compositional changes, where the density values in the summer season (lowest fat content) were highest and comparatively lower (1.0309 g/cm^3) in the autumn samples (with higher fat content). Factors such as somatic cell count(SCC) were not exclusively included in our analysis. SCC is the number of white blood cells, entering the milk as a first line of defense against infections or other damage to the mammary tissue.

However, somatic cell count (SCC) and somatic cell score (SCS) of milk samples were determined for the study period. The average somatic cell count over the period of study was ~93,000 cells/mL, while the average SCS was estimated at 4.66. In the literature, SCC has been shown to impact milk composition, especially the lactose content of milk due to decreased synthesis of lactose (Lindmark-Månsson et al., 2003). However, in our study, SCC was within acceptable limits and, thus, no significant impact of SCC was found on milk composition ($p > 0.05$). The total solids content was also higher in the autumn period compared to the summer and spring periods, but there was no significant variation between the summer and spring periods. This is in line with other studies in the UK and Ireland where the total solids content decreased during the January to April and July to August periods (O'Brien (1) et al., 1999; Chen et al., 2014). As stated earlier, milk yield and compositional characteristics are affected by the stage of lactation and diet. Milk density is dependent on milk fat and SNF content; therefore, the variation in total solids content also impacts milk density, increasing in the autumn season with increasing lactose and total solids contents of milk. The impact of variation in different constituents, i.e., protein and lactose, is also shown in Table 4 and was statistically significant. Fat content showed the highest variation when compared with protein and total solids, which is in line with the general observation that fat is the most sensitive to dietary changes (Walstra et al., 2005; Heck et al., 2009). The density results were determined for major constituents, i.e., milk, total protein and lactose, not segregated for casein (and whey) and/or total solids, to avoid multicollinearity errors in the analysis.

Diet plays a significant role in the variations observed in milk composition (Lindmark-Månsson et al., 2003). During the grazing season in Ireland, cows graze outdoors, and their diet is comprised mostly of fresh grass. Fatty acids form a significant component of milk fat and variation in fatty acid composition has been mainly attributed to the supply of fatty acids through diet and rumen microbial activity (Heck et al., 2009). The main precursors of milk fat,

i.e., acetic and butyric fatty acids—derived from rumen fermentation, can be affected by diet through changes in rumen fermentation or the addition of fats for direct absorption and inclusion into milk fat (Lindmark-Månsson et al., 2003). It has also been shown that the grass consumed by cows during grazing is less mature, and this less mature grass has lower levels of polyunsaturated fatty acids (Ferlay et al., 2006). Oxidative losses in fatty acids due to the wilting and ensiling of grass have also been observed (Dewhurst et al., 2006). This reduces the amount of fatty acids from fresh grass and, thus, causes fluctuations in the fatty acid composition of milk, affecting the total fat content and milk density. Therefore, a combination of these factors and seasonal variation impacted the feed quality for grazing cows, which in turn affected the milk composition and milk density, respectively, as shown in results of this study.

Photoperiod is also known to have a significant impact on the milk production and compositional changes in milk. Photoperiod refers to the length of day or the period of daylight received by an organism (Collier et al.,2011), and the importance of photoperiod on the variations in milk composition has also been highlighted (Bertocchi et al., 2014). In dairy cattle, photoperiod influences a series of hormonal changes which affect the milk yield, composition and feed behaviour, among other parameters. Milk yield and dilution of fat and protein content have been reported to vary considerably with the increase in photoperiod or the length of the daylight period (Dahl et al., 2000; Auldist et al., 2007; Bertocchi et al., 2014). Photoperiod, as a factor, was not studied in this analysis but may contribute to the variation in milk composition and milk density and may thus require further analysis and exploration.

2.3.2 The Effect of Seasonal Variations on Milk Density, Mass Balances and Milk Payment Systems

It is evident from past research and the results of this study that seasonal variations introduce significant fluctuations in fat and protein content, increasing towards the autumn season. The variations in density values can be estimated using the model developed in this study.

Variations in different parameters introduce differences in density values and, therefore, the use of a single density conversion factor is not representative of seasonal variations, including compositional changes, climatic conditions and feed practices.

The method of density analysis is also another important factor that can affect the accuracy of measurements. The results shown in this study indicate a significant impact of the measuring technique on the raw milk density for all the samples studied (Table 2.4, Instrument, $p < 0.001$). The differences in desntiy results between different analytical methods were observed. The pycnometer method was found to have statistically signficant differences with both DMA 35 and DMA 4500 ($p < 0.001$); however, DMA 35 and DMA 4500 results were not significantly different from each other ($p > 0.05$) over the period of study. DMA 35 is used in industry for quick analysis of density (Source: interactions with industry personnel), while DMA 4500 and pycnometer methods are comparatively time-consuming. The results of the pycnometer method were higher than the other two methods, and this may be attributed to different factors, such as accuracy and tolerance limits of the measuring equipment, foreign matter in samples like sediment and particulate matter, entrapped air and bubble formation, viscosity and homogenity of samples, and temperature and temperature history of samples. In this study, the analysis was carried out in a controlled environment using strong experimental protocols to remove errors or bias.

A mass balance may be defined as the consideration of the input, output and distribution of a product/ingredient between streams in a process. For a butter manufacture process, it may be presented as follows [31]:

$$Fat\ intake = Fat\ in\ products + losses + recycled\ fat$$

The use of a density factor is paramount in terms of a mass balance calculation that can help identify different loss-making points in a process, estimate losses in the fat conversion process and, subsequently, make important process-related and investment-related decisions. Milk payment systems across different regions follow the a multiple component pricing model (A +

B - C system), where the value of protein (A) and fat (B) in kg supplied by the farmer to the processor are calculated and the cost of collection and processing (C) in cents per litre, related to the volume of milk supplied by the farmer, is deducted (Geary et al., 2010). Milk volume is converted to weight using the density conversion factor by multiplying the volume collected in litres on each farm by the density factor to obtain the weight of milk in kg.

As stated earlier, the profile of milk in Ireland has considerably changed and a single density conversion factor is not representative of the variations in milk profile due to composition and seasonality. To put this in perspective, a hypothetical example is discussed here. The annual supply of milk in Ireland for the year 2019 was 7990 million L of milk (CSO, 2020) with the seasonal profile as supplied, corresponding to a peak milk supply of 13.4% in the month of May and trough of 2.2% in January. Milk distribution for the year 2019 varied between a maximum of 1072.2 million L in May, with the lowest supply observed in December (243.7 million L) and January (175.3 million L). Thus, using season-based density factors, milk weight was determined, giving a peak of 1105.33 million kg (using a density value of 1.0309 g/cm^3) in May, while the minimum weight of milk was calculated for the December (251.38 million kg) and January (180.72 million kg) period using a density factor of 1.0314 g/cm^3. Peak values of milk weight were obtained towards the end of spring and the beginning of the summer period when the milk supply was also at its highest (May–July). When an average density factor (1.0297 g/cm^3, current industry standard) was used to calculate milk weight as compared to the density factors determined in this study, there was a total difference of 9.39 million kg/year in milk kg produced, with monthly differences as high as up to 1.3 million kg.

The model defined in this study can be a useful tool to predict the milk density value that can be used to estimate weight–volume calculations, based on different parameters such as the season, days in milk etc. Milk weight estimated using the predicted density may then be used to determine the fat and protein (in kg) available for processing. This variation in milk weight and constituents estimated from the use of new density factors will require appropriate

planning. With proper planning and capacity appropriation, the processors can therefore have better operational control in terms of product mix and capacities, as well as a better understanding of their overall mass balance, while also presenting a more accurate financial picture by having the seasonal density factors calculated appropriately.

2.4 Conclusions

The density of milk is dependent upon seasonal variations observed in milk composition throughout the year. This is evident from the results of the present study, with density varying significantly with changes in the constituents' content of the milk. Variations in the composition and ultimately density could be attributed to various factors, such as the stage of lactation, climatic conditions (including microclimatic pattern), the feeding pattern during the period of study, housing conditions in autumn and winter seasons, the genetic group, and temperature, amongst other parameters. Seasonal and annual factors for density conversion used in weight–volume relationships were determined, with an emphasis on usage of a periodic, rather than an average, conversion factor evident from the strength of linear regression models. The distribution of density and individual constituents of milk over the different seasons showed a similar trend, with higher fat and protein content observed in the autumn and winter seasons and the lowest content of these observed during summer. Monthly and season-based density factors were determined, which are relevant for milk-processing planning. Milk density is an important factor in milk processing to estimate the individual milk constituents (weight–volume calculations). The constituent contents thus calculated significantly influence the product portfolio, in conjunction with operating capacities and market demand. The use of season-based density factors, therefore, may improve upon the estimation of individual milk constituents, as shown from this study and, thus, it is vital for the processing industry to plan and control their product mix and operations more effectively. The estimation of new density factors may also enable improvements in the milk payment systems for the production and processing industry.

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Chapter 3

The effects of cow genetic group on the density of raw whole milk

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Abstract

The density of milk is dependent upon various factors including temperature, processing conditions, and animal breed. This study evaluated the effect of different cow genetic groups, Jersey, elite Holstein Friesians (EHF), and national average Holstein Friesians (NAHF) on the compositional and physicochemical properties of milk. Approximately 1040 representative (morning and evening) milk samples (~115 per month during 9 months) were collected once every two weeks. Milk composition was determined with a Bentley Dairyspec instrument. Data were analysed with a mixed linear model that included the fixed effects of sampling month, genetic group, interaction between month and genetic group and the random effects of cow to account for repeated measures on the same animal. Milk density was determined using three different analytical approaches- a portable and a standard desktop density meter and 100-cm^3 calibrated glass pycnometers. Milk density was analysed with the same mixed model as for milk composition but including the analytical method as a fixed effect. Jersey cows had the greatest mean and standard error for fat content (5.69±0.13%), followed by EHF (4.81±0.16%) and NAHF (4.30±0.15%). Milk density was significantly higher (1.0313 g/cm^3 ± 0.00026, $p<0.05$) for the milk of Jersey breed when compared to the EHF (1.0304± 0.00026 g/cm^3) and NAHF (1.0303± 0.00024 g/cm^3) genetic groups. The results from this study can be used by farmers and dairy processors alike to enhance accuracy when calculating the quantity and value of milk solids depending upon the genetic merit of the animal/herd, and may also improve milk payment systems through relating milk solids content and density.

3.1 Introduction

Composition is an important determinant of the processability and nutritive value of milk (Lindmark-Månsson, Fondén, & Pettersson, 2003) and also affects the quality of final products (Amenu & Deeth, 2007). The solids content of milk is significantly affected by the breed of the cow. Differences in milk composition have been observed among different cow breeds and also in individual cows within the same breed, partially attributed to the genetic variations between cows (Bland, Grandison, & Fagan, 2015; Gustavsson et al., 2014; McLean, Graham, Ponzoni, & McKenzie, 1984; Penasa, Tiezzi, Sturaro, Cassandro, & De Marchi, 2014; Stocco, Cipolat-Gotet, Bobbo, Cecchinato, & Bittante, 2017; Tyrisevä, Vahlsten, Ruottinen, & Ojala, 2004; Wedholm et al., 2006). Past experiments have shown that Jersey cows yield higher concentrations of fat and protein as compared to Friesian cows (Auldist, Johnston, White, Fitzsimons, & Boland, 2004; Mackle, Parr, Stakelum, Bryant, & MacMillan, 1996). These variances in fat and protein are attributed to differences in fatty acid and individual protein profiles within the milk and, according to literature, are influenced by breed (Bobe, 2008; Maurice-Van Eijndhoven, Hiemstra, & Calus, 2011; Maurice-Van Eijndhoven, Soyeurt, Dehareng, & Calus, 2013; Peterson, Kelsey, & Bauman, 2002; Soyeurt, 2007). This correlation indicates that genetic selection for milk production affects the composition of milk protein and content of milk fatty acids (DePeters, 1995; McLean et al., 1984). Similar effects of breed on milk fat and fatty acid composition have also been reported for Irish milk (Dillon, 2003; Lawless et al., 1999; O'Callaghan et al., 2016; Walsh et al., 2008)

The density of milk is an important physical characteristic that is widely used for weight-volume calculations, product mix management and profitability calculations. The density of milk is used to convert the volume of milk entering a processing environment to weight/mass of milk. The weight of individual constitutents in milk can then be determined by multiplying the mass of milk entering the processing system by the constituents' percentage. There is a direct correlation between the content of fat and milk solids and milk density (Ueda, 1999). Milk

density is correlated with the size of fat globules (Ueda, 1999) and fat globule size is dependent upon characteristics like feeding treatment, seasonal and compositional changes, breed, physiology of the animal and lactation period (Heck, Van Valenberg, Dijkstra, & Van Hooijdonk, 2009; Kljajevic et al., 2018; Mulder & Walstra, 1974; Walstra, 1969; Parmar et al., 2020). Breed and genetic characterisitics of the animal significantly affect the concentration and ratio of fatty acids in milk fat and affect the processability, i.e., its hardness or softness (MacGibbon, 1996). Processes such as homogenisation lead to smaller fat globules with a larger surface area, and also a higher density (Truong, Palmer, Bansal, & Bhandari, 2016). It has been noted through past research that the content of fatty acids such as stearic, palmitic, and oleic acids is positively correlated with the size of milk fat globules (Wiking, 2004). Milk from different breeds of cows has varying fatty acid content, which affects (Marin et al., 2018) the overall fat concentration and also affects the fat globule size. This has been attributed to the genetic merit and breed characteristics influencing the milk composition (Auldist et al., 2004; Larsen, Hymøller, Brask-Pedersen, & Weisbjerg, 2012; White, 2001).

Breed variations also impact the protein content in milk as observed from various studies (De Marchi, Bittante, Dal Zotto, Dalvit, & Cassandro, 2008; Malacarne et al., 2006; Ng-Kwai-Hang, Hayes, Moxley, & Monardes, 1986). Malacarne et al. (2006) found that protein content (3.49%) and subsequent cheese yield was markedly higher for Italian Brown cows compared to Friesian cows (3.07%). A comparison of Danish Jersey, Swedish red and Danish Holstein cows also showed that there was a significant difference in concentration of protein in milk (Gustavsson et al., 2014). Individual protein concentration and overall content of protein are affected by the genetic variations and influence processing capabilities, including coagulation properties. However, milk density is largely dependent upon factors such as milk fat content, fat globule size and ratio of solid:liquid fat (Ueda, 1999).

Peak season for milk production and supply in Ireland is the period between March – May/June when milk production increases steadily, hitting a peak in May/June, plateauing in July-August

and begins falling (off-peak) from Autumn/winter period (as grass growth begins to decline). This is evident from the data available for milk production and intake of creameries in Ireland for 2018 (CSO, 2018). While the effect of breed on milk composition has been well established through numerous research studies, the effect of genetic group on raw whole milk density has not been studied and is unavailable in the literature. The current study was designed to investigate the interaction between cow genetic group and milk density, measured through different analytical approaches, and observed for one complete season (March-November 2018), including peak and off-peak. The composition of milk samples obtained from different cow genetic groups was also measured and results were evaluated to determine the interaction between genetic group, density and equipment.

3.2 Materials and Methods

3.2.1 Experimental Design and Sampling

The research was carried out over a period of 9 months from March 2018 to November 2018, within one season. Season was defined as spring (March, April and May), summer (June, July, August) and autumn (September, October and November). Raw whole milk samples from the combined evening and morning milking were obtained from a Teagasc Research farm, Kilworth, Co. Cork (Latitude 50°07'N, Longitude 08°16'W). The genetic groups and breeds assessed in this study included Jersey, and two genetic groups of Holstein-Friesian breed, i.e., Elite Holstein-Friesian (EHF) and National average genetic merit Holstein Friesian (NAHF) cows. Elite and National Average Holstein-Friesian were chosen on the basis of economic breeding index (EBI). EBI is a profit index aimed at providing information to farmers regarding selection of cows for breeding herd replacements (Berry et al., 2005). Elite cows had a higher EBI compared to National Average Holstein-Friesian cows. All the cows (n=54) included in the study were milked twice a day. The cows were segregated into three groups on the basis of feed (3 different feed patterns explained below) given to each genetic group and 6 cows were selected for each feed pattern (6*3 = 18 cows per genetic group). Indicative feeding

treatments were as follows:

(a) Control system: Stocking rate (SR) of 2.75 cows/ha, 250 kg N/ha. Concentrate (3 kg) was offered per cow per day immediately post calving to supplement pasture availability in the spring (12 weeks). Pasture was allocated in accordance with best management practice in mid-season (approx. 4.5 cm post grazing residual; 18 weeks). A grass only diet was offered in the autumn period (12 weeks). Post-grazing residual was managed at 4.5 cm in spring and autumn.

(b) High concentrate system: Seven kg concentrate was offered per cow per day immediately post calving to supplement pasture availability in the spring (12-weeks). Four kg/cow/day supplementation was offered in the autumn period (12 weeks). Pasture allocation, stocking rate and post-grazing residual was similar to control.

(c) Lower grass Residual: Concentrate (3 kg) was offered per cow per day immediately post calving to supplement pasture availability in the spring (12 weeks). A grass only diet was offered in the autumn period (12 weeks). Post-grazing residual was 3.5-4 cm in spring and autumn. Pasture allocation and stocking rate was similar to control.

A total of 1040 samples of approx. 150 ml each were collected during this period and each of the samples were tested for compositional profile and whole milk density. The following parameters were tested during the process: fat, protein, total solids content, while raw milk density was evaluated using three different analytical approaches. The milk composition was determined using a Dairyspec FT manual model (Bentley Dairy Systems, Chaska, Minnesota, USA) while the milk density was determined using three different analytical approaches – DMA 35 portable density meter, (Anton Paar, Graz, Austria) ,DMA 4500 desktop density meter, (Anton Paar, Graz, Austria) and 100-cm^3 calibrated glass pycnometers (Blaubrand, Wertheim, Germany). Sampling requirements were in accordance with ISO 707:2008 (Milk and Milk Products: Guidance on sampling) (ISO, 2008).

Evening samples were collected and stored under refrigerated conditions at 5°C for 18 h to

prevent microbial growth and enzymatic activities. Morning samples were collected the next day and mixed with the evening samples to prepare a representative sample. The samples were then tested for composition and density immediately after morning milk recording to prevent alteration to composition or spoilage. Therefore, the analysis was always completed within 24 h of the earliest milk collection.

3.2.2 Methodology

The raw milk density was determined using three different methods i.e. DMA35 portable density meter, a standard desktop density meter DMA4500 and the results from these two methods were then compared against results obtained from measurements using 100-cm3 glass calibrated pycnometers. The samples collected were properly agitated before analysis to ensure thorough mixing of constituents and to remove any errors due to settling. Before analysis, the density meters were also calibrated using distilled water. Once calibrated, one sample at a time was analysed from start to finish on all three analytical methods, while maintaining sample temperature at ~20°C. After completing density measurement for all samples, the samples were then analysed on the Dairyspec infrared manual FT model for milk profile.

DMA4500 and DMA35

DMA35 is used as a method for density measurement across industry due to rapid results, easier handling and manoeuvrability. It works on the principle of hollow oscillating U-tube technology. The principle of operation in the two different pieces of equipment (DMA35 and DMA4500) is based on the principle of changing frequency of a hydrogen filled hollow oscillator when filled with different liquids. The mass and density of the liquid changes the natural frequency of the oscillator due to overall change in mass of the oscillator when a liquid is added into the tube. The DMA4500 is capable of evaluating density with precision of 0.00005 g/cm³ and 0.02 °C with a working temperature range of 0-100°C and requires only 1-2 ml of sample, requires no viscosity-related standards and eliminates temperature-

related fluctuations. The DMA4500 can be calibrated at one temperature and all samples for density can be measured at the set temperature. The equipment is also capable of automated cleansing and introduces immediate temperature equilibrium. The measured density of water at 20°C using DMA35 was 0.9974 g/cm3 and, for DMA4500, it was noted to be 0.99826 g/cm3, close to the theoretical value of 0.99820 g/cm3 for water at 20°C.

AOAC standard method using glass pycnometers

The third method used to measure density was the AOAC 925.22 official method for determining the specific gravity of a liquid using pycnometry. Calibrated 100-cm3 glass density pycnometers (Make Blaubrand BR43338, Wertheim, Germany) were used to determine the density of the milk samples. The densities of liquids attained from pycnometry method are compared against water. In this method, firstly, the empty glass bottle was weighed and noted. The glass bottle was then filled with distilled water and wiped dry to remove any water on the outer surface of the bottle. This filled mass was then measured and noted, after which the bottle was emptied completely. The bottle was then filled with liquid (milk) and the outer surface was wiped dry and weighed again. Excess liquid or water from the bottle was removed from the bottle through a capillary action of the bottle lid. The density of the liquid against that of water was measured using the formula

$$Density = \frac{WS - WE}{WW - WE}$$

Where WS is the weight of a sample-filled bottle, WE is the weight of an empty bottle and WW is the weight of a water-filled bottle.

The sample was firstly tested on DMA35 with approx. 1-2 ml of sample drawn directly from the sample container and the density was noted from the display screen of the equipment. Secondly, two ml syringes were used to inject the samples into the oscillating tubes of the DMA4500 equipment, preventing the flow of air into the sample. The desktop model DMA4500 was adjusted to note the density of milk samples at 20 °C for all samples using the temperature settings available on the panel. The milk density of samples was then noted

using the glass bottles from the standard AOAC 905.22 method and formula. The same procedure was applied to measure the density of all the samples collected during every run (18 samples for each genetic group each month). The glass pycnometer method requires a minimum of 100-cm3 sample for density measurement and thus needs to account for insufficient milk produced and collected at the farm, spillage and/or wastage. The number of sample points for the pycnometry method in this study are therefore less (approx. 740), compared to the other two methods (approx. 1040 for the other two approaches).

After analysis of density was completed, the milk compositional profile, i.e., milk fat, protein and total solids content, was assessed by infrared spectrophotometry. An approx. volume of 30 ml sample was required to be tested on the Dairyspec infrared manual FT model (Make-Bentley Instruments Inc.) calibrated for raw whole milk compositional analysis. The Dairyspec machine is based on FTIR (Fourier transform infra-red spectroscopy) principle.

3.2.2 Statistical Analysis

All dependent variables were analysed using the statistical package SAS version 9.4 (SAS Institute Inc., Cary, NC, USA). Descriptive statistics were obtained using the MEANS procedure. Least squares and standard errors for factors affecting milk composition and density were obtained using the MIXED procedure. The model for milk composition traits included the fixed effects of genetic group, feeding treatment, parity, days in milk with linear and quadratic effect as covariates, and random effects of cow and residual error. Milk density was analysed with the same mixed model as for milk composition with the addition of analytical method (DMA4500, DMA35 and glass pycnometers) as fixed effect. Least squares means were used for multiple mean comparisons using the Fisher's least significant difference test as implement in the option LSMEANS and significant differences were defined at $p<0.05$. Variance components for cow (σ^2cow) and residual (σ^2e) were used to estimate repeatability of the trait, calculated as

rep = σ^2cow/σ^2total where σ^2total = σ^2cow + σ^2e.

3.3. Results

Descriptive statistics for milk composition for all samples collected during the period of study was determined. The average fat, protein, lactose, total solids, casein and weighted average density values were 4.73±1.30%, 3.85±0.56%, 4.70±0.30%, 14.03±2.21%, 2.88±0.58% and 1.0308±0.002 g/cm3, respectively. Coefficient of variation was also determined for each of the constituents analysed and are shown in Table 3.1.

Table 3.1 Mean, standard deviation (SD), Coefficient of Variation (CV) and minimum and maximum values of milk composition (n = 1044) and density - samples (n=2836) collected from three cow genetic groups (averaged results).

Trait	Mean	SD	CV	Minimum	Maximum
Fat (%)	4.73	1.30	27	2.14	14.86
Protein (%)	3.85	0.56	16	1.76	5.95
Total solids (%)	14.03	2.21	19	8.57	22.48
Casein (%)	2.88	0.58	20	0.61	5.00
Lactose (%)	4.70	0.30	6	2.45	5.61
Density g/cm^3	1.0308	0.002	0.20	1.0153	1.0378

Table 3.2 presents the least mean square values along with the standard errors for the constituents and density based on the genetic groups analysed. Fat content was estimated at 4.81±0.16% for Elite HF milk, while fat content was highest and significantly different for Jersey cows at 5.69±0.13%, compared to Elite and NA Holstein-Friesian cows. Fat content for Jersey milk was approx. 30% higher compared to National average Holstein-Friesian cow milk. Overall milk density least mean squared value for Jersey milk was estimated to be significantly different ($p<0.05$, 1.0313±0.00021g/cm3) from Elite and NA cows (1.0304±0.00026 g/cm3 and 1.0303±0.00024 g/cm3, a difference of 0.001 g/cm3 between NA Holstein-Friesian and Jersey cow milk density). The numerical difference between

density for Jersey cow milk and Holstein-Friesian cow milk was observed to be small but statistically significant ($p<0.05$).

Table 3.2 Least squares means (LSMean), number of samples (=n) and standard errors (SE) of milk composition traits (Fat, protein, total solids, lactose and casein, %) and density (ρ, g/cm^3) from three cow genetic groups (Elite, National Average Holstein-Friesian and Jersey

cows)

Trait	Genetic group[1]	n	LSMean	SE
Fat, %	Elite HF	357	4.81[b]	0.165
	Jersey	341	5.69[c]	0.131
	NA HF	346	4.30[a]	0.154
Protein, %	Elite HF	357	3.82[a]	0.063
	Jersey	341	4.18[b]	0.050
	NA HF	346	3.73[a]	0.058
Total solids, %	Elite HF	357	14.11[b]	0.242
	Jersey	341	15.36[c]	0.185
	NA HF	346	13.34[a]	0.227
Lactose, %	Elite HF	357	4.63[a]	0.031
	Jersey	341	4.67[a]	0.037
	NA HF	346	4.61[a]	0.026
Casein, %	Elite HF	357	2.89[a]	0.065
	Jersey	341	3.15[b]	0.052
	NA HF	346	2.82[a]	0.060
Density	Elite HF	330	1.0304[a]	0.00026
(g/cm^3)	Jersey	301	1.0313[b]	0.00021
	NA HF	314	1.0303[a]	0.00024

[1] Elite HF = Elite Holstein-Friesian, NA HF = national average Holstein-Friesian.

[a,b,c] LSMeans with different superscript within each milk component are significantly different ($p<0.05$).

Table 3.3 presents the density values for each of the genetic groups estimated with all three measurement techniques. The maximum (and significant, $p<0.05$) variation in density was

seen for pycnometer method for all genetic groups, while density values obtained from DMA35 and DMA4500 methods were not significantly different ($p>0.05$). The number of milk samples used for density measurement was different for the pycnometer method compared to DMA35 and DMA4500 (same number of samples used). This is attributed to the fact that the pycnometer method requires a minimum of 100-cm3 sample to estimate density, which was not feasible due to limited milk production and thus, sampling. Table 3.3 presents the analysis for density when the same number of samples (n) was used for estimation of density for all three measurement techniques.

Table 3.3 Least squares means (LSMean) and standard errors (SE) of genetic group-wise (Elite, National Average Holstein-Friesian and Jersey cows) milk density (ρ, g/cm^3) determined by three analytical methods (Pycnometer, DMA35 and DMA4500).

Genetic group	Method	LSMean	SE
Elite Holstein-Friesian	Pycnometer	1.0319^a	0.00024
	DMA35	1.0296^b	0.00024
	DMA4500	1.0296^b	0.00024
Jersey	Pycnometer	1.0327^a	0.00021
	DMA35	1.0308^b	0.00021
	DMA4500	1.0308^b	0.00021
National average Holstein-Friesian	Pycnometer	1.0318^a	0.00023
	DMA35	1.0295^b	0.00023
	DMA4500	1.0296^b	0.00023

^{a,b} LSMeans with different superscript within each genetic group are significantly different ($p<0.05$).
^{A,B,C,D,E,F} LSMeans with different superscript within each genetic group are significantly different (for the analytical method used, $p<0.05$)

The pycnometer method showed the highest estimate of density at 1.0321 g/cm3 and pycnometer density results were significantly higher ($p<0.05$) from those of the other two

methods (DMA 35 – 1.0300 g/cm3 and DMA4500 – 1.0300 g/cm3, no significant difference between DMA 35 and DMA 4500 ($p>0.05$)) The results estimated in Table 3.4 were observed from the same samples (n=744) after removing any missing data from all measurement techniques.

Table 3.4 Least squares means (LSMean), number of samples (N) and standard errors (SE) of milk density determined by three analytical methods (Pycnometer, DMA35 and DMA4500) (to assess the effect of each measurement technique)

Method[1]	N	LSMean	SE
Pycnometer	744	1.0321[b]	0.0001
DMA35	744	1.0300[a]	0.0001
DMA4500	744	1.0300[a]	0.0001

[a,b] LSMeans with different superscript are significantly different ($P<0.05$).
[1]Analytical methods used for measurement of milk density, discussed in detail in Materials and Methods

Table 3.5 shows the Pearson's correlation coefficient determined to compare the three methods. Pycnometer method was established as the gold standard and the other two methods compared against it. The correlation coefficient for DMA35 and DMA4500 were not significantly different from each other (0.82 and 0.83).

Table 3.5 Pearson Correlation coefficients determined to compare the three measurement techniques -Pycnometer method as a gold standard; DMA35 and DMA4500 compared with

the standard

Pearson Correlation Coefficients			
	Pycnometer	DMA35	DMA4500
Pycnometer	1.00	0.82	0.83
Pycnometer		<.0001	<.0001
DMA35	0.82	1.00	0.92
DMA35	<.0001		<.0001
DMA4500	0.83	0.92	1.00
DMA4500	<.0001	<.0001	

Lastly, in Table 3.6, covariance parameters were determined to test the repeatability of effect of cow on density variation over the sampling period. Random cow effects on density accounted for 20.45% of between-cow effects and 79.54% for within-cow effects, which could be attributed to genetic merit and inter-genetic group differences.

Table 3.6 Estimates of variance components and repeatability of milk density, ρ for three cow genetic groups (Elite, National Average Holstein-Friesian and Jersey cows)

Trait	Between cows	Within cow	Total	Repeatability (%)
Fat	0.24	0.69	0.93	26.22
Protein	0.04	0.08	0.12	30.26
Density	6.779E-7	2.636E-6	3.31E-6	20.45

3.4 Discussion

3.4.1 Effect of genetic group on raw milk density

The impact of breed on different characteristics of milk such as composition profile, fatty acid profile, processability etc. has been well established in the literature (Bland et al., 2015; Kelsey, Corl, Collier, & Bauman, 2003; Lock & Bauman, 2004; Malossini, Bovolenta, Piras, Dalla Rosa, & Ventura, 1996; Penasa et al., 2014; Stocco et al., 2017; TX Yang, 2013; Tyrisevä et al., 2004). However, the impact of breed and the use of different types of analytical approaches to measure raw milk density have not been widely addressed, to the best of our knowledge. The effect of genetic group on milk composition, e.g., fat and protein levels, fatty acid composition and protein polymorphisms has been discussed widely (De Marchi et al., 2008; Heck et al., 2009; Kljajevic et al., 2018; Malacarne et al., 2006; Ng-Kwai-Hang et al., 1986). Because of genetic background and traits, milk samples collected from different cattle genetic groups have diverse compositional profile. A similar trend was observed in the results of this study, where fat, protein and total solids content varied across different genetic groups throughout the season. In this study, the milk composition (fat and protein contents) obtained from three different genetic groups were significantly different ($p<0.05$) under the same feeding conditions.

Sample-related factors include temperature history of the sample, inclusion of air and concentration of fat and solids-non-fat. Other factors affecting milk physical characteristics and composition may be genetic merit of the cow, feeding treatment, lactation cycle and period and inter- and intra-herd variations (Gustavsson et al., 2014; McLean et al., 1984; Wedholm et al., 2006). Sample-related factors such as temperature and temperature history of the sample have been described (Hlaváč & Božiková, 2011; Richmond, Davis, & Macdonald, 1953; Short, 1955). The results for milk density from this study show the highest density value for Jersey milk (1.0313 g/cm^3), while it was measured as 1.0304 and 1.0303 g/cm^3 for milk of elite and national average Holstein-Friesian cows. This may be attributed to genetic merit of the animal and variations in milk fat concentration due to genetic group effects.

Genetic merit and its impact on milk composition has been extensively studied in literature.

Milk fat is mainly present in globule form as an oil-in-water emulsion (MacGibbon, 1996) and fat is comprised of approx. 400 different types of fatty acids, out of which approx. 70% are saturated fatty acids and the remaining 30% are unsaturated (Lindmark Månsson, 2008). The fatty acid profile of milk is dependent upon different factors: animal breed, stage of lactation, feed, and microbial activity in the rumen of the animal (Lindmark Månsson, 2008). The main pre-cursors of milk fat, i.e., acetic and butyric fatty acids - derived from rumen fermentation, can be affected by diet through changes in rumen fermentation, directly dependent upon the genetic variations in cows (Lindmark Månsson, 2008). The impact of genetic variations and background significantly affects the fatty acid composition in individual breeds, for example, a higher content of short chain fatty acids and to some extent, medium chain fatty acids were observed in Danish Holstein cows compared to the Danish Jersey breed (Poulsen et al., 2012) It has been noted through past research that the content of fatty acid such as stearic, palmitic, and oleic acid is positively correlated to the size of milk fat globule (Wiking, 2004). Mulder & Walstra (1974) suggested that the majority (94%) of fat globules are sized between 2-8 µm and fat globule size is dependent upon characteristics like breed, physiology of the animal and lactation period. Milk fat globule size directly impacts the milk density and the size of globules increase with an increase in fat content of milk, due to limited membrane production (Wiking, 2004). Therefore, it is clear that the changes in milk fat globule size and subsequent milk density are directly correlated to the genetic merit of the animal, as shown from the results of this study (Table 3.2). This outcome was also corroborated by other studies available in literature (Larsen et al., 2012; White, 2001) and is independent of dietary effects on composition and only due to genetic traits and breed differences (Beaulieu & Palmquist, 1995). Thus, the size of milk fat globules critically effects the stability, technological and physical properties of milk, such as density, and is reliant on characteristics like breed and physiology of cows (Heck et al., 2009; Kljajevic et al., 2018). Disintegration of fat globules during proccessing also impacts the size of milk globule and, therefore, affects the milk density.

A related assessment for effect of breed on protein profile and individual protein content was

conducted by Gustavsson et al. (2014). The results from their study showed a significant impact of breed on the relative overall concentrations of proteins (as shown in the results of this study, milk of Jersey cows have highest protein content, compared to Elite and NA strains of Holstein Friesian). Protein content, as well as its composition, is known to impact the processability of milk (Ketto et al., 2017; Malossini et al., 1996; Poulsen, Glantz, Rosengaard, Paulsson, & Larsen, 2017; Tyrisevä et al., 2004; Wedholm et al., 2006). The impact of seasonal and compositional variation on milk density has been assessed in a study by the same authors (Parmar et al., 2020), which showed that variation in milk constituents over different seasons significantly impacted milk density ($p<0.05$).

Other studies in the literature have observed an inverse relationship between milk fat content and milk density values (Czerniewicz et al., 2006). Milk fat content along with solid-non-fat content including protein content had a significant impact on the density of milk. Extrinsic factors such as days in milk, season, feeding treatment, and measurement technique also have statistically significant impacts on milk density.

3.4.2 Effect of analytical technique on the measurement of raw milk density

The results from this study indicate a significant impact of the measuring technique on the raw milk density for all the samples studied. The results were significantly affected by measurement method ($p<0.05$) with 100-cm^3 glass pycnometers recording the highest values of density for all genetic groups. The results of density measured from 100-cm3 glass pycnometers, as per the AOAC method, revealed a higher value of density (1.0321 g/cm^3) as compared to the results of DMA 35 and DMA 4500 (1.0300 and 1.0300 g/cm^3 respectively), with all samples undergoing the same treatment (storage conditions). This may be attributed to the precision and tolerance limits of the measurement technique, along with variations in density introduced due to temperature history of the samples and Recknagel's phenomenon. Recknagel's phenomenon refers to the density of sample measured immediately after milking being lower compared to milk stored for longer periods of time especially at lower temperatures. This is observed due to the increase in hydration of protein

at lower temperatures instead of the escape of air bubbles (IASRI, 2012). Another critical factor affecting density measurement using different equipment is the temperature history of the samples. The samples collected in the evening were stored in a refrigerator overnight at 5°C and were mixed with freshly collected samples from the morning milking. This affected the temperature of the representative sample subsequently used for density measurement. The temperature of measurement for the DMA 4500 was standardised at 20 °C for all samples while temperature variations could have been introduced into density measurement when assessed on the DMA 35 and 100-cm3 glass bottles. This may be attributed to the temperature sensitivity of DMA 35 and no temperature control was used during the use of pycnometers for density measurement. Past research has highlighted the need to determine the controlled temperature history necessary for high precision and accurate density measurement (Hilker & Caldwell, 1961; Sharp & Hart, 1936; Vanstone, 1960). Other factors affecting the density measurement using bottles may include the possible presence of foreign particles in sample, entrapped air, bubble formation, temperature influence, and/or viscosity-related errors.

3.5. Conclusion

Genetic traits and merit of the animal significantly impacts on whole milk density, in conjunction with other factors like composition, feed treatment, seasonality, processing environment and temperature. To the best of our knowledge, this is the first of its kind of research, especially for the Irish dairy sector, wherein the breed of the animal has been studied to analyse its impact on milk density, which is an integral parameter in weight-volume calculations in a dairy processing environment. Milk density factors established for different genetic groups in this study may be helpful in estimating weight-volume relationships based on milk supplied from different herds (genetic groups). This will also help in calculating the weight of milk constituents received for processing. The relationship between genetic group and density, thus, established, may enable the inclusion of breed as a support parameter in decision making for milk payments.

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Chapter 4

Effect of temperature on raw whole milk density and its potential impact on milk payment in the dairy industry

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Declaration: Milk collection, analysis and determination of milk composition and density were conducted by Puneet Parmar at Teagasc, Moorepark Food Research Centre. Dr. Laurence Shalloo gave guidance regarding the experimental design and analysis. All experimental data were analysed and the chapter written by Puneet Parmar, with corrections and comments from Prof. Alan L. Kelly, and Dr. Shane V. Crowley from University College Cork, Dr. Laurence Shalloo, Dr. Frank Buckley, Dr. John T. Tobin and Dr. Eoin Murphy from Teagasc Moorepark and Dr. Nicolas Lopez-Villalobos from Massey University.

Abstract

The objective of this study was to determine the effect of temperature on whole milk density measured at four different temperatures :5, 10, 15, and 20°C. A total of 93 samples were collected from morning milking of 32 Holstein Friesian dairy cows, of national average genetic merit, once every two weeks over a period of 6 weeks and were assessed by Fourier Transform Infrared Spectroscopy for milk composition analysis. Density of the milk was evaluated using two different analytical methods: a portable density meter DMA35 and a standard desktop model DMA4500M (Anton Paar GmbH, UK). Milk density was analysed with a linear mixed model with the fixed effects of sampling period, temperature and analysis method; triple interaction of sampling period x analysis method x temperature, and the random effect of cow to account for repeated measures. The effect of temperature on milk density(ρ) was also evaluated including temperature (*t*) as covariate with linear and quadratic effects within each analytic method. The regression equation describing the curvature and density-temperature relationship for the DMA35 instrument was $\rho = 1.0338 - 0.00017T - 0.0000122T^2$ while it was $\rho = 1.0334 + 0.000057T - 0.00001T^2$ for DMA4500 instrument. The mean density determined with DMA4500 at 5 °C was 1.0334 g/cm^3, with corresponding figures of 1.0330, 1.0320 and 1.0305 g/cm^3 at 10, 15 and 20 °C, respectively. The milk density values obtained in this study at specific temperatures will help to address any bias in weight-volume calculations and thus may also improve the financial and operational control for the dairy processors in Ireland and internationally.

4.1 Introduction

The dairy processing sector contributes significantly to the economy of many countries such as Ireland, Netherlands, New Zealand, Denmark and the USA. For example, in 2017, Irish dairy's economic contribution accounted for approx. one-third, or € 4.02 billion, of the total €12.6 billion exports from the food and drink sector, rising by approx. 19% compared to 2016 (Cornall, 2018). In view of this, milk composition is considered as an important parameter for process-ability and quality of final products (Amenu and Deeth, 2007), as well as the yield of products produced from the milk. The composition of raw whole milk procured by dairy processing companies plays a vital role in the profitability of the business and is a key determinant of the value of milk (Lindmark-Månsson, Fondén and Pettersson, 2003). A significant amount of research has been conducted globally to study the physico-chemical properties and variations in milk composition during the course of the year. Variations in composition of milk are dependent on a number of factors, such as season, lactation stage, health of cow, feeding regime and cow genetics (Heck et al., 2009; Kljajevic et al., 2018). As a result, the composition of milk and its associated functional properties can vary significantly throughout the year (Chen, Lewis & Grandison, 2014). This is particularly true where pasture-based feeding is practiced, i.e., in New Zealand, Australia and Ireland. The associated changes in feeding pattern affect the yield and composition of milk throughout the year (Grimley, Grandison & Lewis, 2009).

Milk density is a function of inherent and external factors. Density is impacted by external factors such as processing, agitation, homogenization, composition at a given temperature and pressure (Walstra and Jenness, 1984). Density is particularly important in milk processing, where milk intake is typically measured on a volume basis (L); however, process and final product yields are typically calculated on a weight basis (kg). Thus, density is calculated as mass = volume x density. Changes in density are closely related to solids-not-fat content, fat

content and temperature of milk (Short, 1955). Past research suggests that density of milk fluctuates between 1.025 to 1.035 g/cm^3 (Scott et al., 1998). Milk density is also dependent upon external factors like processing, agitation, and homogenization of milk, along with inherent factors such as animal genotype, stage of lactation, and seasonal variation (Heck et al., 2009; Kljajevic et al., 2018; Parmar et al., 2020; Rutz, Whitnah & Baetz, 1955; Short, 1956). The effect of temperature on milk density has also been studied, and it has been previously shown that milk density decreased as the temperature is increased up to 40 °C (Short, 1955,1956). Past research also found that pasteurization affected the milk density negligibly, but that sterilization of milk at high temperature 95°C decreased the density for both whole and skim milk (Short, 1956). Thermal treatment of milk affects the size of fat globules by impacting the crystallization of fat, which directly impacts on density (Huppertz and Kelly, 2006; Mulder and Walstra, 1974; Van Boekel and Walstra, 1995).

To the best of our knowledge, milk density-temperature relationships have not been analysed for the dairy industry recently, and the past research on this relationship has been completed many years ago (Short, 1955;1956). The compositional profile of milk has altered considerably since then, due to improvements in animal genetics, health and physiology, management practices, feeding regimes and other factors, thus requiring the current density factors to be evaluated. This study also enables to establish a link between milk density, variations in milk density due to temperature and its usage and impact on milk payment systems

The current study was designed to assess the impact of temperature on whole milk density for the milk production and processing sector. The temperatures identified to conduct density trials are important during milk processing within a dairy plant and, therefore, can be used to establish weight-volume relationships and to estimate the variations in yield of products and profitability of the milk conversion processes. It is also worthy to note that, in practice, most density measurements are completed at 20°C at the dairy plant sites, while milk is collected

from farms at 4-5 °C. This difference in temperature (between collection and processing) leads to variance in milk density estimation. This study, therefore, aimed to establish density factors at different temperatures, i.e., 5, 10, 15 and 20 °C, for use in weight-volume calculations.

4.2 Materials and Methods

4.2.1 Milk Samples

Data was available from the 'Next Generation Herd' project at the Teagasc research farm in Kilworth (Co. Cork, Ireland) in 2018. A detailed description of this study has been published previously [19]. The farm comprised of an effective area of 93 ha, with a capacity of 200-250 spring-calving cows. For this study, 32 Holstein Friesian individual cows of national average genetic merit were selected for sampling and were chosen on the basis of economic breeding index (EBI), which is a profit index aimed at providing helpful information to farmers regarding selection of cows for breeding herd replacements (Berry and Amer, 2005). Raw milk samples (100 ml each) were collected from Teagasc Kilworth Research Farm, Kilworth, Co. Cork, Ireland (Latitude 50°07'N, Longitude 08°16'W).

Morning samples were collected from a group of cows once every 2 weeks over a 6-week period. A total of 93 samples were collected for a period of approx. 6 weeks between July and August 2018 to assess the variations in density associated with temperature. The composition and physical properties were measured every two weeks. The following parameters were measured: fat, protein, total solids, temperature and milk density. Approx. 100 ml samples were collected from each of the selected cows milked using a 20-unit herringbone parlour (Make- DairyMaster, Cincinnati, OH, USA) with daily electronic milk weighing and sampling. Milk samples collected were stored overnight at 4-5°C to prevent spoilage and bacterial growth before each analysis.

4.2.2 Sample Analysis

The compositional characteristics of whole milk samples, i.e., fat, protein and total solids were

determined at 5 °C by Fourier Transform Infrared Spectroscopy using a Dairyspec FT manual model (Bentley systems, Chaska, MN, USA) to determine the variation in fat, protein and total solids content over the monitored period. However, there were no significant differences noted in the constituents for the three sampling periods. The temperature of samples was adjusted using a cooling circulator waterbath, (CC K-6, Make- Huber Kältemaschinenbau AG, Offenburg, Germany). The sampling chamber (20 ml) was heated to the required temperature (first measurement done at 5°C and then heated up to the temperature, 10, 15 and 20 °C) by circulating water though the surrounding jacket for 1 min using a Huber water bath CC-K6 (cooling circulator) (Make- Huber Kältemaschinenbau, Offenburg, Germany). A screw nut arrangement at the bottom of the sampling chamber allowed for drainage of each sample and the chamber was cleaned after every sample.

Density of the samples was determined using 2 different methods: DMA35 portable density meter (Make- Anton Paar, Hertfordshire, UK) and DMA4500 desktop density meter, (Make- Anton Paar, Hertfordshire, UK). The DMA35 has a working temperature range of 0 to 40 °C and density tolerance limit of 0.001 g/cm^3. Current industry practice includes the use of a portable hand-held density meter (DMA35) (for quicker results, source: interaction with industry personnel). The DMA4500 has a temperature range of 0 to 100°C and density tolerance limits of 0.00005 g/cm^3. The DMA4500 is capable of automated cleansing, introduces immediate temperature equilibrium and there are no temperature-related aging effects on the measuring cell. All measurements were made at 5, 10, 15 and 20 °C after storing samples at 4-5 °C for 24 hours. Both instruments were calibrated using distilled water to ensure that the measured density of water was within the permitted range (1.0000 at 4°C – 0.9980 g/cm^3 at 20°C) (USGS,2018).

For DMA35, the calibrated density value for water was 0.9971 g/cm^3 and for DMA4500, it was noted to be 0.9988 g/cm^3. For the first batch of samples tested at 5°C, the samples were

maintained at the treatment temperature in the water bath and density was measured using the 2 measurement approaches. After measuring density at 5°C, the samples were heated to 10°C by adjusting the temperature of water bath (an equilibration time of approx. 90 sec) and density was again measured using DMA35 and DMA4500. The sample remained in the water bath chamber for the duration of density measurement.

New milk samples were collected once every 2 weeks and the process was repeated for the other temperature combinations, i.e., 5 and 15 °C and 5 and 20 °C, giving a set of measurements for every batch. The analysis provided a set of three readings for density at 5 °C and one set of readings for each of the temperatures monitored, i.e., 10, 15 and 20 °C across different samples. The three sets of readings obtained at 5 °C were then statistically analysed.

4.2.3 Statistical Analysis

The data was analysed using SAS 9.4 (SAS Institute Inc., Cary, NC, USA) to determine the effect of temperature on the density of the milk. Least squares and standard errors for factors affecting density were obtained using the MIXED procedure. The mixed linear model included the fixed effect of sampling period, temperature, analytical method and the triple interaction of sampling period x analysis method x temperature, as well as the random effect of cow to account for repeated measures on the same cow. Least squares means were obtained for the fixed effects and used for multiple mean comparisons using the Fisher's least significant difference test as implemented in the option LSMEANS. Significant differences were defined at $P<0.05$. Variance components for cow (σ^2_{cow}) and residual (σ^2_e) were used to estimate repeatability of the trait calculated as rep = $\sigma^2_{cow}/\sigma^2_{total}$ where $\sigma^2_{total} = \sigma^2_{cow} + \sigma^2_e$. The effect of temperature on milk density was also determined considering temperature as a covariate in the model described above with linear and quadratic effects within each analytic method. From the model estimates of the regression coefficients, standard errors and P-

values were obtained to model milk density on temperature.

4.3 Results

The milk composition was analysed to determine the mean fat, protein and total solids content. Sampling periods 1, 2 and 3 were defined as the period of sampling milk, i.e., every 2 weeks during July-August 2018. The samples were analysed separately for three temperature combinations, i.e., 5-10 °C, 5-15 °C and 5-20 °C. The changes in density value increased as the temperature increased from 5 to 10°C and beyond. Table 4.1 depicts the least squares means of milk density for combinations between sampling period, analytical method and temperature. Least squares mean of milk density for the DMA35 instrument at 5 °C was 1.0330 g/cm^3, at 10 °C was 1.0322 g/cm^3, at 15 °C was 1.0311 g/cm^3, and at 20 °C was 1.0296 g/cm^3.

Table 4.1 Least squares means (LSMean) and standard errors of mean (SEM) of milk density (n=93) determined by 2 analytical methods (DMA 35 and DMA 4500), adjusted for interactions between different sampling periods and temperatures of milk samples

Analytical method	Sampling point (2018)	Temperature	Density LSMeans (g/cm^3)	SEM
DMA35	2nd Aug	5	1.0330[a]	0.0001
		10	1.0322[b]	0.0001
	15th Aug	5	1.0331[a]	0.0002
		15	1.0311[b]	0.0002
	30th Aug	5	1.0328[a]	0.0002
		20	1.0296[b]	0.0002
DMA4500	2nd Aug	5	1.0339[a]	0.0001
		10	1.0334[b]	0.0001
	15th Aug	5	1.0335[a]	0.0002
		15	1.0319[b]	0.0002
	30th Aug	5	1.0330[a]	0.0002
		20	1.0303[b]	0.0002

[a,b] LSMeans within each date for each instrument with different superscripts are significant different ($P<0.05$).

The least squares mean milk density values were comparatively higher for DMA4500 for similar test conditions. Table 4.2 shows the least squares means of milk density at the different temperatures measured with the DMA4500 instrument; density values were 1.0334 g/cm^3 at 5 °C and 1.0305 g/cm^3 at 20 °C

Table 4.2 Least squares means (LSMean) and standard errors of mean(SEM) for milk density (ρ, g/cm^3) for different temperature (5,10, 15 and 20°C) corrected for effect of sampling period, analytical approach and random cow effects

Effect		LSMean (g/cm^3)	SEM
Temperature	5	1.0334^a	0.0001
	10	1.0330^b	0.0002
	15	1.0320^c	0.0002
	20	1.0305^d	0.0002

^{a,b,c,d} LSMeans within each effect with different superscripts are significant different (P<0.05).

Density values for temperature shown here are for the DMA4500 instrument.

Table 4.3 shows the estimates of regression coefficients of milk density on temperature with linear (β_1) and quadratic (β_2) effects in each of the analytic method. The two equations corresponding to each of method are shown below.

Table 4.3 Estimates of regression coefficients ± standard error (and p-value) of milk density (ρ, g/cm^3) on temperature for different analytical methods (DMA35 and DMA4500).

Regression coefficient	Analytical method	
	DMA35	DMA4500
	Estimate±SE	Estimate±SE
β_0	1.03380±0.00033	1.03340±0.00033
β_1	-0.0001726±0.000056 (p = 0.0024)	0.000057±0.000056 (p = 0.3104)
β_2	-1.22E-06±2.386E-6 (p = 0.6102)	-0.00001±2.386E-6 (p < .0001)

The quadratic effect of temperature on milk density was significant (P<0.0001) only when estimated in the DMA4500 instrument, indicating curvature in the density-temperature relationship (Fig. 4.1). The figure also highlights the scale of variation in density values for the 2 analytical devices measured at different temperatures.

For DMA35, the equation was $\rho = 1.0338-0.0001726T-0.0000122T^2$ Equation (1)

And for DMA4500, the equation was $\rho = 1.0334+0.000057T-0.00001T^2$ Equation (2)

where, ρ = milk density in g/cm^3 and T = temperature in °C

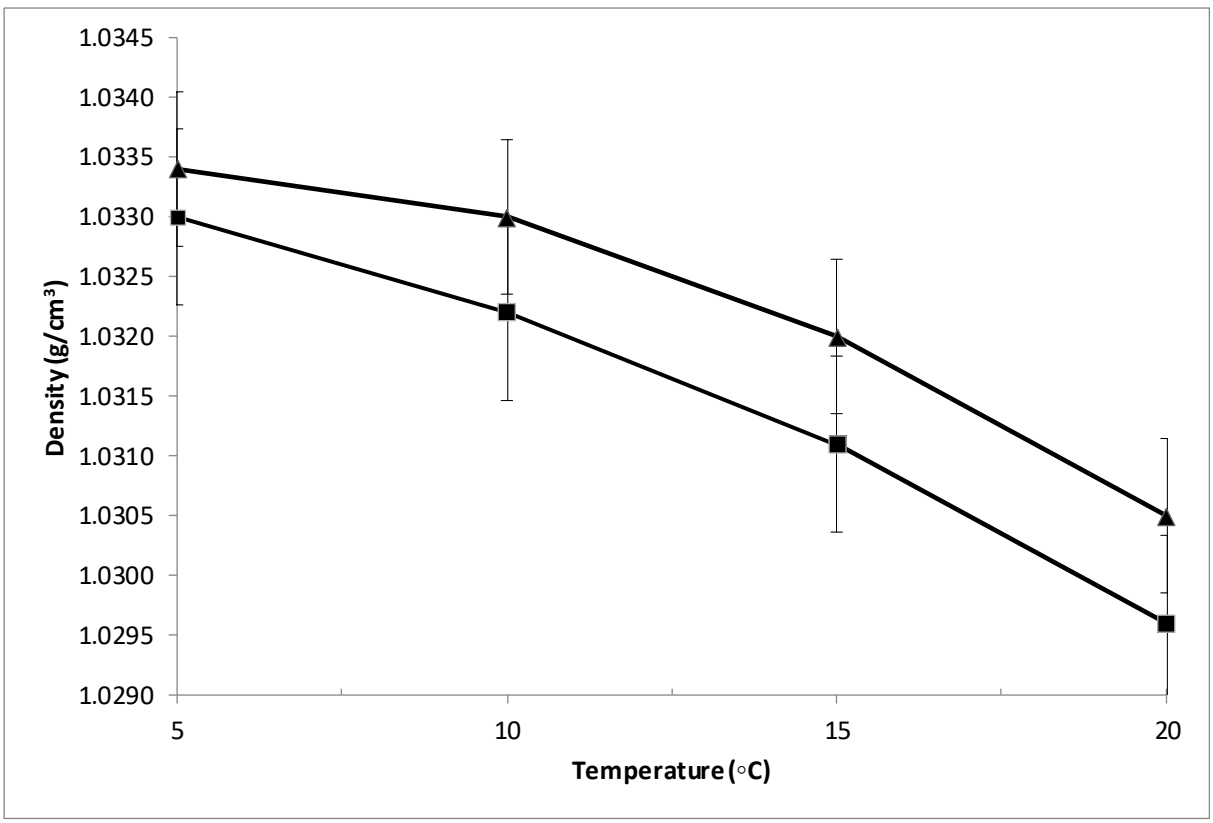


Figure 4.1 Density as a function of temperature for 2 different measuring devices, DMA4500 (▲) and DMA35 (■) adjusted for sampling period, effect of measurement technique and random effects of cow

Table 4.4 highlights the significance of density as a conversion factor in weight-volume calculations for the dairy industry. The effect of milk density on the milk payment for the farmers was evaluated considering the estimation of total milk solids at different temperatures. The data shown in Table 4 was obtained from the Irish Central Statistics Office for the year 2018 and the current density value, 1.0297 g/cm^3, used for weight-volume calculations, was obtained from interactions with industry personnel. Irish dairy farmers produced approx. 7.576 x10^9 L of milk in 2018, which when converted to weight using the current density factor of 1.0297 g/cm^3 gives approx. 7.801 x10^9 kg of milk. The same produced volume multiplied by a

density factor of 1.0334 g/cm^3, as determined in this study, gives approx. 7.83 billion kg of milk, a difference of approx. 28.03 million kg of milk (Table 4.4). When this is equated to kilograms of fat and protein across the entire industry as a whole in 2018, it represents just over 1 million kilograms of protein and over 1.1 million kilograms of fat.

Table 4.4 Weight-volume relationships for Irish milk volumes in 2018, showing differences in fat mass for historical density factor (1.0297 g/cm^3) and the new density factor (1.0334 g/cm^3) at 5 °C.

Description	Density factor (g/cm^3)	Quantity (in millions)
Estimated volume of milk produced (L)		7,576.00
Estimated weight of milk (kg)	1.0297	7,801.01
	1.0334	7,829.04
Difference in weight estimation (kg)		28.03
Variance in fat at 4.14% (kg)		1.16
Variance in protein at 3.61% (kg)		1.01

Table 4.5 shows the estimates of variance components for cow, residual and total variation; this parameter was analysed to determine the effect of cow on milk density. Variation between cows accounted for 61.1% of the total variation for milk density, and 39% of the total variation was explained by other environmental factors not accounted for in the model.

Table 4.5 Estimates of variance components for random animal effects and repeatability of milk density

Cov Parm[1]	Estimate (x10^{-7})	Repeatability
Cow	6.04	61.13%
Residual	3.84	
	9.87	

Total	

[1]= Covariance parameter

4.4 Discussion

Changes in milk density on changing temperature are dependent upon its constituents, especially water and fat (Hlaváč and Božiková, 2011; Parmar et al., 2020; Short, 1955) and may be attributed to the thermal expansion characteristics of fat in milk (Richmond and Davis, 1953). The estimate of repeatability for milk density in our study (61%) was similar to the estimates of repeatability for contents of fat, protein and lactose (Costa et al., 2019) meaning that genetic and permanents effects of the cows are important in explaining the phenotypic variation for milk density during the lactation. The analysis of variance indicated that 39% of the phenotypic variance was explained by environmental factors. Research conducted in the past shows that the changes in density and volume of milk are greater than when compared to water when subjected to different temperatures (Short, 1955). A study to determine the density of water (Lewin, 1972) showed that the density of water peaks at 3.98 °C and maintains a linear relationship with temperature; the density of water does not vary significantly with increasing temperature (1.000 at 4°C to 0.99802 at 20°C) (USGS, 2018).

Previous research suggested that the density of milk decreases with increasing temperature up to 40 °C (Short, 1955; 1956). Another study (Hilker and Caldwell, 1961) measured density of milk between 2.2 °C and 74 °C, and found that minimum density was observed at 74 °C, while the highest value was observed at the lowest temperature. Additionally, it has been reported that the maximum density value for milk was reported between 2 different

temperatures, i.e., -0.6°C and -0.3°C (Davies, 1939; Olson, 1950), respectively. All the density results from past studies are in line with results from our study, with the highest density being recorded at the lowest temperature and vice versa. Watson & Tittsler (1961) assessed the density of raw milk between 1 and 10 °C to replicate a range of milk handling conditions and determined a predictive best-fit equation that could be used to estimate density using fat, solids-not-fat (SNF) and temperature parameters. These authors evaluated density at 4 °C and obtained an average value of 1.0344. However, it was found that most density values were overestimated, and the residual errors became larger as the predicted density increased. This may be attributed to the method used for determining milk density (Ueda, 1999). Further research corroborating this point was shown when the Babcock and Mojonnier method were compared (Goff & Hill, 1993), where the fat content estimated by the Babcock method produced higher results than the Mojonnier method. Research from the USDA (1965) also pointed out that specific gravity measured by a Lactometer in the method used (Watson & Tittsler, 1961) was lower than that determined with the Babcock bottle method (USDA, 1965). In addition, solid and liquid fat fractions in milk affect density, and are determined by temperature at the time of measurement and the temperature history of the sample (Hlaváč & Božiková, 2011; McCarthy & Singh, 2009). Milk fat is liquid at temperatures above 40 °C and is solidified at -40°C; it is in intermediate state as a mixture of, crystals and oil at temperatures between 40 and -40°C (Walstra, 1999). Temperature affects the physical state of fat available in milk and the fat begins to crystallize as the temperature drops. Increasing the fat crystallization process leads to an increase in milk density. Milk density, as measured in this study, was highest at 5°C (1.0334 g/cm^3) and, as the temperature increased, melting of fats decreased density. It may also be noted that, the higher the fat content in milk, the more density varies with increasing temperature, because the volume of fat varies more with temperature compared to water.

4.4.1 Effect of analytical method on density results

Milk density measured for the samples in this study at different temperatures was also impacted by the use of different measuring methods. Referring to the results of milk density for both DMA35 and DMA4500 at the measured temperature, both systems showed a very similar trend, although there were differences in the absolute numbers, with the DMA35 showing a consistently lower density than the DMA4500.

The DMA35 is used regularly in the dairy industry for rapid measurement for milk density (personal communications from industry personnel) and measures the density based on oscillating U-tube technology. The frequency of the oscillator changes due to introduction of liquids, and this variation in natural frequency of the oscillator enables density measurement (Paar, 2009). The effect of instrument was assessed calculating the density of milk at different temperatures after adjusting for any variations introduced due to sampling period, instrument and random effects of cow. Several researchers have determined the controlled temperature history necessary for high precision and accurate density measurement (Sharp & Hart, 1936; Vanstone & Dougall 1960; Hilker & Caldwell, 1961). However, for this study, the temperature history did not affect the results because all the samples were subjected to the same procedure and temperature history (equilibrated at each temperature for same time duration).

4.4.2 Implications of milk density measured at temperature (5°C) on milk payment

Previous research (Shalloo, Dillon & Wallace, 2008) suggested that milk procured from dairy farmers should be paid for based on a multi-component pricing system, i.e., A+B-C system, which has been used in many countries around the world (e.g. Denmark, Australia, Holland, New Zealand etc.), including Ireland, for approximately 10 years. This system works by putting a value on the kg of protein (A) and fat (B) supplied by farmer to the processor and deducts the cost of collection and processing (C) related to the volume of milk supplied by the farmer.

Currently, milk is collected at the farm at ~ 4-5°C and, presently, the processors' payment system quantifies the amount of fat and protein using milk volume in litres, milk fat and protein concentration and a density factor of 1.0297 g/cm^3 for the weight-volume relationship. The reduction in the density of milk with increasing temperature has been noted. Furthermore, it has been found that as the fat content of milk increases, there are larger density changes with temperature variations (Paar, 2009). The density factor is used to convert the volume of milk from litres to weight (kilos) by multiplying the volume of milk with the density factor, i.e., 1 L of milk at density factor 1.0297 g/cm^3 weighs 1.0297 kg. The density factor is also used when calculating the amount of fat and protein in milk by multiplying the volume of milk in litres to estimate the weight of milk and multiplying by the concentrations of fat and protein (%) in milk, which generates the mass of fat and protein in milk, respectively. As revealed by the results of this study, milk density varies at different temperatures (reducing with increasing temperature) and significantly impacts the weight-volume calculations.

Density may also be used to calculate the amount of milk solids as depicted by Fleischmann's formula (Ullmann et al.,1985):

$$\mathrm{TS} = 1.2 * \mathrm{F} + 266.5 * \frac{(\mathrm{S} - 1)}{\mathrm{S}}$$

where TS is total milk solids, F is the fat content in milk (both in %) and S is the density

The above formula shows the importance of milk density and thus implies that total milk solids content estimated at lower temperature (5 °C) will be higher than total milk solids estimated at higher temperatures of approx. 20 °C. The results of density estimated in this study were based on a mass per mass basis. The new density factor of 1.0334 g/cm^3 may be used for volume-weight conversion, i.e., 1 L of milk with the new factor will weigh 1.0334 kg. This may enable a more precise estimation of fat and protein quantity in milk. An example of the use of the density conversion at the same temperature (5°C) in weight volume relationships is shown in Table 4.4. For total milk produced in Ireland in the year 2018, a significant difference in

mass estimation of individual constituents (1.16 million kg in fat and 1.011 million kg in protein) is observed between the use of historical factor, 1.0297 g/cm^3, and the new factor, 1.0334 g/cm^3. While saying this, it is important to note that while there may have been more kilograms of fat and protein in the milk (at milk density 1.0334 g/cm^3) than the conversion factor of 1.0297 g/cm^3, in reality, this does not mean that there will be more money to pay out in milk price, but will mean that allocation of payment is aligned with increased levels of milk solids.

However, over time, improving milk payment systems is one of the key areas in developing better communication mechanisms between the farmer and the processor. Ensuring the accuracy of this communication is key to ensuring thrust on both sides. Within the processing plant, accurate measurement of incoming milk constituents, process control and monitoring allocation for product mix under different processing conditions will ensure that any issues that become apparent are identified early and appropriate remedies are put in place in an efficient manner. Accurate monitoring and measurement of temperature and its effect on raw milk density through the quadratic model suggested earlier will enable improvement in milk payment models and impact on the appropriate product mix for processors and profitability of both dairy farmers and processors.

The model developed may enable farmers to estimate density changes based on changes in temperature of milk samples, and the density factor thus estimated can help in measuring the total solids content in milk. The volume of milk produced and supplied from Irish dairy farms has significantly increased since the removal of EU milk quotas, and this research aligns with the current trend, enabling accurate measurement of milk solids and directly impacting the profitability of both dairy farmers and processing industries. The results of this study can be effectively utilised by processors during weight-volume calculations to accurately record the amount of total solids incoming at the plant gates and also monitor and control the milk constituents' conversion process with better efficiency. The temperatures observed in the

study were in line with prevalent processing conditions observed at dairy plants (personal communication with dairy plant managers and professionals).

4.5 Conclusion

The intake temperature of milk on farm significantly affects whole milk density, along with other external factors such as composition and processing conditions. There is an inverse relationship between temperature and density, i.e., density of milk decreases with increasing temperature, and there is also a quadratic effect of temperature on milk density. To the best of our knowledge, this study is the first of its kind for the Irish dairy sector and generates a new density conversion factor to be used, for example, in the A+B-C milk payment system currently followed in the Irish dairy sector. The results from this study for measurement of density at specific temperatures will help to address any bias in weight-volume calculations and thus may also improve the financial and operational control for the dairy processors in Ireland and internationally.

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Chapter 5

Development and evaluation of a processing sector model for butter manufacture using a mass balance technique at two dairy processing sites

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Abstract

The butter manufacturing process at two different commercial dairy processing sites in Ireland was evaluated using a mass balance approach to develop, evaluate and validate a processing sector model of the flow of milk fat from intake to final product. The mass balance was represented as a function of fat intake = fat in products + fat losses + recycled fat. Representative samples of all products, namely whole milk, cream, skim milk, butter, buttermilk and cleaning-in-place streams (cream silo flush, butter churn residue and sludge), were collected from two different sites. Milk fat levels and product quantities were measured to obtain the fat outputs. Total fat losses at the end of butter production ranged between 1.90% and 2.25% of the total fat input for both sites. Three different scenarios were examined to evaluate the model: S1 (Animal Breed) high genetic merit (Elite) and national average (NA) Holstein Friesian (HF) cows were evaluated, for their effect on the net value of milk; S2 (Product Portfolio) a mixed product portfolio of cheese, butter and skim milk powder (SMP) was compared to a product portfolio comprised of butter alone; and S3 (Process Efficiency) the impact of varying process losses on net values of milk and the quantities of products produced was simulated. The value per 1000 L of milk for S1 was €410.69 and €393.20 for Elite and NA cow's milk, respectively. For S2, the butter-only product portfolio returned €355.10, whereas the mixed-products portfolio returned €369.60. Lastly, S3 corresponding returns for 1%, 2.2% and 5% losses was €365.90, €361.47 and €351.12, respectively.

5.1 Introduction

Milk and dairy products are major constituents of western diets and there has been an ever growing demand for high quality dairy products in these regions (Heck et al. 2009). Various factors affect the demand and supply of dairy products, including product formulations, variations in milk supply, seasonality, consumer perceptions, and fluctuations in customer demand (Chen et al. 2014). Seasonal variations in milk composition pose a significant challenge to the processing industry and the ultimate product mix and quality of products (Auldist et al. 1998; Lindmark-Månsson et al. 2003). With the removal of European Union (EU) milk quotas in 2015, there has been a consequent increase in production across the EU. For example, in the Irish dairy industry, milk production has increased from ~under 5 billion litres at the end of 2009 to 7.57 billion litres at the end of 2018 (CSO, 2018). This has occurred at a time where there is significant price volatility, which presents a challenge to the dairy industry. Demand variations govern the price of dairy products, and a small change in demand can have a significant impact on prices (Vitaliano, 2016; Stephenson and Nicholson, 2018). Such uncertain scenarios are manifested by steep changes in the prices of dairy products, especially butter prices, which have almost doubled from 2015 to 2018 (CLAL, 2019a; GDT, 2019a). One of the reasons behind the increasing prices of butter could be the 're-profiling' of butter as a healthy food product. Exports for butter from the EU region has increased significantly (7.4% export growth in first quarter of 2019) (IFA, 2019). The increase in production has been attributed to growing demand from developing countries, while consumption from western countries has been stagnant or dropped slightly (Vitaliano, 2016; Kiernan, 2019). Demand for dairy proteins and powders such as SMP has increased considerably, with the SMP price index rising by 3.2% in 2018 (O'Brien, 2019), while SMP production from the Irish dairy sector alone has also increased considerably from ~120,000 tons at the end of 2017 to ~134,000 tons in 2018 (CLAL, 2019b). These changes pose a

significant challenge for processors, and processors who are responsive to this variability in the market will attain a higher rate of return and thus be more economically sustainable (Geary et al. 2010).

Various processing models have been developed and studied around the world using a mass balance approach. Mass balance approaches have been in practice for a long time and have been applied across diverse fields like climate studies (Medwedeff and Roe, 2017), environmental monitoring (Ashfaq et al. 2017; Irvine et al. 2017), chemical analysis (Little et al. 2014; Chen et al. 2015), engineering (Fahrenfeld et al. 2014) and energy balance analysis (Brock et al. 2000). The mass balance approach is central to the evaluation of processing efficiency as regards to yields of products and waste.

In dairy processing, mass balances have been implemented across diverse segments ranging from the estimation of milk constituents like fat protein and lactose (Bangstra et al. 1988; Garrick and Lopez-Villalobos, 2000; Bailey et al. 2005; Geary et al. 2010; Sneddon et al. 2016), and comparing process-based models for nitrogen, phosphorus and greenhouse gases (GHG) impact developing models associated with associated with milk production at the animal, field and farm-scale (Spears et al. 2003; Veltman et al. 2017). The objective of this study was to develop, evaluate and validate a mass balance model for the milk fat conversion process and to apply the model across two dairy processing sites in Ireland.

5.2 Materials and Methods

5.2.1 Mass balance approach

The principle of a mass balance is based on the law of conservation of mass. The mass balance equation (Warn and Brew, 1980) is represented as :

Mass in = Mass out + Mass stored

Or

Raw Materials = Products + Wastes+ Stored Materials

For a butter manufacture process, it may be stated as

Fat Intake = Fat in products + Fat losses + Recycled Fat + Excess fat sold

Where, Fat intake = fat content of the total milk volume processed (kg); Fat in products = fat in each of the products produced (kg); Fat losses = fat lost during processing (kg); Recycled fat = fat collected from cleaning-in-place (CIP) activities such as cream silo flush and churn residue flush and sent into separation again (kg); Excess fat sold = any fat not used in the production of products sold to internal/external customers (kg).

A protocol was shared with all the participating sites to organize the mass balance exercise. The exercise of following fat conversion within the dairy processing environment was monitored as a batch with one or two silos of whole milk being processed to butter in a closed loop procedure. The process was divided over a period of two days, with the first day being dedicated to gathering samples and data for raw milk and the separation process while the second day was dedicated to the butter manufacture process, with samples collected for butter, buttermilk, and CIP streams. Raw milk arrived at plant sites in bulk tankers and samples were collected off the back of the tanker to test for antibiotics. Once the sample passed the antibiotic test, it was transferred to designated silos for the mass balance study. Composite representative samples of milk were taken as the silos were emptied for separation process. During the separation process, representative skim milk samples were collected once every hour.

For Day 2, representative samples of cream stored overnight (continuously agitated to avoid fat separation) was collected as the cream was emptied for butter manufacture. As the butter manufacture process continued, buttermilk produced was collected in an assigned silo to measure the total volume produced. Once the butter manufacture process was completed, the total weight of butter and volume of buttermilk produced was measured. A CIP process was initiated at the end of butter manufacture with CIP being completed in the cream silo, packing

line and butter churn. CIP flows from these points were collected in individual intermediate bulk containers and weighed to determine the volume generated. Three samples of each of the CIP streams were collected for testing. After sample collection, the samples were tested for fat content using Rose Gottlieb Method.

The fat mass balance was calculated by multiplying the volume of incoming milk from intake by the density conversion factor to obtain weight of milk. The weight of incoming milk was then multiplied by the fat content in whole milk to attain the fat available for conversion to butter and other products. Similarly, the cream and skim volumes produced were multiplied by the fat contents to obtain the fat mass in cream and skim available. This exercise was completed at all the stages within the fat processing value chain for other products, i.e., buttermilk, butter, CIP streams – cream silo flush, butter churn residue and final sludge by multiplying the fat content with weight of each product produced. The difference of fat from intake to end of separation and butter manufacture process was calculated as:

Loss at Separation = Fat Intake – (Fat in cream + Fat in Skim milk)

Loss at butter production = Fat in cream – (Fat in Butter + Fat in buttermilk)

5.2.2 Model Description

All inputs, outputs and losses within the dairy processing steps were accounted for in the mass balance model. The model is a mathematical representation of the conversion of milk fat into the butter. The model inputs included the volumes and composition of milk intake and product portfolio, i.e., butter, buttermilk, skim milk and CIP discharges and their composition, all of which were used in the mass balance calculations. The final quantities of each of the products in the portfolio were also noted and used as inputs in the model. Costs of milk processing, along with costs of collection and standardization were estimated to determine the economics of the mass balance model. The availability of actual cost estimates was a challenge in developing and refining this model. However, lack of the real-time costs information was

overcome by using data from past studies (Quinlan et al. 2006; Breen et al. 2007; Geary et al. 2010), control reports and rigorous consultation with dairy industry professionals. Different scenarios were examined and their impact on net returns was estimated. The schematic diagram of the dairy processing sector model for fat conversion is shown in Figure 5.1. Site 1 and Site 2 were located in different regions of Ireland with differences in their milk supply profile, processing capacities, demand and customer requirements, plant set-up (number of silos studied, number of separators, CIP practices), period of analysis and management practices at the plants.

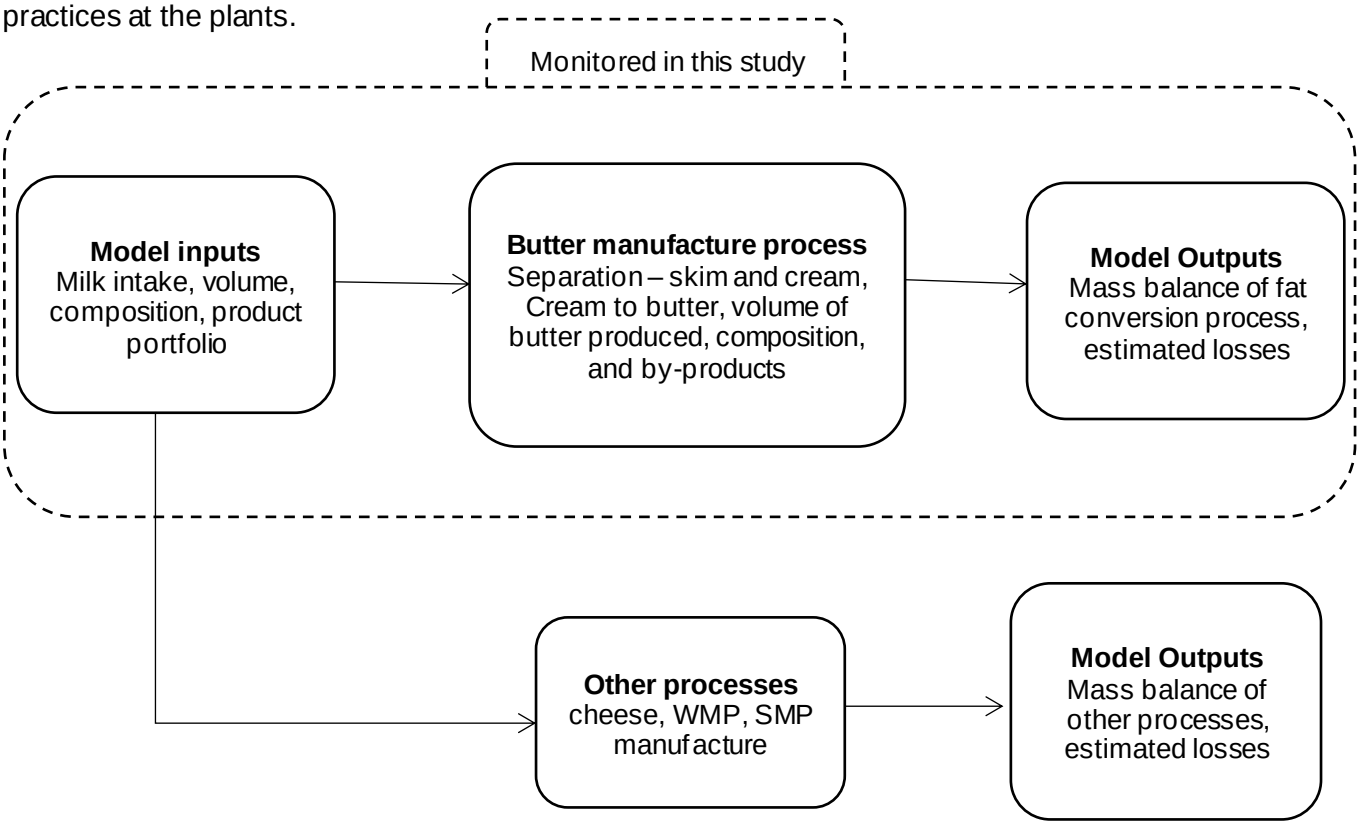


Figure 5.1 Schematic representation of the dairy processing sector model for butter manufacture. (WMP= Whole milk powder, SMP= Skim milk powder)

5.2.3 Financial Components

Market returns The market values for calculating returns from the mass balance model were taken from the Global Dairy Trade (GDT) website (https://www.globaldairytrade.info/) (GDT,

2019a) and were representative of a 2-year average for 2017-2018. The market price obtained from the 2-year average for butter was ~USD 5,027/T. Average product prices for other products for the 2-year period are shown in Table 5.3.

Processing cost

Processing costs, including volume-related costs associated with collection, standardisation and processing of milk, were gathered from a study on Irish dairy processing cost analysis (Breen et al. 2007) and using different indices such as industrial price index, wholesale price index and information the Central Statistics Office of Ireland (CSO, 2018), along with consultation with dairy industry professionals and experts. Processing costs also included product-related costs as associated with processing, packaging, transportation and storage and marketing costs, all adjusted for 2018 levels (Quinlan et al. 2006; Geary et al. 2010; Heinschink et al. 2012). The processing costs for manufacture of butter and other products are summarized in Table 5.1.

Milk price

Marginal rate of technical substitution (MRTS) was used to determine the value of milk fat and protein per kg. It is described as the amount by which one input can be reduced when one additional unit of another input is used so that the overall outcome remains constant. In this case, one unit of protein can be reduced to add one extra unit of fat to keep the overall milk value constant. It is represented mathematically as:

$$\mathrm{MRTS}\ (\mathrm{x1},\mathrm{x2}) = \frac{\Delta \mathrm{x2}}{\Delta \mathrm{x1}} = \frac{-\ \mathrm{MP1}}{\mathrm{MP2}}$$

Where, MP1 and MP2 are the marginal products of input 1 and input 2, respectively. For every additional kg of input, fat or protein, the overall revenue from milk will be increased depending upon product portfolio, processing costs and market value.

Value of Milk The net value of milk may be calculated by subtracting the costs of converting milk to butter from the total volume of butter produced and the market value of butter obtained.

Net value of milk = $\sum(v * p) - c$

where v is the volume of butter produced, p is the market price of butter and c is the costs of processing milk to butter.

5.2.4 Model Evaluation

Scenario Analysis

Three different scenarios (S1, S2 and S3) were explored for variations in milk values depending on: (i) two genetic groups representative of high genetic merit (Elite) and national average (NA) Holstein Friesian (HF) cows; (ii) a plant producing only butter as a product compared to a product mix of cheese (37.6%), Skim milk powder (SMP) (22.5%) and butter (39.9%); and (iii) an increase or decrease in processing efficiencies at the plants. The product portfolio details were derived from Central Statistics Office, Ireland website (CSO, 2018) and included mainly cheese, butter and skimmed milk powder (SMP). The percentages for cheese, butter, SMP, and whole milk powder (WMP) were calculated as a proportion of the cumulative tonnes that were produced.

Scenario 1 (S1)

The first scenario used the model to evaluate net value of milk and the quantities of product produced from 1000 litres (L) milk from Elite and NA HF cows assessed, with 39.9% of milk intake used in the production of butter, 37.6% into cheese, and 22.5% into SMP (CSO, 2018). The by-products of this portfolio are Whey and Buttermilk Powder (BMP).

Scenario 2 (S2)

In the second scenario, a product portfolio comprised of cheese, butter and SMP was investigated with 39.9% of milk intake used in the production of butter, 37.6% into cheese, and 22.5% into SMP as compared to if only butter as an end product (100% milk allocated for butter manufacture) was manufactured. SMP and BMP are by-products of this product portfolio.

Scenario 3 (S3)

In the last scenario, the impact of varying process efficiencies (increasing or decreasing losses) on net values of milk and the quantities of products produced was examined. The composition of products simulated in the model is shown in Table 5.2.

5.3. Results

Table 5.1 presents the processing cost, including volume-related and product-related costs, for a product mix, i.e., butter, buttermilk powder, skim milk powder, cheese and whole milk powder, taken from literature (Quinlan et al. 2006; Breen et al. 2007) and adjusted up to 2018 levels using inflation and price indices. The average processing costs including volume-related, and product costs such as marketing, packaging, storage and distribution etc. were ~0.04 cents/litre (c/L).

Table 5.1 Processing costs including volume-related and product-related costs for butter adjusted up to 2018 levels (Quinlan et al., 2006, Breen et al., 2007)

Cost	Butter	Cheese	WMP	SMP	BMP
Volume costs , €/L					
Collection[1]	0.0126	0.0126	0.0126	0.0126	0.0126
Standardization	0.0051	0.0051	0.0051	0.0051	0.0051
Processing[2]	0.0042	0.0128	0.0101	0.0128	0.0101
Product costs, €/MT					
Processing[2]	99.89	127.44	178.34	175.68	178.34
Packaging[2]	31.78	41.45	41.45	41.45	41.45
Storage[3]	75.10	44.06	28.66	7.96	28.66
Distribution[2]	73.00	73.00	73.00	73.00	73.00

Marketing	50.70	50.70	50.70	50.70	50.70

(WMP- Whole milk powder, BMP- buttermilk powder and SMP- skim milk powder)

[1] Quinlan, C, Keane, M, Enright, P and O'Connor, D (2006) The milk transport cost implications of alternative dairy factory locations. Agribusiness Discussion Paper.

[2] Breen, J, Wallace, M, Crosse, S and O'Callaghan, D (2007) A new direction for the payment of milk: Technological and seasonality considerations in multiple component milk pricing of milk (liquid and manufacturing) for a diversifying dairy industry.

[3] Dairy professionals and expert consultation.

Table 5.2 shows the composition of different products simulated using the model for estimation of net value of milk. The milk constituent content for each product in the product mix were in line with Codex Standards, i.e., fat in butter was taken as 82%, while the fat level was 35% in cheese.

Table 5.2 Composition of different products simulated using the model of Geary et al. (2010) for estimation of net value of milk

Item	Cheese	Butter	WMP	SMP	Whey	BMP
Fat, %	35.00	82.00	26.50	1.00	1.00	8.30
Protein, %	24.50	0.59	25.10	33.00	15.15	41.72
Lactose, %	1.39	0.79	39.80	54.00	77.15	40.32
Minerals, %	2.15	0.12	5.90	8.00	4.32	4.66
Water, %	35.26	16.50	2.70	4.00	2.38	5.00

WMP= Whole milk powder, SMP= Skim milk Powder, BMP = Buttermilk powder

Table 5.3 shows the average market value of the products simulated using the model, with average butter price for years 2017-2018 at approx. 5027 USD/MT (metric ton), buttermilk powder ~USD 2170/MT, SMP priced at @USD 2000/MT and cheese price of USD ~3720/MT.

	Butter		BMP		SMP		WMP		Cheese	
Period	**2017**	**2018**	**2017**	**2018**	**2017**	**2018**	**2017**	**2018**	**2017**	**2018**
Jan	4,345.0	4,699.0	2,726.5	1,866.0	2,636.0	1,758.5	3,288.5	2,948.0	3,917.0	3,401.5
Feb	4,595.5	5,305.5	2,172.0	2,039.0	2,591.0	1,882.0	3,251.5	3,236.0	3,694.0	3,712.5
Mar	4,781.5	5,280.5	1,846.0	1,959.0	2,033.0	1,969.0	2,818.5	3,229.0	3,420.5	3,684.0
Apr	4,821.5	5,574.0	1,654.0	1,988.0	1,978.5	1,881.0	2,961.0	3,294.5	3,375.0	3,767.0
May	5,195.0	5,717.0	1,919.5	1,990.0	1,990.0	2,023.0	3,272.5	3,228.5	3,696.0	4,114.5
Jun	5,699.5	5,596.0	2,092.0	2,314.0	2,187.0	2,027.0	3,082.5	3,197.0	4,203.0	3,922.5
Jul	5,889.5	5,171.5	2,264.0	2,352.5	2,057.0	1,936.0	3,112.5	2,939.0	4,081.5	3,654.5
Aug	5,741.0	4,597.0	2,198.0	2,441.0	1,967.0	1,961.5	3,149.0	2,920.5	3,968.5	3,573.5
Sept	5,990.0	4,270.5	2,026.0	2,474.0	1,932.0	1,992.5	3,111.0	2,794.5	4,075.0	3,567.0
Oct	5,786.5	4,065.0	1,804.0	2,522.0	1,846.0	1,979.5	3,025.5	2,741.0	4,108.0	3,436.0
Nov	5,330.0	3,841.0	1,931.0	2,568.0	1,759.5	1,981.0	2,815.0	2,627.0	3,916.0	3,251.0
Dec	4,524.5	3,836.5	1,957.0	2,973.0	1,724.5	2,006.0	2,792.5	2,670.5	3,542.5	3,223.5
Avg	5,225.0	4,829.5	2,049.2	2,290.5	2,058.5	1,949.8	3,056.7	2,985.5	3,833.1	3,609.0
	5,027.2		**2,169.9**		**2,004.1**		**3,021.1**		**3,721.0**	

BMP- Buttermilk Powder, SMP- Skim milk powder, WMP- Whole milk Powder
[1]MT= Metric ton = A unit of **weight** equal to 1,000 kilograms

Table 5.4 Quantities of products produced, and net value of milk after subtracting processing costs for each of the scenarios[1] simulated from the model

		Product mix (kg / 1000L Milk)						
	Desc.	**Cheese**	**SMP**	**Butter**	**Whey**	**BMP**	**Collection and processing value/L, €**	**Net milk value(€/1,000 L)**
S1	Elite[1]	48.20	52.40	36.50	23.60	4.40	−0.04	410.69
	NA[1]	46.60	52.40	33.80	23.10	3.90	−0.04	393.20
S2	All products	44.40	50.20	31.50	21.70	3.50	-0.04	369.60
	Only Butter	-	80.30	50.80	-	5.60	-0.04	355.10
S3 Loss	1% loss	44.00	49.60	31.20	21.50	3.40	-0.04	365.90
	2.2% loss	43.50	49.00	30.80	21.30	3.40	-0.04	361.47
	5% loss	42.20	47.60	30.00	20.60	3.30	-0.04	351.12

[1] Scenario 1 (S1) evaluated milk from high genetic merit (Elite) and national average (NA) Holstein-Friesian cows. Scenario 2 (S2) evaluated milk processed into two product portfolios, one with milk used for butter (39.9%), cheese (37.6%) and SMP (22.5%), and other with milk used for butter (100%). Scenario 3 (S3) evaluated different process efficiencies.

BMP- Buttermilk Powder, SMP- Skim milk powder

Site results

A total of ~521 MT of milk was processed at site 1, with an average fat content of 4.37%. Fat mass available for butter manufacture at site 1 was 22.76 MT (weight of milk * % fat). The milk collected was passed through a separation process, producing a total of 56.2 MT of cream

(38.77% fat) and ~468.57 MT skim milk with 0.10% fat (Table 5.5). Fat mass available in cream at site 1 was calculated as 21.79 MT and fat mass from skim milk was 0.47 MT. From the available fat mass in cream, butter was manufactured, with butter mass produced at site 1 ~ 26.83 MT at 80.50% fat (butter fat = 21.59 MT) (Table 5.5). Similarly, the weight of butter milk produced as a by-product of the process was also noted (28.25 MT buttermilk at 0.66% fat = 0.19 MT fat mass). The CIP process inputs, i.e., churn residue melt and cream silo flush were collected and weighed in an intermediate bulk container (IBC) to complete the mass balance approach (i.e., fat intake = fat in products + losses + recycled fat). Churn residue and silo steamed flush (steaming out the cream residue from cream silo) at site 1 was measured at 1.50 MT and 1.90 MT with fat content of 38.19% and 17.47% respectively (Table 5.5).

Table 5.5 Fat percentage, quantity of each product produced and fat mass in each product at each sub-stage of fat conversion process at the two sites monitored.

		Site 1			Site 2		
Stage	Product	Fat (%)	Qty prod. (MT)	Fat at each stage (MT)	Fat (%)	Qty prod. (MT)	Fat at each stage (MT)
Intake	Milk	4.37	520.88	22.76	4.03	555.21	22.39
Separation	Cream	38.77	56.20	21.79	47.64	46.00	21.92
	Skim	0.10	468.57	0.47	0.04	512.58	0.20
Butter	Butter	80.50	26.83	21.59	82.15	25.78	21.17
Process	BM	0.66	28.25	0.19	0.72	21.09	0.15
CIP	CHR	38.19	1.50	0.57	36.66	0.77	0.28
	St. Cr.	17.47	1.90	0.33	41.87	0.35	0.15

CIP = Cleaning in Place, BM = Buttermilk, CHR =Churn Residue, St. Cr. = Silo steamed Cream

The loss values from butter manufacture process, when divided by the incoming fat content (weight) from cream gave the percentage loss experienced during butter manufacture. Therefore, total fat mass obtained in butter and buttermilk, when subtracted from fat available for butter manufacture process (from cream), gave an estimate of the losses in the butter manufacture process (0.16 MT) at site 1. The calculation of total loss percentage formed the final step of the mass balance with cumulative losses from separation and butter manufacture process was 0.43 MT for site 1. The cumulative loss values (1.93%), when divided by the total incoming fat from milk, gave the total losses at the end of butter manufacture process (Table 5.6).

For site 2, ~555 MT of milk was collected and processed at site 2 with a fat content of 4.03% (fat mass ~ 22.16 MT). There was 46 MT fat in cream, with 512.58 MT of skim milk (0.04% fat) at site 2 (Table 5.5). The fat mass obtained in cream from site 2 was 21.92 MT and the difference (incoming fat – fat in cream + fat in skim) when divided by the total incoming fat in milk gave the fat loss % at separation stage = 2.21% (Table 5.6). Amount of butter produced was ~25.78 MT at 82.15% fat while butter fat mass was 21.17 MT for site 2 (Table 5.6). Processing at site 2 produced 21.09 MT buttermilk with 0.72% fat (0.15 MT fat mass). Site 2 churn residues and silo steamed cream were 0.77 MT and 0.35 MT with 36.66% and 41.87% fat content, respectively. Total fat mass obtained in butter and buttermilk when subtracted from fat available for butter manufacture process (from cream) gave the estimate of loss at butter manufacture process. Thus, the total loss for butter manufacture was 0.01 MT at site 2; the cumulative losses from separation and butter manufacture process was 0.51 MT at site 2 (2.25% of the total fat mass in the system).

Table 5.6 Fat content at each sub-stage and using mass balance approach to determine the losses for different processes for Sites 1 and 2 monitored

SNo	Fat content in each process	Site 1 (MT)	Site 2 (MT)
1	Incoming fat in milk	22.39	22.76
2	Fat in cream	21.92	21.79
3	Fat in skim milk	0.20	0.47
4	Difference	0.27	0.50
5	% loss in separation process	1.21	2.21
6	Fat in butter process (butter+ buttermilk)	21.76	21.78
7	Difference ((2) – (6)) (T)	0.16	0.01
8	% loss in butter manufacture process ((7)/(2))	0.73	0.05
9	Total difference ((6)+ (3)) (T)	0.43	0.51
10	Total fat loss at the end of butter production (%)	1.93	2.25

(SNo = Serial number)

Scenario Analysis

S1: 1000 L of milk from Elite HF and NA HF cows yielded 48.20 and 46.60 kg cheese, 52.40 kg SMP for both genetic groups; 36.50 and 33.80 kg butter, 23.60 and 23.10 kg of Whey; and 4.40 and 3.90 kg BMP, respectively. The net value of milk after deducting processing costs of milk was € 410.69 and € 393.20 for Elite and NA HF cows, respectively. These values might seem a bit higher than expected since the estimated value of milk here does not include a margin.

S2: 1000 L of milk yielded 44.40 kg cheese, 50.20 kg SMP, 31.50 kg butter, 21.70 kg Whey

and 3.50 kg BMP for a plant producing the complete product mix, with net value of milk being € 369.60. For a plant producing only butter, the quantities of products produced were 80.30 kg SMP, 50.80 kg butter and 5.60 kg BMP, while the net value of milk was € 355.10. The values estimated under scenario 2 also follow trend from scenario 1 and do not include a margin, thus seeming relatively higher.

S3: In the final scenario, losses occurring in the processing of products were assessed at three different levels, i.e., 1%, 2.2% (as per the mass balance studies discussed in this paper) and a higher level of loss at 5% of the total milk processed (1000 L). The product portfolio included all products simulated, i.e., cheese, butter, SMP, Whey and BMP. As expected, the lower the losses, the higher the net value of milk generated. Losses at 1% in processing corresponded to 44.00 kg cheese, 49.60 kg SMP and 31.20 kg butter produced with Whey By-product and BMP. The net value of milk for 1% loss was the highest of the three % losses simulated, at € 365.90/1000L of milk. Losses in processing (as evaluated from mass balance exercise in this study ~ 2.2%) reflected lower quantities of products produced and a slight drop in net value of milk (€361.47/1000L milk). Lastly, a high loss of 5% was simulated, returning the lowest net value of milk per 1000L at € 351.12.

5.4. Discussion

5.4.1 Processing models

Various processing sector models have been developed and evolved over time as decision support tools for industry. One of the earlier models, developed by Pratt et al. (1997), was used to simulate and determine optimum mix for milk and milk products in terms of production and marketing. Similarly, another model by Benseman (1986) was developed for the New Zealand dairy industry for determining the most profitable product mix. Papadatos et al. (2002) developed a model for determining the revenue generated for cheese manufacturing process.

Geary et al. (2010) also developed a similar model simulating different production scenarios to identify the optimal product mix yielding best returns and accounting for variables such as market returns, processing costs and compositional changes for the Irish dairy industry. Burke (2006) used two mathematical models based on linear programming to quantify different parameters, such as net cost, revenues, and volume of cheese produced; the model developed for butter manufacture accounted for variations in product composition (fat content), processing costs, using market value to determine returns and net value of milk. Garrick and Lopez-Villalobos (2000) developed cost-price models to describe collection, processing and marketing activities for milk and dairy products, such as butter, cheese, casein and powders, for the New Zealand dairy industry. Milk processing is a highly complex process with challenges around variables such as seasonality, volume available, market demand, product portfolio and labour requirements (Burke, 2006; Geary et al. 2010). The impact of seasonality has not been addressed in the model developed in this study, and seasonality has a significant impact on milk composition, supply profile, associated production and labour costs, and demand. Adding these factors into the model may thus enhance the effectiveness of the model.

The transition of the Irish dairy sector from quota- and EU-support-based system to a global, market-driven scenario has presented many challenges in terms of price and income volatility, with higher investments into efficient dairy processing. Investments in the dairy processing sector have been strong, with approx. 1.2 c/l per year being invested back (2015-2017) into expansion of facilities in milk processing sector since the end of milk quotas in 2015 (Moran, 2018) and a further € 300 million investment being expected between 2018-2020. The increased investment has been justified by the relative increase of milk production and processed products.

5.4.2. Mass balance

In this study, the fat conversion process was studied as a batch or a closed-loop procedure to account for all the intakes, processing, and outflows, including sludge and CIP procedures. In the dairy industry, the application of mass balances may help in enhancing process efficiencies by underlining key focus areas and identifying key areas where there are losses within the system. The initial estimate for overall fat losses in fat conversion process, from interactions with the site professionals, was ~2.0-2.5%, but the actual points of loss within the whole fat conversion process were unknown.

Results in Table 5.5 highlight the efficacy of the mass balance approach allowing industry partners to identify key focus areas within the fat conversion process. For example, the fat content in skim milk at site 1 after separation was 0.10%, as compared to the industry benchmark of 0.06% (source: interactions with industry personnel). The results of fat content from skim and cream highlight the need to address the separation process, with focused investment or solutions needed to improve the separation efficiencies within plants. Higher separation efficiencies will mean lesser losses into skim milk, and thus, higher returns for processors.

Site 1 produced a larger volume of churn residue melt and cream silo flush compared to site 2 (Table 5.5). Although the melted CIP volumes were recycled for further fat recovery at both sites, dairy processing is a water- and energy-intensive process. A better control over CIP outflows would allow for reduced energy consumption and make the process more efficient. Improved monitoring of CIP process outflows, better process efficiency in terms of churn configuration, and operation, and optimised process controls like temperature and churn speed, were identified as improvement areas as a result of the mass balance model exercise for both sites.

5.4.3. Effect of density on the calculation of available milk fat

The milk intake at dairy processing plants is calculated in terms of volume (litres) but the production data is generated in terms of mass of fat or products produced. For the conversion of volume to weight, milk density is used wherein the volume of milk is multiplied by a density factor to give the effective weight of milk. Thus, a higher density factor will allow for a higher estimation of fat mass in milk (volume × density = mass) compared to a lower density factor. For example, 500,000 litres of milk with 4% fat converted to weight using 1.0297 g/cm^3 gives a fat mass of 22.59 MT, while using a density factor of 1.0320 g/cm^3 gives an estimated fat mass of 22.64 MT or 0.22% increase in fat estimation. Considering process efficiency values, if the correct conversion factor is not included, this will add to the incorrect assumptions around fat conversion. However, the density of milk is dependent upon the fat content of milk, and there are significant variations in milk fat content due to factors including composition and seasonal variation in milk, genotype and processing conditions, among other factors (Kelsey et al. 2003; Lock and Garnsworthy, 2003; Grimley et al. 2009; Heck et al. 2009; Chen et al. 2014; Liu et al. 2017).

The weight-volume conversion at both sites was being completed using a single annual average density factor which is not representative of changes in milk profile observed over the period of milk supply, i.e., compositional changes, supply conditions and lactation cycle and has not been changed in ~25 years within the industry (source - interactions with industry and academia professionals). Both sites monitored in this study used a single density conversion factor of 1.0297 g/cm^3 year-round to convert volume to weight during processing (source: interactions with industry personnel). Changes in practices such as farm management, animal welfare, improvement in genetics and other factors have led to variations in milk constituents and have significantly increased the milk solids content in Ireland. The average fat content in milk in Ireland has increased from 3.67% in 1998 to 4.14% in 2018, while the average protein

content has increased from 3.24% in 1998 to 3.48% in 2018 (CSO, 2018). As stated earlier, density variations are highly susceptible to changes in milk fat and solids-non-fat(SNF) content in milk. The increase in fat and SNF content from 1998 to 2018 would suggest that the corresponding density factor should also increase. These variations in milk fat and milk solids content have raised the question about accuracy and validity of using the old density factor and, therefore, appropriate density factors need to be developed to enable a well-rounded, accurate mass balance.

5.4.4. Breed and genetic merit

Cow breed has a major impact on the content of each of the constituents of milk (Auldist et al. 2004; Bailey et al. 2005). Milk composition has a significant bearing on the products portfolio produced, along with net value of milk constituents, net value of milk and total returns. Several researchers in the past have studied the impact of breed on variations in milk constituents (Garrick and Lopez-Villalobos, 2000; Auldist et al. 2004; Bailey et al. 2005; Geary et al. 2010). Fat and protein content were found to be higher in Jersey milk compared to HF milk, which greatly enhanced the returns while producing a product mix of 70% fluid milk and 30% cheese/WMP (Bailey et al. 2005). Similarly, casein content was also noted to be higher for Jersey milk, thus giving a higher yield of cheese and higher returns (Auldist et al. 2004). In our study, two genetic types of HF cows , Elite and NA, were analysed, and the results were in line with past studies (Garrick and Lopez-Villalobos, 2000; Auldist et al. 2004; Bailey et al. 2005; Geary et al. 2010), showing milk with higher solids content to yield higher product outputs and returns. Milk composition attributes, if included with genetics and breeding programs, can be highly effective in increasing the milk solids content and product yields and reducing costs by reducing energy, processing and fuel costs, and ultimately, yielding higher net returns for milk producers and processors.

5.5. Conclusion

Weight-volume calculations have been indentified as an issue for dairy processors in Ireland. Data analysis based on a mass balance approach helped to identify loss points in the process and also enabled processors to reconcile their test results. From an economic perspective, this study identified unaccounted-for losses in the process and helped monitor the overall financial performance. The model was developed to predict the net value of milk or returns based on different scenarios. The model may be a useful tool in determining the 'best-possible' scenario by evaluating all the possible variables like density factor, breed, and processing variations when scenario planning for processing. While, this model was developed for milk fat processing, i.e., the fat conversion process at a single point of time, with all milk intake being directed to butter manufacture. In reality, milk processing in a dairy environment involves different product mixes, dependent upon various parameters such as supply and demand profile, compositional variances, capacity constraints, estimating actual costs and returns. If all these parameters, along with a season-based density factor, are incorporated and analysed, it may enable the model to be more adaptive and precise decision-support to the changing dynamics of the dairy industry throughout the year.

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Chapter 6

General discussion

Declaration: This chapter was written by Puneet Parmar, with corrections and comments from Prof. Alan L. Kelly, Dr. Shane V. Crowley from University College Cork, and Dr. Laurence Shalloo from Teagasc Moorepark.

Since the removal of milk production quotas in 2015, Milk production in Ireland has been expanding and increased significantly. Milk production was reported at 5 billion litres in 2014 and it was projected to increase to 7.5 billion litres by 2020; however, this production target was already reached in 2018 (Central Statistics Office, 2019). This scenario is an opportunity for Irish dairy processors to expand its market worldwide; however, internal issues like low efficiency, unidentified losses etc. within the plant are major causes of concern and need to be addressed. One of the issues of unidentified losses may be attributed to the use of a single density factor for weight-volume calculations in mass balance to estimate the weight of milk constituents available for processing. Inaccurate estimation of milk constituents leads to an increased losses in the process and thus, lower efficiency and profitability.

To address the issue of appropriate density factors, the density factors were calculated based on multiple factors as highlighted in this thesis. The effect of compositional changes over a period of 9 months was studied in chapter 2 to determine season-based density factors which would be beneficial in improving the accuracy of mass balance, thus, enabling better calculation of milk constituents. Other parameters were categorized as separate studies and their impact on density factors was also evaluated. Chapter 3 studied the impact of cow genetic group on milk density also used to estimate density factors. It was observed that the breed Jersey, had the highest density value compared to the two genetic groups of Holstein-Friesian. The density of milk is dependent upon several factors, both internal and external, such as composition of milk, SNF content in milk, animal breed, stage of lactation etc. and external factors such as processing, agitation and homogenization. The results of this study may be used by farmers and processors in calculating the quantity of milk solids based on the genetic merit and may also be helpful in improving the milk payment systems. Another important factor playing a critical role in its impact on milk density, temperature, was also assessed in chapter

4. It was observed that as the temperature of milk sample increased, the density of milk reduced. This study enabled the establishment of a negative correlation between temperature and milk density. The density values obtained in this study at specific temperatures may be beneficial to address bias in weight-volume calculations and may help to improve operational control for dairy processors.

6.1. Mass balance process

A mass balance may be defined as the consideration of the inputs, outputs and distribution of a product/ingredient between streams in a process. For a butter manufacturing process, for example, it may be presented as follows (Short, 1955):

$$Fat\ intake = Fat\ in\ products + losses + recycled\ fat$$

The use of a density factor is important in mass balance calculations for identifying different loss-making points in a process, estimating losses in the fat conversion process and, subsequently, facilitating important process-related and investment-related decisions. Milk payment systems across different regions follow the multiple component pricing model (A + B - C system), where the value of protein (A) and fat (B) in kg supplied by the farmer to the processor are calculated and the cost of collection and processing (C) in cents per litre, related to the volume of milk supplied by the farmer, is deducted. Milk volume is converted to weight using the density conversion factor by multiplying the volume collected in litres on each farm by the density factor to obtain the weight of milk in kg.

The fat conversion process maybe studied as a batch or a closed-loop procedure to account for all the intakes, processing, and outflows, including sludge and CIP procedures. In the dairy industry, the application of mass balances helps in enhancing process efficiencies by underlining key focus areas and identifying key areas where there are losses within the system. The initial estimate for overall fat losses in fat conversion process was ~2.0-2.5%, but the actual points of loss within the whole fat conversion process were unknown. This estimate

of 2-2.5% was observed at a majority of the industry partner sites visited. The extensive mass balance study conducted at two partner sites (chapter 5) enabled the determination of actual loss points and loss values for both sites under analysis. Results in Chapter 5 highlight the efficacy of the mass balance approach allowing industry partners to identify key focus areas within the fat conversion process. For example, the fat content in skim milk at site 1 after separation was 0.10%, as compared to the industry benchmark of 0.06% (source: interactions with industry personnel). The fat content results for skim and cream highlight the need to address the separation process, with focused investment or solutions needed to improve the separation efficiencies within plants. Higher separation efficiencies will mean lesser losses into skim milk, and thus higher returns for processors.

Site 1 produced a larger volume of churn residue melt and cream silo flush compared to site 2. Although the melted CIP volumes were recycled for further fat recovery at both sites, dairy processing is a water- and energy-intensive process. A better control over CIP outflows would allow for reduced energy consumption and make the process more efficient. Improved monitoring of CIP process outflows, better process efficiency in terms of churn operating efficiency, and optimised process controls like temperature and churn speed, which were all identified as improvement areas as a result of the mass balance model exercise for both sites. The mass balance developed in our study as a result of the analysis of two dairy production sites depicted the first rigorous statistical analysis of data available from a dairy processing environment. Further collection of data and analysis is recommended to assess more scenarios based on market needs (supply/demand/production), with the view to enhance the robustness of this model and develop more comprehensive models integrating all the available data. This would be beneficial to develop a precise, more reliable decision-making tool based on comprehensive data.

6.2. Effect of density on milk fat calculation

The milk intake at dairy processing plants is calculated in terms of volume (litres) but the production data is generated in terms of mass of fat or products produced. For the conversion of volume to weight, milk density is used, wherein the volume of milk is multiplied by a density factor to give the effective weight of milk. Thus, a higher density factor will allow for a higher estimation of fat mass in milk (volume * density = mass) compared to a lower density factor. For example, 500,000 litres of milk with 4% fat converted to weight using a factor of 1.0297 g/cm^3 gives a fat mass of 22.59 MT, while using a density factor of 1.0320 g/cm^3 gives an estimated fat mass of 22.64 MT or 0.22% increase in fat estimation. Considering process efficiency values, if the correct conversion factor is not included, this will add to the incorrect assumptions around fat conversion. However, the density of milk is dependent upon the fat content of milk, and there are significant variations in milk fat content due to factors including composition and seasonal variation in milk, breed and processing conditions, among other factors (Kelsey et al. 2003; Lock and Garnsworthy, 2003; Grimley et al. 2009; Heck et al. 2009; Chen et al. 2014; Liu et al. 2017).

The weight-volume conversion at sites assessed was calculated using a single annual average density factor which is not representative of changes in milk profile observed over the period of milk supply, i.e., compositional changes, supply conditions and lactation stage. The density factor had not been changed in ~25 years within the industry (source: interactions with personnel in industry and academia). Both sites monitored in this study used a single density conversion factor of 1.0297 g/cm^3 year-round to convert volume to weight during processing (source: interactions with industry personnel).

Changes in practices such as farm management, animal welfare, improvements in genetics and other factors in recent years have led to variations in milk constituents and have significantly increased the milk solids content in Ireland. The average fat content in milk in Ireland has increased from 3.67% in 1998 to 4.14% in 2018, while the average protein content

has increased from 3.24% in 1998 to 3.48% in 2018 (CSO, 2018). As stated earlier, density variations are highly susceptible to changes in milk fat and SNF content in milk. These variations in milk solids have raised questions about accuracy and validity of using the old density factor and, therefore, appropriate density factors need to be developed to enable a well-rounded, accurate mass balance. Several other factors have direct or indirect impact on the density of milk and therefore, further analysis of the data is desirable, with the view of developing more comprehensive and robust density factors and associated models, which have an ability to integrate the different sets of data available from the experimental analysis.

6.3 Processing models

Milk processing is a highly complex process with challenges around variables such as seasonality, volume available, market demand, product portfolio and labour requirements (Burke, 2006; Geary et al. 2010). Various processing sector models have been developed and evolved over time as decision support tools for industry. One of the early models, by Benseman (1986) was developed for the New Zealand dairy industry to determine the most profitable product mix. Similarly, another model developed by Pratt et al. (1997), was used to simulate and determine optimum mix for milk and milk products in terms of production and marketing. Papadatos et al. (2002) developed a model for determining the revenue generated for a cheese manufacture process. Geary et al. (2010) also developed a similar model simulating different production scenarios to identify the optimal product mix yielding best returns and accounting for variables such as market returns, processing costs and compositional changes for the Irish dairy industry. Burke (2006) used two mathematical models based on linear programming to quantify different parameters, such as net cost, revenues, and volume of cheese produced; the model developed for butter manufacture accounted for variations in product composition (fat content), processing costs, using market value to determine returns and net value of milk. Garrick and Lopez-Villalobos (2000)

developed cost-price models to describe collection, processing and marketing activities for milk and dairy products, such as butter, cheese, casein and powders, for the New Zealand dairy industry.

The transition of the Irish dairy sector from quota- and EU-support-based system to a global, market-driven scenario has presented many challenges and opportunities in terms of price and income volatility, with higher investments into efficient dairy processing. Investments in the dairy processing sector have been strong, with approx. 1.2 c/L of all milk processed per year being invested back (2015-2017) into expansion of facilities in milk processing sector since the end of milk quotas in 2015 (Moran, 2018). In a report completed in 2020 it was shown that Dairygold who process approximately 18% of the national milk pool spent €389 million building additional processing capacity (Shalloo et al., 2020).

A processing model for fat conversion process (butter manufacture) was developed (chapter 5) using data captured from two dairy processing sites in Ireland. The processing model developed in chapter 5 incorporated the new density factors developed under chapter 2 to estimate the accurate content of milk solids at the two sites, which accounted for input into the model, alongwith the milk volume available for processing. The model utilized this value of milk fat available at the start of butter manufacture process, and data about the fat available in each sub-process, i.e., separation, butter manufacture and CIP. The model also considered three different scenarios- S1, where, net value of milk and quantity of product produced from 1000L of milk from two genetic groups of Holstein-Friesian cows, was calculated. S2- where two different product portfolios were compared and the net value of milk determined and S3- the impact of varying processing efficiencies on milk value was calculated. This model may thus be a useful tool to determine the most optimal scenario for a dairy processor based on several factors such as milk density, animal breed and processing efficiency within the plant.

6.4. Breed and genetic merit

Cow breed has a major impact on the content of each of the constituents of milk (Auldist et al. 2004; Bailey et al. 2005) along with different characteristics of milk such as composition profile, fatty acid profile, processability etc. (Bland et al., 2015; Kelsey, Corl, Collier, & Bauman, 2003; Lock & Bauman, 2004; Malossini, Bovolenta, Piras, Dalla Rosa, & Ventura, 1996; Penasa et al., 2014; Stocco et al., 2017; Yang, 2013; Tyrisevä et al., 2004). Milk composition has a significant bearing on the products portfolio produced, along with net value of milk constituents, net value of milk and total returns. Several researchers in the past have studied the impact of breed on variations in milk constituents (Garrick and Lopez-Villalobos, 2000; Auldist et al. 2004; Bailey et al. 2005; Geary et al. 2010). Fat and protein content were found to be higher in Jersey milk compared to HF (Holstein-Friesian) milk, which greatly enhanced the returns while producing a product mix of 70% fluid milk and 30% cheese/WMP (Bailey et al. 2005). Similarly, casein content was also noted to be higher for Jersey milk compared to HF cow milk, thus giving a higher yield of cheese and higher returns (Auldist et al. 2004). However, the impact of breed on raw milk density have not been widely addressed, to the best of our knowledge.

Because of genetic background and traits, milk samples collected from different cattle genetic groups have diverse compositional profile. Milk density is directly dependent upon the variations in sizes of milk fat globule, which in turn, is directly impacted by the content of fatty acids in milk (Wiking, 2004), which is directly correlated to the genetic merit and breed of the animal (Lindmark Månsson, 2003). A similar trend was observed in the results of this study (Chapter 3), where fat, protein and total solids content varied across different genetic groups throughout the season. In our study, two genetic types of HF cows, Elite and NA, were analysed, and the results were in line with past studies (Garrick and Lopez-Villalobos, 2000; Auldist et al. 2004; Bailey et al. 2005; Geary et al. 2010), showing that milk with higher

solids content yielded higher product outputs and returns. Milk composition attributes, if included with genetics and breeding programs, can be highly effective in increasing the milk solids content and product yields and reducing costs by reducing energy, processing and fuel costs, and, ultimately, yielding higher net returns for milk producers and processors.

6.5. Seasonality

The effect of seasonal variation and other factors on milk compositional profile has been extensively studied in the literature in the past (Ferlay et al., 2006; Stoop et al., 2009; Grimley et al., 2009; O'Callaghan et al., 2016). However, the most important parameters that affect milk composition are diet/feed and stage of lactation (Grimley et al., 2009; Sodini et al., 2004). The lactation period significantly affected milk composition, with late-lactation milk having higher fat and protein content as compared to mid-lactation (Sodini et al., 2004). The results presented in Chapter 2 align with those of Sodini et al., (2004), wherein the fat and protein contents were higher during the later phase of lactation, lowest in the spring period and highest in the autumn period.

The density of milk has previously been shown to be dependent on fat and solids-non-fat (SNF) content in milk. The results in Chapter 2 show the variation in milk density with season and compositional changes, where the density values were highest in the summer season (lowest fat content) and comparatively lower (1.0309 g/cm^3) in the autumn season (with higher fat content). The total solids content was also higher in the autumn period compared to the summer and spring periods, but there was no significant variation between the summer and spring periods. This is in line with other studies in the UK and Ireland where the total solids content decreased during the January to April and July to August periods (Geary et al., 2010). As stated earlier, milk yield and compositional characteristics are affected by the stage of lactation and diet. Milk density is dependent on milk fat and SNF content;

therefore, the variation in total solids content also impacts on milk density, increasing in the autumn season with increasing lactose and total solids contents of milk.

During the grazing season in Ireland cows graze outdoors and their diet is comprised mostly of fresh grass. Fatty acids form a significant component of milk fat and variation in fatty acid composition has been mainly attributed to the supply of fatty acids through diet and rumen microbial activity (Murthy et al., 2016). The main precursors of milk fat, i.e., acetic and butyric fatty acids derived from rumen fermentation, can be affected by diet through changes in rumen fermentation or the addition of fats for direct absorption and inclusion into milk fat (Ferlay et al., 2006). Oxidative losses in fatty acids due to the wilting and ensiling of grass have also been observed (O'Callaghan et al., 2016). This reduces the amount of fatty acids from fresh grass and, thus, causes fluctuations in the fatty acid composition of milk, affecting the total fat content and milk density. Therefore, a combination of these factors and seasonal variation impacted the feed quality for grazing cows, which in turn affects milk composition and milk density, respectively.

6.6. Use of different analytical methods and impact on milk density

Milk density measured for the samples in our studies at different temperatures was impacted by the use of different measuring methods. Referring to the results of milk density for both DMA35 and DMA4500 at the measured temperature in Chapter 4, both systems showed a very similar trend, although there were differences in the absolute numbers, with the DMA35 showing a consistently lower density than the DMA4500.

The DMA35 is used regularly in the dairy industry for rapid measurement for milk density (personal communications from industry personnel) and measures the density based on oscillating U-tube technology. The frequency of the oscillator changes due to introduction of liquids, and this variation in natural frequency of the oscillator enables density measurement (Paar, 2009). The effect of instrument was assessed calculating the density of milk at different

temperatures after adjusting for any variations introduced due to sampling period, instrument, and the random effects of cow. Other factors that may also affect the density of samples using different equipment include temperature history of the sample, which introduces small variations in density, Recknagel's phenomenon, which refers to the increased density of stored cold milk, and the level of trapped air. The amount of entrapped air in fresh milk could be as high as 6%, and this entrapped air may influence the milk density measurement and lead to errors in measuring results and poor repeatability (Hyfoma, 2019). Past research has shown that entrapped air does not significantly impact milk density directly but needs to be removed to improve measurement accuracy (Bouvier et al., 2013; Sharp & Hart, 1936). For this study, air bubbles on the oscillating tube were visible on the display screen of DMA4500 during density measurement and can be removed by pushing in more sample using a 2-ml syringe. This, therefore, enables more accurate measurement of density without any air-induced errors.

Several researchers have determined the controlled temperature history necessary for high precision and accurate density measurement (Sharp & Hart, 1936; Vanstone & Dougall 1960; Hilker & Caldwell, 1961). However, for this study, the temperature history did not affect the results because all the samples were subjected to the same procedure and temperature history (equilibrated at each temperature for the same time duration).

6.7. Milk density, payment systems and temperature

Past research indicated that the milk procured from dairy farmers be paid on a multi-component pricing system (MCP) (Shalloo, Dillon & Wallace, 2008). MCP referred to putting a value on the mass (kg) of protein (A) added to the value of mass (kg) of fat (B) in the system and subtracting the cost of collection and processing (C) related to volume of milk supplied. This was named as the A+B-C system, which has been used in many countries around the world (e.g., Denmark, Australia, Holland, New Zealand etc.), including Ireland, for

approximately 10 years.

Milk is collected from the farm at a temperature of ~ 4-5°C and, under the current system, the payment system followed by the processor quantifies the amount of fat and protein using milk volume in litres, milk fat and protein concentration, multiplied by a density factor of 1.0297 g/cm^3 for the weight-volume relationship. It has been noted that as the temperature of milk is increased, the density of milk reduces. A majority of changes in milk density can be attributed to the changes in milk fat concentration (Paar, 2009). The density factor is used to convert the volume of milk from litres to weight (kilos) by multiplying the volume of milk by the density factor, i.e., 1 L of milk at density factor 1.0297 g/cm^3 weighs 1.0297 kg. The density factor is also used when calculating the amount of fat and protein in milk by multiplying the volume of milk in litres to estimate the weight of milk and multiplying by the concentrations of fat and protein (%) in milk, which generates the mass of fat and protein in milk, respectively. In our study, milk density was measured at four different temperatures in Chapter 4 and it was shown that milk density varies at different temperatures (reducing with increasing temperature), recorded highest density at 5°C and lowest value of density measured at 20°C and significantly impacts the weight-volume calculations.

Past method of measuring milk solids content utilized the density as shown by Fleischmann's formula (Ullmann et al.,1985):

$$\mathrm{TS} = 1.2 * \mathrm{F} + 266.5 * \frac{(\mathrm{S}-1)}{\mathrm{S}}$$

where TS is total milk solids, F is the fat content in milk (both in %) and S is the density

The above formula shows the importance of milk density and thus implies that total milk solids content estimated at a lower temperature (5 °C) will be higher than total milk solids estimated at higher temperatures of approx. 20 °C. From our study (chapter 4), the density factor of 1.0334 g/cm^3 may be used for volume-weight conversion, i.e., 1 L of milk with the new factor will weigh 1.0334 kg. This is significantly different from using the current value of

1.0297 g/cm^3 being used in the mass balance. Using the new density factor of 1.0334 g/cm^3 may enable a more precise estimation of fat and protein quantity in milk. An example of the use of the density conversion at the same temperature (5°C) in weight volume relationships is shown in Table 4.4. For total milk produced in Ireland in the year 2018, a significant difference in mass estimation of individual constituents (1.16 million kg in fat and 1.011 million kg in protein) is observed between the use of historical factor, 1.0297 g/cm^3, and the new factor, 1.0334 g/cm^3. While saying this, it is important to note that, while there may have been more kilograms of fat and protein in the milk (at milk density 1.0334 g/cm^3) than at the conversion factor of 1.0297 g/cm^3, in reality, this does not mean that there will be more money to pay out in milk price, but will mean that allocation of payment is aligned with an increased levels of milk solids.

The model developed may enable farmers to estimate density changes based on changes in temperature of milk samples, and the density factor thus estimated can help in measuring the total solids content in milk. The volume of milk produced and supplied from Irish dairy farms has significantly increased since the removal of EU milk quotas, and this research aligns with the current trend, enabling accurate measurement of milk solids and directly impacting the profitability of both dairy farmers and processing industries. The results of this study can be effectively utilised by processors during weight-volume calculations to accurately record the amount of total solids incoming at the plant gates and also monitor and control the milk constituents' conversion process with better efficiency.

The density of milk is dependent upon seasonal variations observed in milk composition throughout the year. This is evident from the results in Chapter 2, with density varying significantly with changes in the constituent's contents of the milk. Monthly and season-based density factors were determined, which are relevant for milk-processing planning. Variations in the composition and ultimately density could be attributed to various

factors, such as the stage of lactation, climatic conditions (including microclimatic pattern), the feeding pattern during the period of study, housing conditions in autumn and winter seasons, the genetic group, and temperature, amongst other parameters. Seasonal and annual factors for density conversion used in weight–volume relationships were determined, with an emphasis on usage of a periodic, rather than an average, conversion factor evident from the strength of linear regression models. Secondly, the impact of genetic traits and breed on milk density were also estimated. A comparison of density values for milk samples obtained from Jersey and two strains of Holstein-Friesian cows was obtained. Thirdly, the relationship between milk density and temperature was determined. It was observed that the intake temperature of milk on farm significantly affects whole milk density, along with other external factors such as composition and processing conditions. There is an inverse relationship between temperature and density, i.e., density of milk decreases with increasing temperature, and there is also a quadratic effect of temperature on milk density. To the best of our knowledge, this analysis was the first of its kind for the Irish dairy sector and generated a new density conversion factor to be used at 5ºC, for example, in the A+B-C milk payment system currently followed in the Irish dairy sector.

Milk density is an important factor in milk processing to estimate the individual milk constituents (weight–volume calculations). The constituent contents thus calculated significantly influence the product portfolio, in conjunction with operating capacities and market demand. Weight-volume calculations have been identified as an issue for dairy processors in Ireland. Data analysis based on a mass balance approach helped to identify loss points in the process and also enabled processors to reconcile their test results. From an economic perspective, this study identified unaccounted-for losses in the process and helped monitor the overall financial performance. The model was developed to predict the net value of milk or returns based on different scenarios. The model may be a useful tool in determining the 'best-

possible' scenario by evaluating all the possible variables like density factor, breed, and processing variations when scenario planning for processing. This model was developed for milk fat processing, i.e., the fat conversion process at a single point of time, with all milk intake being directed to butter manufacture. In reality, milk processing in a dairy environment involves different product mixes, dependent upon various parameters such as supply and demand profile, compositional variances, capacity constraints, estimating actual costs and returns. If all these parameters, i.e., temperature, breed, and seasonal variation in composition, along with a season-based density factor, are incorporated and analysed, it may enable the model to be more adaptive and precise decision-support to the changing dynamics of the dairy industry throughout the year.

6.7. Proposals for further research

The following suggested studies would provide further understanding of the role of milk density in the broader financial and operational efficiency analysis in a dairy processing environment:

- Including new density factors in estimation of weight-volume conversion at dairy processing sites. The present research and interactions with industry professionals highlighted the use of a single density factor as a point of concern and resulted in under-estimation of milk constituents. Use of season-based density factors could be useful in accurate estimation of milk constituents and therefore, useful in operational planning in a dairy processing environment;
- Developing a holistic model comprising of various elements like seasonality, animal breed product mixes, dependent upon various parameters such as supply and demand profile, compositional variances, capacity constraints, estimating actual costs and returns, in conjunction with. This may enable the model to be more adaptive and a

precise decision support tool to be developed that is responsive to the changing dynamics of the dairy industry throughout the year;

- The use of density parameter was noted in all the by-product streams like skim milk and buttermilk in the processing environment. As is the case of milk density, there is currently a single density factor being used year-round for each of the by-product streams. As observed in the present thesis, development of new density factors for the skim milk and buttermilk processing processes will enable further reduction in losses observed in these processes and may improve the overall dairy processing efficiency and subsequently, financial performance.

6.8. References

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Appendix

Conference Contributions

Evaluation of butter manufacture process – Mass balance approach to assess fat conversion process in a dairy processing unit

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Abstract

The dairy industry is one of Ireland's most important indigenous industries and comprises a vital part of the agri-food sector. Dietary preferences of consumers and scientific research including product formulations have brought milk fat into limelight, whereby demand and butter prices are increasing across dairy markets around the world, pushing further research into this domain. The present study analysed the fat conversion process in a commercial dairy processing unit. The objective of the study was to identify and account losses within butter manufacture process using a mass balance approach.

Representative samples were collected from a local dairy industry in Ireland. Milkoscan (Foss Instruments) was used to estimate fat content of the samples at site. Rose Gottlieb was used to test fat content in the lab at Teagasc, Moorepark. The volumes and quantities of milk and product entering and leaving the process were quantified for mass balance studies. Fat content of samples at site was 4.033% obtained through rapid testing. Rose-Gottlieb conducted in lab obtained fat results of 3.992% (tankers) and 4.032% (silos). Fat results (rapid) of cream and skim at site were 48.58% and 0.07% whereas lab (standard) results were estimated at 47.64% and 0.039% respectively. Mass balance is fundamental to regulate processing especially product yields and outputs and is represented as

$$Fat\ intake = Fat\ in\ products + losses + recycled\ fat^{*}$$

(* – fat returned to separation from butter churn melting and steamed silos)

Fat intake was approx. 22050 kilos for processing into butter. Fat content in butter (end product) yield was 21175 kilos. Mass balance applied on fat results obtained a loss of 0.44% (~100 kilos) for tankers and 1.414% (~300 kilos) for silos against industry assumed losses of 2.0-2.5% (~450-570 kilos).

Variation in results obtained at site and lab could be attributed to the changes in milk composition on different farms and also milk collected from different herds. Rapid testing methodologies impacted the fat results due to calibration or manual errors. Load cell calibrations at silo level induced inaccuracy in fat estimation. Milk density fluctuates with season and an average conversion factor 1.035 might not be representative of milk

composition throughout the year. Mass balance determined the losses, recognised loss points, and estimated a lower fat loss percentage than assumed by the industry whilst identifying the reasons accountable for higher values. This study had direct financial impact in determining the profitability of the site. A study to analyse seasonal variation in density and milk composition is proposed as a result of this work.

Key words: Milk fat, mass balance, conversion factor

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Effect of temperature variation on raw whole milk density and its impact on milk payment system for Irish dairy Industry

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Abstract:

The objective of this study was to determine the effect in whole milk density due to variations in temperature. Whole milk samples were collected from morning milking of 32 individual dairy cows of national average genetic merit once every two weeks over a period of 6 weeks from the Teagasc research farm, in Kilworth, Co. Cork, Ireland. A total of 93 samples were assessed on the rapid testing technique – Dairyspec FT manual system (Make- Bentley) for milk compositional analysis. Density of milk was evaluated using two methods - a portable density meter DMA 35 and desktop version DMA 4500M. Statistical analysis using analysis of variance (ANOVA) showed a significant difference in means of densities (F, 78.866> F-crit., 3.947 and $p<0.01$) measured at different temperatures. The results were then analysed using PROC GLM procedure, SAS software to develop a quadratic model and identify the relationship (linear or curved) between temperature and density. The output indicated a significant non-linear relationship ($p=0.0008$) with the model equation defining the curvature and density-temperature relationship ($r^2 = 0.659$) as

$$Density = 1.033 + 0.0000632 * temp - 0.0000114 * temp^2$$

In general, there was an inverse correlation between whole milk density and temperature (i.e. as temperature increased, milk density decreased). Mean density calculated

at 5 °C was 1.03319 g/cm^3 with corresponding figures of 1.03277, 1.03148 and 1.02994 g/cm^3 at 10, 15 and 20 °C respectively. This implies that the volume of milk and subsequent total milk solids content estimated at lower temperature (5 °C) will be higher than the values estimated at a higher temperature (20 °C)

Keywords: raw milk, whole milk, density, temperature, payment

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Effect of seasonal and composition variation on whole milk density and determining season-based density factors for Irish Dairy

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Abstract

The objective of this study was to determine the effect of seasonal variation on milk composition and establish an equation to predict density based on milk composition, which could enable the calculation of real-time, season-based density conversion calculations. Raw whole milk samples were collected from the morning and evening milking of ~60 individual cows of three different genetic merits i.e. Jersey, Elite and National Average, once every two weeks for a period of 9 months (March-November, 2018) from the Teagasc research farm, in Kilworth, Co. Cork, Ireland. A total of approx. 1035 samples were assessed for fat, protein and lactose content on the rapid testing technique – Dairyspec FT manual system (Make- Bentley). Density of the milk was evaluated using three methods - a portable density meter DMA 35, a standard desktop version DMA 4500M and AOAC method using 100 cc glass bottles. Statistical analysis of the data using PROC Mixed for each season presented a significant difference in density of milk samples ($p < 0.0001$) across stage of lactation (days in milk),

season and compositional variations corrected for the effects of parity, animal breed and measurement technique. The means and standard deviations of milk composition were 4.72±1.18% fat, 3.85±0.45% protein and 4.69±0.25% lactose. Season, days in milk, and percentages of fat, protein and lactose were predictors of milk density. Peak density values were observed at the beginning and end of lactation (1.0309 g/cm^3 for March, 1.0313 g/cm^3 for October, 1.0316 g/cm^3 for November). The density values estimated for each season i.e. spring, summer and autumn were 1.0304 g/cm^3, 1.0315 g/cm^3 and 1.0309 g/cm^3 respectively.
Keywords: seasonal variation, raw milk, whole milk, composition, milk density, conversion factor

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ORIGINAL

RESEARCH Development and evaluation of a processing sector model for butter manufacture using a mass balance technique at two dairy processing sites

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The butter manufacturing process at two different commercial dairy processing sites in Ireland was evaluated using a mass balance approach to develop, evaluate and validate a processing sector model of the flow of milk fat from intake to final product. The mass balance was represented as a function of fat intake = fat in products + fat losses + recycled fat. *Representative samples of all products, namely whole milk, cream, skim milk, butter, buttermilk and cleaning-in-place streams (cream silo flush, butter churn residue and sludge), were collected from two different sites. Milk fat levels and product quantities were measured to obtain the fat outputs. Total fat losses at the end of butter production ranged between 1.90% and 2.25% of the total fat input for both sites. Three dif- ferent scenarios were examined to evaluate the model: S1 (Animal Breed) high genetic merit (Elite) and national average (NA) Holstein Friesian (HF) cows were evaluated, for their effect on the net value of milk; S2 (Product Portfolio) a mixed product portfolio of cheese, butter and skim milk powder (SMP) was compared to a product portfolio comprised of butter alone; and S3 (Process Efficiency) the impact of varying process losses on net values of milk and the quantities of products produced was simulated. The value per 1000 L of milk for S1 was €410.69 and €393.20 for Elite and NA cow's milk, respectively. For S2, the butter-only product portfolio returned €355.10, whereas the mixed-products portfolio returned €369.60. Lastly, S3 corresponding returns for 1%, 2.2% and 5% losses was €365.90, €361.47 and €351.12, respectively.*

Keywords Dairy, Milk fat, Mass balance, Processing, Model.

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INTRODUCTION

Milk and dairy products are major constituents of Western diets, and there has been an ever-grow- ing demand for high-quality dairy products in these regions (Heck *et al.* 2009). Various factors affect the demand and supply of dairy products, including product formulations, variations in milk supply, seasonality, consumer perceptions and fluctuations in customer demand (Chen *et al.* 2014). Seasonal

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variations in milk composition pose a significant challenge to the processing industry and the ultimate product mix and quality

of products (Auldist *et al*. 1998; Lindmark-Månsson *et al*. 2003). With the removal of European Union (EU) milk quotas in 2015, there has been a consequent increase in production across the EU. For example, in the Irish dairy industry, milk production has increased from ~under 5 billion litres at the end of 2009 to 7.57 billion litres at the end of 2018 (Central Statistics Office (CSO), 2018). This has occurred at a time where there is significant price volatility, which presents a challenge to the dairy industry.

Demand variations govern the price of dairy products, and a small change in demand can

have a significant impact on prices (Vitaliano, 2016; Stephenson and Nicholson, 2018). Such uncertain scenarios are manifested by steep changes in the prices of dairy prod- ucts, especially butter prices, which have almost doubled from 2015 to 2018 (Global Dairy Trade (GDT), 2019a; CLAL, 2019a). One of the reasons behind the increasing prices of butter could be the 're-profiling' of butter as a healthy food product. Exports for butter from the EU region have increased significantly (7.4% export growth in first quarter of 2019) (Irish Farmers Association (IFA), 2019). The increase in production has been attributed to growing demand from developing countries, while consumption from Western countries has been stagnant or dropped slightly (Vitaliano, 2016; Kiernan, 2019). Demand for dairy proteins and powders such as SMP has increased considerably, with the SMP price index rising by 3.2% in 2018 (O'Brien, 2019), while SMP production from the Irish dairy sector alone has also increased considerably from ~120 000 tons at the end of 2017 to ~134 000 tons in 2018 (CLAL, 2019b). These changes pose a significant challenge for processors, and processors who are responsive to this variability in the market will attain a higher rate of return and thus be more economically sustainable (Geary *et al*. 2010).

Various processing models have been developed and stud- ied around the world using a mass balance approach. Mass balance approaches have been in practice for a long time and have been applied across diverse fields such as climate studies (Medwedeff and Roe, 2017), environmental monitor- ing (Ashfaq *et al*. 2017; Irvine *et al*. 2017), chemical analy- sis (Little *et al*. 2014; Chen *et al*. 2015), engineering (Fahrenfeld *et al*. 2014) and energy balance analysis (Brock *et al*. 2000). The mass balance approach is central to the evaluation of processing efficiency as regard to yields of products and waste.

In dairy processing, mass balances have been imple- mented across diverse segments ranging from the estimation of milk constituents such as fat protein and lactose (Bang- stra *et al*. 1988; Garrick and Lopez-Villalobos, 2000; Bailey *et al*. 2005; Geary *et al*. 2010; Sneddon *et al*. 2016), and comparing process-based models for nitrogen, phosphorus and greenhouse gases impact developing models associated with associated with milk production at the animal, field and farm-scale (Spears *et al*. 2003; Veltman *et al*. 2017). The objective of this study was to develop, evaluate and validate a mass balance model for the milk fat conversion process and to apply the model across two dairy processing sites in Ireland.

MATERIALS AND METHODS

Mass balance approach

The principle of a mass balance is based on the law of con- servation of mass. The mass balance equation (Warn and Brew, 1980) is represented as follows:

Massin ¼ Massout þ Massstored

or

Rawmaterials ¼ Products þ Wastes þ StoredMaterials For

a butter manufacture process, it may be stated as

Fatintake ¼ Fatinproducts þ Fatlosses þ Recycledfat

þ Excessfatsold

where Fat intake = fat content of the total milk volume pro- cessed (kg); Fat in products = fat in each of the products produced (kg); Fat losses = fat lost during processing (kg); Recycled fat = fat collected from cleaning-in-place (CIP) activities such as cream silo flush and churn residue flush and sent into separation again (kg); Excess fat sold = any fat not used in the production of products sold to internal/ external customers (kg).

A protocol was shared with all the participating sites to organise the mass balance exercise. The exercise of follow- ing fat conversion within the dairy processing environment was monitored as a batch with one or two silos of whole milk being processed to butter in a closed-loop procedure. The process was divided over a period of 2 days, with the first day being dedicated to gathering samples and data for raw milk and the separation process while the second day was dedicated to the butter manufacture process, with sam- ples collected for butter, buttermilk and CIP streams. Raw milk arrived at plant sites in bulk tankers and samples was collected off the back of the tanker to test for antibiotics. Once the sample passed the antibiotic test, it was transferred to designated silos for the mass balance study. Composite representative samples of milk were taken as the silos were emptied for separation process. During the separation pro- cess, representative skim milk samples were collected once every hour.

For Day 2, representative samples of cream stored over- night (continuously agitated to avoid fat separation) were collected as the cream was emptied for butter manufacture. As the butter manufacture process continued, buttermilk produced was collected in an assigned silo to measure the total volume produced. Once the butter manufacture process was completed, the total weight of butter and volume of buttermilk produced was measured. A CIP process was initi- ated at the end of butter manufacture with CIP being com- pleted in the cream silo, packing line and butter churn. Cleaning-in-place flows from these points were collected in individual intermediate bulk containers (IBC) and weighed to determine the volume generated. Three samples of each of the CIP streams were collected for testing. After sample collection, the samples were tested for fat content using Rose Gottlieb Method.

The fat mass balance was calculated by multiplying the volume of incoming milk from intake by the density

conversion factor to obtain weight of milk. The weight of incoming milk was then multiplied by the fat content in whole milk to attain the fat available for conversion to but- ter and other products. Similarly, the cream and skim vol- umes produced were multiplied by the fat contents to obtain the fat mass in cream and skim available. This exercise was completed at all the stages within the fat processing value chain for other products, that is, buttermilk, butter, CIP streams – cream silo flush, butter churn residue and final sludge by multiplying the fat content with weight of each product produced. The difference of fat from intake to end of separation and butter manufacture process was calculated as follows:

Lossatseparation ¼ Fatintake — ðFatincream þ Fatinskimmilk

Lossatbutterproduction ¼ Fatincream — ðFatinbutter þ Fatinbuttermilkþ

Model description

All inputs, outputs and losses within the dairy processing steps were accounted for in the mass balance model. The model is a mathematical representation of the conversion of milk fat into the butter. The model inputs included the vol- umes and composition of milk intake and product portfolio, that is, butter, buttermilk, skim milk and CIP discharges and their composition, all of which were used in the mass bal- ance calculations. The final quantities of each of the prod-

ucts in the portfolio were also noted and used as inputs in the model. Costs of milk processing, along with costs of collection and standardisation, were estimated to determine

the economics of the mass balance model. The availability of actual cost estimates was a challenge in developing and refining this model. However, lack of the real-time costs information was overcome by using data from past studies (Quinlan *et al*. 2006; Breen *et al*. 2007; Geary *et al*. 2010), control reports and rigorous consultation with dairy industry professionals. Different scenarios were examined, and their impact on net returns was estimated. The schematic diagram of the dairy processing sector model for fat conversion is shown in Figure 1. Site 1 and Site 2 were located in differ- ent regions of Ireland with differences in their milk supply profile, processing capacities, demand and customer require- ments, plant set-up (number of silos studied, number of sep- arators, CIP practices), period of analysis and management practices at the plants.

Financial components

Market returns

The market values for calculating returns from the mass bal- ance model were taken from the Global Dairy Trade (GDT) website (https://www.globaldairytrade.info/) (GDT, 2019a) and were representative of a 2-year average for 2017–2018. The market price obtained from the 2-year average for butter was ~USD 5027/T. Average product prices for other products for the 2-year period are shown in Table 3.

Processing cost

Processing costs, including volume-related costs associated with collection, standardisation and processing of milk, were gathered from a study on Irish dairy processing cost analysis (Breen *et al*. 2007) and using different indices such as industrial price index, wholesale price index and information the Central Statistics Office of Ireland (CSO, 2018), along with consultation with dairy industry professionals and experts. Processing costs also included product-related costs as associated with processing, packaging, transportation and storage and marketing costs, all adjusted for 2018 levels þ (Quinlan *et al*. 2006; Geary *et al*. 2010; Heinschink *et al*. 2012). The processing costs for manufacture of butter and other products are summarised in Table 1.

Milk price

Marginal rate of technical substitution was used to deter- mine the value of milk fat and protein per kg. It is described as the amount by which one input can be reduced when one additional unit of another input is used so that the overall outcome remains constant. In this case, one unit of protein can be reduced to add one extra unit of fat to keep the over- all milk value constant. It is represented mathematically as follows:

$$\mathrm{MRTS}(x1, x2) = \frac{\Delta x2}{\Delta x1} = \frac{-\mathrm{MP1}}{\mathrm{MP2}}$$

where MP1 and MP2 are the marginal products of input 1 and input 2, respectively. For every additional kg of input, fat or protein, the overall revenue from milk will be increased depending upon product portfolio, processing costs and market value.

Value of milk

The net value of milk may be calculated by subtracting the costs of converting milk to butter from the total volume of butter produced and the market value of butter obtained.

$$\text{Net value of milk} = \sum(v \times p) - c$$

where v is the volume of butter produced, p is the market price of butter and c is the costs of processing milk to but- ter.

Model evaluation

Scenario analysis

Three different scenarios (S1, S2 and S3) were explored for variations in milk values depending on (i) two genetic groups representative of high genetic merit (Elite) and national average (NA) Holstein Friesian (HF) cows; (ii) a plant producing only butter as a product compared to a

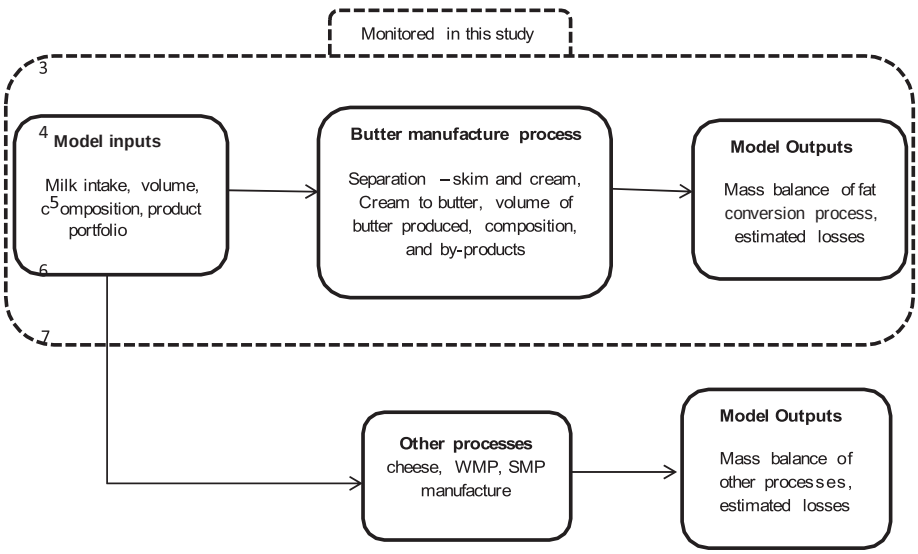


Figure 1 Schematic representation of the dairy processing sector model for butter manufacture. (SMP, skim milk powder; WMP, whole milk powder).

Table 1 Processing costs including volume-related and product-related costs for butter adjusted up to 2018 levels (Quinlan *et al.*, 2006; Breen *et al.*, 2007)

Cost	*Butter*	*Cheese*	*WMP*	*SMP*	*BMP*
Volume costs, €/L					
Collection[a]	0.0126	0.0126	0.0126	0.0126	0.0126
Standardisation	0.0051	0.0051	0.0051	0.0051	0.0051
Processing[b]	0.0042	0.0128	0.0101	0.0128	0.0101
Product costs, €/MT					
Processing[b]	99.89	127.44	178.34	175.68	178.34
Packaging[b]	31.78	41.45	41.45	41.45	41.45
Storage[c]	75.10	44.06	28.66	7.96	28.66
Distribution[b]	73.00	73.00	73.00	73.00	73.00
Marketing	50.70	50.70	50.70	50.70	50.70

BMP, buttermilk powder; SMP, skim milk powder; WMP, whole milk powder.

[a]Quinlan *et al*. (2006).

[b]Breen *et al*. (2007).

[c]Consultation with dairy professionals and expert.

product mix of cheese (37.6%), skim milk powder (SMP) (22.5%) and butter (39.9%); and (iii) an increase or decrease in processing efficiencies at the plants. The product portfolio details were derived from Central Statistics Office, Ireland website (CSO, 2018) and included mainly cheese, butter and SMP. The percentages for cheese, butter, SMP and whole milk powder (WMP) were calculated as a proportion of the cumulative tonnes that were produced.

Scenario 1 (S1): The first scenario used the model to evaluate net value of milk and the quantities of product produced from 1000 L milk from Elite and NA HF cows assessed, with 39.9% of milk intake used in the production of butter, 37.6% into cheese and 22.5% into SMP (Central Statistics Office (CSO), 2018). The by-products of this portfolio are Whey and buttermilk powder (BMP).

Scenario 2 (S2): In the second scenario, a product portfolio comprised of cheese, butter and SMP was investigated with 39.9% of milk intake used in the production of butter, 37.6% into cheese and 22.5% into SMP as compared to if only butter as an end product (100% milk allocated for butter manufacture) was manufactured. Skim milk powder and BMP are by-products of this product portfolio.

Scenario 3 (S3): In the last scenario, the impact of varying process efficiencies (increasing or decreasing losses) on

net values of milk and the quantities of products produced was examined. The composition of products simulated in the model is shown in Table 2.

RESULTS

Table 1 presents the processing cost, including volume-related and product-related costs, for a product mix, that is, butter, BMP, SMP, cheese and WMP, taken from literature (Quinlan *et al.* 2006; Breen *et al.* 2007) and adjusted up to 2018 levels using inflation and price indices. The average processing costs including volume-related, and product costs such as marketing, packaging, storage and distribution were ~0.04 c/L. Table 2 shows the composition of different products simulated using the model for estimation of net value of milk. The milk constituent content for each product in the product mix was in line with Codex Standards; that is, fat in butter was taken as 82%, while the fat level was 35% in cheese. Table 3 shows the average market value of the products simulated using the model, with average butter price for years 2017–2018 at ~5027 USD/MT (metric ton), BMP ~USD 2170/MT, SMP priced at @USD 2000/MT and cheese price of USD ~3720/MT. Table 4 shows the net value of milk obtained from the model for the different scenarios simulated, and the results of the scenario analysis have been explained in later section.

Site results

A total of ~521 MT of milk was processed at site 1, with an average fat content of 4.37%. Fat mass available for butter manufacture at site 1 was 22.76 MT (weight of milk * % fat). The milk collected was passed through a separation process, producing a total of 56.2 MT of cream (38.77% fat) and ~468.57 MT skim milk with 0.10% fat (Table 5). Fat mass available in cream at site 1 was calculated as 21.79 MT, and fat mass from skim milk was 0.47 MT. From the available fat mass in cream, butter was manufactured, with butter mass produced at site 1 ~26.83 MT at 80.50% fat (butterfat = 21.59 MT) (Table 5). Similarly, the weight of buttermilk produced as a by-product of the process was also noted (28.25 MT buttermilk at 0.66% fat = 0.19 MT fat mass). The CIP process inputs, that is, churn residue melt and cream silo flush, were collected and weighed in an IBC to complete the mass balance approach (i.e. fat intake = fat in products + losses + recycled fat). Churn residue and silo steamed flush (steaming out the cream residue from cream silo) at site 1 were measured at 1.50 and 1.90 MT with fat content of 38.19% and 17.47%, respectively (Table 5).

The loss values from butter manufacture process, when divided by the incoming fat content (weight) from cream, gave the percentage loss experienced during butter manufacture. Therefore, total fat mass obtained in butter and buttermilk, when subtracted from fat available for butter manufacture process (from cream), gave an estimate of the losses in the butter manufacture process (0.16 MT) at site 1. The calculation of total loss percentage formed the final step of the mass balance with cumulative losses from separation and butter manufacture process was 0.43 MT for site 1. The cumulative loss values (1.93%), when divided by the total incoming fat from milk, gave the total losses at the end of butter manufacture process (Table 6).

For site 2, ~555 MT of milk was collected and processed at site 2 with a fat content of 4.03% (fat mass ~ 22.16 MT). There was 46 MT fat in cream, with 512.58 MT of skim milk (0.04% fat) at site 2 (Table 5). The fat mass obtained in cream from site 2 was 21.92 MT and the difference (in-coming fat − fat in cream + fat in skim) when divided by the total incoming fat in milk gave the fat loss % at separation stage = 2.21% (Table 6). Amount of butter produced was ~25.78 MT at 82.15% fat, while butterfat mass was 21.17 MT for site 2 (Table 5). Processing at site 2 produced 21.09 MT buttermilk with 0.72% fat (0.15 MT fat mass). Site 2 churn residues and silo steamed cream were 0.77 and 0.35 MT with 36.66% and 41.87% fat content, respectively. Total fat mass obtained in butter and buttermilk when subtracted from fat available for butter manufacture process (from cream) gave the estimate of loss at butter manufacture process. Thus, the total loss for butter manufacture was 0.01 MT at site 2; the cumulative losses from separation and butter manufacture process were 0.51 MT at site 2 (2.25% of the total fat mass in the system).

Table 2 Composition of different products simulated using the model of Geary *et al.* (2010) for estimation of net value of milk

Item	*Cheese*	*Butter*	*WMP*	*SMP*	*Whey*	*BMP*
Fat, %	35.00	82.00	26.50	1.00	1.00	8.30
Protein, %	24.50	0.59	25.10	33.00	15.15	41.72
Lactose, %	1.39	0.79	39.80	54.00	77.15	40.32
Minerals, %	2.15	0.12	5.90	8.00	4.32	4.66
Water, %	35.26	16.50	2.70	4.00	2.38	5.00

BMP, buttermilk powder; SMP, skim milk powder; WMP, whole milk powder.

Scenario analysis

S1: 1000 L of milk from Elite HF and NA HF cows yielded 48.20 and 46.60 kg cheese, 52.40 kg SMP for both genetic groups; 36.50 and 33.80 kg butter, 23.60 and 23.10 kg of whey; and 4.40 and 3.90 kg BMP, respectively. The net value of milk after deducting processing costs of milk was € 410.69 and 393.20 for Elite and NA HF cows, respectively. These values might seem a bit higher than expected since the estimated value of milk here does not include a margin.

Table 3 Average butter and other product prices (USD/MT[a]) for the year 2017 and 2018 obtained from the global dairy trade website (GDT, 2019b)

	Butter		*BMP*		*SMP*		*WMP*		*Cheese*	
Period	*2017*	*2018*	*2017*	*2018*	*2017*	*2018*	*2017*	*2018*	*2017*	*2018*
January	4345.0	4699.0	2726.5	1866.0	2636.0	1758.5	3288.5	2948.0	3917.0	3401.5
February	4595.5	5305.5	2172.0	2039.0	2591.0	1882.0	3251.5	3236.0	3694.0	3712.5
March	4781.5	5280.5	1846.0	1959.0	2033.0	1969.0	2818.5	3229.0	3420.5	3684.0
April	4821.5	5574.0	1654.0	1988.0	1978.5	1881.0	2961.0	3294.5	3375.0	3767.0
May	5195.0	5717.0	1919.5	1990.0	1990.0	2023.0	3272.5	3228.5	3696.0	4114.5
June	5699.5	5596.0	2092.0	2314.0	2187.0	2027.0	3082.5	3197.0	4203.0	3922.5
July	5889.5	5171.5	2264.0	2352.5	2057.0	1936.0	3112.5	2939.0	4081.5	3654.5
August	5741.0	4597.0	2198.0	2441.0	1967.0	1961.5	3149.0	2920.5	3968.5	3573.5
September	5990.0	4270.5	2026.0	2474.0	1932.0	1992.5	3111.0	2794.5	4075.0	3567.0
October	5786.5	4065.0	1804.0	2522.0	1846.0	1979.5	3025.5	2741.0	4108.0	3436.0
November	5330.0	3841.0	1931.0	2568.0	1759.5	1981.0	2815.0	2627.0	3916.0	3251.0
December	4524.5	3836.5	1957.0	2973.0	1724.5	2006.0	2792.5	2670.5	3542.5	3223.5
Avg	5225.0	4829.5	2049.2	2290.5	2058.5	1949.8	3056.7	2985.5	3833.1	3609.0
	5027.2		2169.9		2004.1		3021.1		3721.0	

BMP, buttermilk powder; SMP, skim milk powder; WMP, whole milk powder.

[a]MT, Metric ton: A unit of weight equal to 1000 kg.

Table 4 Quantities of products produced, and net value of milk after subtracting processing costs for each of the scenarios[a] simulated from the model

Product mix (kg/1000 L milk)								
Desc.		*Cheese*	*SMP*	*Butter*	*Whey*	*BMP*	*Collection and processing value/L, €*	*Net milk value (€/1000 L)*
S1	Elite[a]	48.20	52.40	36.50	23.60	4.40	−0.04	410.69
NA[a]		46.60	52.40	33.80	23.10	3.90	−0.04	393.20
S2	All products	44.40	50.20	31.50	21.70	3.50	−0.04	369.60
Only butter		–	80.30	50.80	–	5.60	−0.04	355.10
S3	1% loss	44.00	49.60	31.20	21.50	3.40	−0.04	365.90
Loss	2.2% loss	43.50	49.00	30.80	21.30	3.40	−0.04	361.47
5% loss		42.20	47.60	30.00	20.60	3.30	−0.04	351.12

BMP, buttermilk powder; SMP, skim milk powder.

[a]Scenario 1 (S1) evaluated milk from high genetic merit (Elite) and national average (NA) Holstein Friesian cows. Scenario 2 (S2) evaluated milk processed into two product portfolios, one with milk used for butter (39.9%), cheese (37.6%) and SMP (22.5%), and other with milk used for butter (100%). Scenario 3 (S3) evaluated different process efficiencies.

S2: 1000 L of milk yielded 44.40 kg cheese, 50.20 kg SMP, 31.50 kg butter, 21.70 kg whey and 3.50 kg BMP for a plant producing the complete product mix, with net value of milk being €369.60. For a plant producing only butter, the quantities of products produced were 80.30 kg SMP, 50.80 kg butter and 5.60 kg BMP, while the net value of milk was € 355.10. The values estimated under scenario 2 also follow trend from scenario 1 and do not include a mar- gin, thus seeming relatively higher.

S3: In the final scenario, losses occurring in the process- ing of products were assessed at three different levels, that is, 1%, 2.2% (as per the mass balance studies discussed in this paper) and a higher level of loss at 5% of the total milk processed (1000 L). The product portfolio included all prod- ucts simulated, that is cheese, butter, SMP, whey and BMP. As expected, the lower the losses, the higher the net value of milk generated. Losses at 1% in processing corresponded to 44.00 kg cheese, 49.60 kg SMP and 31.20 kg butter pro- duced with whey by-product and BMP. The net value of milk for 1% loss was the highest of the three % losses sim- ulated, at € 365.90/1000 L of milk. Losses in processing (as evaluated from mass balance exercise in this study ~2.2%)

Table 5 Fat percentage, quantity of each product produced and fat mass in each product at each sub-stage of fat conversion process at the sites monitored

		Site 1			Site 2		
Stage	*Product*	*Fat (%)*	*Qty prod. (MT)*	*Fat at each stage (MT)*	*Fat (%)*	*Qty prod. (MT)*	*Fat at each stage (MT)*
Intake	Milk	4.37	520.88	22.76	4.03	555.21	22.39
Separation	Cream	38.77	56.20	21.79	47.64	46.00	21.92
	Skim	0.10	468.57	0.47	0.04	512.58	0.20
Butter process	Butter	80.50	26.83	21.59	82.15	25.78	21.17
	BM	0.66	28.25	0.19	0.72	21.09	0.15
CIP	CHR	38.19	1.50	0.57	36.66	0.77	0.28
	St. Cr.	17.47	1.90	0.33	41.87	0.35	0.15

BM, buttermilk; CHR, churn residue; CIP, cleaning in place; St. Cr., Silo steamed cream.

Table 6 Fat content at each sub-stage and using mass balance approach to determine the losses for different processes for Sites 1 and 2 monitored

Serial number	*Fat content in each process*	*Site 1 (MT)*	*Site 2 (MT)*
1	Incoming fat in milk	22.39	22.76
2	Fat in cream	21.92	21.79
3	Fat in skim milk	0.20	0.47
4	Difference	0.27	0.50
5	% loss in separation process	1.21	2.21
6	Fat in butter process	21.76	21.78
7	(butter + buttermilk) Difference ((2) – (6)) (T)	0.16	0.01
8	% loss in butter manufacture process ((7)/(2))	0.73	0.05
9	Total difference ((6)+ (3)) (T)	0.43	0.51
10	Total fat loss at the end of butter production (%)	1.93	2.25

reflected lower quantities of products produced and a slight drop in net value of milk (€361.47/1000 L milk). Lastly, a high loss of 5% was simulated, returning the lowest net value of milk per 1000 L at € 351.12.

DISCUSSION

Processing models

Various processing sector models have been developed and evolved over time as decision support tools for industry. One of the earlier models, developed by Pratt *et al*. (1997), was used to simulate and determine optimum mix for milk and milk products in terms of production and marketing. Similarly, another model by Benseman (1986) was devel- oped for the New Zealand dairy industry for determining the most profitable product mix. Papadatos *et al*. (2002) developed a model for determining the revenue generated for cheese manufacturing process. Geary *et al*. (2010) also developed a similar model simulating different production scenarios to identify the optimal product mix yielding best returns and accounting for variables such as market returns, processing costs and compositional changes for the Irish dairy industry. Burke (2006) used two mathematical models based on linear programming to quantify different parame- ters, such as net cost, revenues and volume of cheese pro- duced; the model developed for butter manufacture accounted for variations in product composition (fat con- tent), processing costs, using market value to determine returns and net value of milk. Garrick and Lopez-Villalobos (2000) developed cost-price models to describe collection, processing and marketing activities for milk and dairy prod- ucts, such as butter, cheese, casein and powders, for the New Zealand dairy industry. Milk processing is a highly complex process with challenges around variables such as seasonality, volume available, market demand, product port- folio and labour requirements (Burke, 2006; Geary *et al*. 2010). The impact of seasonality has not been addressed in the model developed in this study, and seasonality has a sig- nificant impact on milk composition, supply profile, associ- ated production and labour costs, and demand. Adding these factors into the model may thus enhance the effectiveness of the model.

The transition of the Irish dairy sector from quota- and EU-support-based system to a global, market-driven sce- nario has presented many challenges in terms of price and income volatility, with higher investments into efficient dairy processing. Investments in the dairy processing sector have been strong, with ~1.2 c/L per year being invested back (2015–2017) into expansion of facilities in milk pro- cessing sector since the end of milk quotas in 2015 (Moran, 2018) and a further € 300 million investment being expected between 2018 and 2020. The increased investment

has been justified by the relative increase in milk production and processed products.

Mass balance

In this study, the fat conversion process was studied as a batch or a closed-loop procedure to account for all the intakes, processing and outflows, including sludge and CIP procedures. In the dairy industry, the application of mass balances may help in enhancing process efficiencies by underlining key focus areas and identifying key areas where there are losses within the system. The initial estimate for overall fat losses in fat conversion process, from interactions
with the site professionals, was ~2.0–2.5%, but the actual points of loss within the whole fat conversion process were unknown.

Results in Table 5 highlight the efficacy of the mass bal- ance approach allowing industry partners to identify key focus areas within the fat conversion process. For example, the fat content in skim milk at site 1 after separation was 0.10%, as compared to the industry benchmark of 0.06% (source: interactions with industry personnel). The results of fat content from skim and cream highlight the need to address the separation process, with focused investment or solutions needed to improve the separation efficiencies within plants. Higher separation efficiencies will mean lesser losses into skim milk, and thus, higher returns for proces- sors.

Site 1 produced a larger volume of churn residue melt and cream silo flush compared to site 2 (Table 5). Although the melted CIP volumes were recycled for further fat recov- ery at both sites, dairy processing is a water- and energy-in- tensive process. A better control over CIP outflows would allow for reduced energy consumption and make the process more efficient. Improved monitoring of CIP process out- flows, better process efficiency in terms of churn configura- tion, and operation, and optimised process controls like temperature and churn speed, were identified as improve- ment areas as a result of the mass balance model exercise for both sites.

Effect of density on the calculation of available milk fat

The milk intake at dairy processing plants is calculated in terms of volume (litres), but the production data are gener- ated in terms of mass of fat or products produced. For the conversion of volume to weight, milk density is used wherein the volume of milk is multiplied by a density factor to give the effective weight of milk. Thus, a higher density factor will allow for a higher estimation of fat mass in milk (volume × density = mass) compared to a lower density factor. For example, 500 000 L of milk with 4% fat con- verted to weight using 1.0297 g/cm^3 gives a fat mass of
22.59 MT, while using a density factor of 1.0320 g/cm^3 gives an estimated fat mass of 22.64 MT or 0.22% increase in fat estimation. Considering process efficiency values, if the correct conversion factor is not included, this will add to the incorrect assumptions around fat conversion. However, the density of milk is dependent upon the fat content of milk, and there are significant variations in milk fat content due to factors including composition and seasonal variation in milk, genotype and processing conditions, among other factors (Kelsey *et al.* 2003; Lock and Garnsworthy, 2003; Grimley *et al.* 2009; Heck *et al.* 2009; Chen *et al.* 2014; Liu *et al.* 2017).

The weight-volume conversion at both sites was being completed using a single annual average density factor which is not representative of changes in milk profile observed over the period of milk supply, that is, composi- tional changes, supply conditions and lactation cycle and has not been changed in ~25 years within the industry (source interactions with industry and academia profession- als). Both sites monitored in this study used a single density conversion factor of 1.0297 g/cm^3 year-round to convert volume to weight during processing (source: interactions with industry personnel). Changes in practices such as farm management, animal welfare, improvement in genetics and other factors have led to variations in milk constituents and have significantly increased the milk solid content in Ire- land. The average fat content in milk in Ireland has increased from 3.67% in 1998 to 4.14% in 2018, while the average protein content has increased from 3.24% in 1998 to 3.48% in 2018 (Central Statistics Office (CSO), 2018). As stated earlier, density variations are highly susceptible to changes in milk fat content. The increase in fat content from 1998 to 2018 would suggest that the corresponding density factor should also increase. These variations in milk solids have raised the question about accuracy and validity of using the old density factor, and therefore, appropriate den- sity factors need to be developed to enable a well-rounded, accurate mass balance.

Breed and genetic merit

Cow breed has a major impact on the content of each of the constituents of milk (Auldist *et al.* 2004; Bailey *et al.* 2005). Milk composition has a significant bearing on the products portfolio produced, along with net value of milk constituents, net value of milk and total returns. Several researchers in the past have studied the impact of breed on variations in milk constituents (Garrick and Lopez-Villalo- bos, 2000; Auldist *et al.* 2004; Bailey *et al.* 2005; Geary *et al.* 2010). Fat and protein content was found to be higher in Jersey milk compared to HF milk, which greatly enhanced the returns while producing a product mix of 70% fluid milk and 30% cheese/WMP (Bailey *et al.* 2005). Simi- larly, casein content was also noted to be higher for Jersey milk, thus giving a higher yield of cheese and higher returns (Auldist *et al.* 2004). In our study, two genetic types of HF cows, Elite and NA, were analysed, and the results were in line with past studies (Garrick and Lopez-Villalobos, 2000;

Auldist *et al*. 2004; Bailey *et al*. 2005; Geary *et al*. 2010), showing milk with higher solids content to yield higher pro- duct outputs and returns. Milk composition attributes, if included with genetics and breeding programs, can be highly effective in increasing the milk solid content and pro- duct yields and reducing costs by reducing energy, process- ing and fuel costs, and ultimately, yielding higher net returns for milk producers and processors.

CONCLUSIONS

Weight-volume calculations have been identified as an issue for dairy processors in Ireland. Data analysis based on a mass balance approach helped to identify loss points in the process and also enabled processors to reconcile their test results. From an economic perspective, this study identified unaccounted-for losses in the process and helped monitor the overall financial performance. The model was developed to predict the net value of milk or returns based on different scenarios. The model may be a useful tool in determining the 'best-possible' scenario by evaluating all the possible variables such as density factor, breed and processing varia- tions when scenario planning for processing, while this model was developed for milk fat processing, that is, the fat conversion process at a single point of time, with all milk intake being directed to butter manufacture. In reality, milk processing in a dairy environment involves different product mixes, dependent upon various parameters such as supply and demand profile, compositional variances, capacity con- straints, estimating actual costs and returns. If all these parameters, along with a season-based density factor, are incorporated and analysed, it may enable the model to be more adaptive and precise decision support to the changing dynamics of the dairy industry throughout the year.

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AUTHOR CONTRIBUTIONS

Puneet Parmar: Conceptualization, Data curation, Formal analysis, Investigation, Software, Writing-original draft. Nicolas Villalobos: Data curation, Formal analysis, Soft- ware, Writing-review & editing. John Tobin: Funding acquisition, Resources, Supervision, Writing-review & editing. Eoin Murphy: Resources, Supervision, Writing-review & editing. Shane Crowley: Project administration, Supervision, Writing-review & editing. ALan Kelly: Project administration, Supervision, Writing-review & editing. Laurence Shalloo: Conceptualization, Funding acquisition, Methodology, Project administration, Resources, Supervi- sion, Writing-review & editing

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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Article

The Effect of Compositional Changes Due to Seasonal Variation on Milk Density and the Determination of Season-Based Density Conversion Factors for Use in the Dairy Industry

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Abstract: The objective of this study was to determine the effect of seasonal variation on milk composition and establish an algorithm to predict density based on milk composition to enable the calculation of season-based density conversion calculations. A total of 1035 raw whole milk samples were collected from morning and evening milking of 60 spring-calving individual cows of different genetic groups, namely Jersey, Elite HF (Holstein–Friesian) and National Average HF, once every two weeks for a period of 9 months (March–November, 2018). The average mean and standard deviation for milk compositional traits were 4.72 ± 1.30% fat, 3.85 ± 0.61% protein and 4.69 ± 0.30% lactose and density was estimated at 1.0308 ± 0.002 g/cm^3. The density of the milk samples was evaluated using three methods: a portable density meter, DMA 35; a standard desktop version, DMA 4500M; and an Association of Official Agricultural Chemists (AOAC) method using 100-mL glass pycnometers. Statistical analysis using a linear mixed model showed a significant difference in density of milk samples ($p < 0.05$) across seasonal and compositional variations adjusted for the effects of days in milk, parity, the feeding treatment, the genetic group and the measurement technique. The mean density values and standard error of mean estimated for milk samples in each season, i.e., spring, summer and autumn were 1.0304 ± 0.00008 g/cm^3, 1.0314 ± 0.00005 g/cm^3 and 1.0309 ± 0.00007 g/cm^3, respectively.

Keywords: seasonal variation; raw milk; whole milk; composition; milk density; conversion

1. Introduction

Milk and dairy products are important components in the majority of western diets. The composition of milk significantly impacts the quality of final products, acceptability by consumers, and profitability of the dairy industry [1]. Over the past years, multiple studies have been performed to assess variations in the composition of milk. Several factors have been found to be directly or indirectly linked to the changes in milk composition [2–5]. Some of these factors include breed and genotype effects, changes in feeding systems, and the impact of seasonal changes and climatic conditions [6–10]. Climatic conditions may include high temperature variations, microclimate and cold weather conditions. High temperatures may induce heat stress in animals and heat stress has been observed for milk characteristics in Italy [11] and fatty acid composition in Swiss [12], Swedish [2] and Dutch milk [5].

Other factors linked to milk composition include lactation stage [13], animal health [14], herd management and farm and feed management practices [15,16]. The effect of processing on milk composition such as chemical composition, amino acids and fatty acid profile were studied in Ireland [9,17–21] and other parts of the world [22–24]. It has been reported that the availability and concentrations of different constituents of milk, such as fat and protein along with other physico-chemical properties, vary throughout a year [24,25]. This has been mainly attributed to the changes in feeding pattern and the stage of lactation [4]. When cows are grazed outdoors, changes in the feed are induced due to variable climatic conditions and growth stages of the grass that can introduce changes into the milk composition on a frequent basis. Change in the feed type and its effect on milk composition was studied [26] while significant compositional variations were observed when the diet was switched from silage-based to pasture-based and vice versa [27].

Significant variations in fat concentration, fatty acid profile and cheese yield in relation to feed patterns were reported in the past [8,9,28]. Similarly, alterations in feed leading to changes in milk composition have a significant effect on product quality [9,29]. Milk fat and protein content are the two main components that vary significantly due to seasonal variability in feed [30]. A study in the UK showed that the fat content in bovine milk collected between 2009–2013 decreased from January to July, followed by a sharp increase in August and September, remaining constant thereafter [25], while protein content declined steadily from November to April (3.35% to 3.23%), remained constant (April to July), and increased marginally thereafter [25].

Milk composition affects physical attributes like density and, thus, the basis of weight–volume calculations in the dairy processing industry. Changes in density are closely related to solids-non-fat content and fat content of milk [31], higher milk fat represents lower density and vice versa. The density of milk fluctuates between 1.025 to 1.035 g/cm^3 [32] with seasonal changes throughout the year, resulting in higher densities in summer and lower in winter [24]. Density has also been noted to be dependent upon other factors such as temperature and processing conditions like agitation and homogenization [33,34].

The density of milk within a temperature range of 0–60 °C has been studied [35]; the density reduced from 1.0338 g/cm^3 at 0.5 °C and to 1.0296 g/cm^3 at 20 °C, while further decreasing with increasing temperature (1.0220 g/cm^3 at 40 °C and 1.0132 g/cm^3 at 60 °C). The physical state of fat globules becomes important at different temperatures, with crystallisation at lower temperatures (higher density) and melting of fat at higher temperatures (lower density) [36]. The impact of seasonal variation in milk composition profile has been assessed by various studies in the past, but its impact on milk density has not been studied extensively. Milk density is an important parameter in the dairy industry for estimating weight–volume relationships. In dairy processing, milk is supplied in volume (litres) while the final product mix is usually measured as mass/weight (kg), which may introduce variations in measurement. Current practice includes using an average single annual density factor to convert weight to volume; however, milk composition profile varies with different parameters, as stated earlier. Therefore, the use of a single density conversion factor for the weight–volume relationship in a processing environment is not representative of the seasonal changes in milk composition and may cause incorrect estimation of milk constituents (as it does not account for variations in composition observed over different seasons) highlighted in later sections.

The current study was designed to assess seasonal changes observed in raw milk composition by monitoring variations in individual milk constituents over a period of 9 months, covering spring, summer and autumn periods in Ireland. These seasonal changes in raw milk profile were then correlated with milk density to establish a density–

composition relationship. The density–composition relationship helped to evaluate patterns of variation in density across different seasons and determine

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season-based density conversion factors which can be used by dairy processors to accurately estimate the yield of products and profitability of individual processors and the dairy industry as a whole.

2. Material and Methods

2.1 Experimental Design and Sample Collection

The experiment was carried out over a period of approximately 9 months from March 2018 to November 2018, divided into spring (March, April and May), summer (June, July, August) and autumn (September, October and November) seasons. Raw whole milk samples from spring-calved cows was collected from evening and morning milking from the Teagasc Research farm, Kilworth, Co. Cork (Latitude 50°07^{I} N, Longitude 08°16^{I} W). In a spring calving system, cows are calved close to the time when grass grows rapidly, allowing farmers to maximise production from grazed grass, subsequently positively impacting the profitability of their farm. Cows were selected based on their economic breeding index (EBI) (genetic merit) and the individual animal performance. The genetic groups assessed in this study included Jersey and Elite and National Average genetic merit Holstein–Friesian cows. All the cows (n = 60 total, 20 of each genetic group) included in the study were healthy and milked twice a day at 0700 and 1500 h.

Days in milk (DIM) was used as a parameter in the analysis for variation in milk density with season and stage of lactation. The spring calving period for the cows used in this study started at the end of January and continued until the third week of March. Spring season was classified for samples collected between March to May (DIM = 1–123), summer season for samples collected between June to August (DIM = 79–210) and autumn season for samples collected between September to November (DIM = 173–299), respectively.

The cows were also segregated into three groups, for each breed, based on feed. Between six and seven cows from each genetic group were selected based on EBI to be included for each diet pattern and were classified as control, high concentrate and low grass allowance groups [37]. The description of feed allowance is given below.

- High grass allowance: Stocking rate of 2.75 cows/ha, 250 kg N/year. Three kg of concentrate was offered per cow per day immediately post calving to supplement pasture availability in the spring for 12 weeks. Pasture was allocated in accordance with best management practice (approx. 4.5 cm post grazing residual). A grass only diet was offered in the autumn period for 12 weeks.
- High concentrate system: Stocking rate of 2.75 cows/ha. Concentrate (7 kg) was offered per cow per day immediately post-calving to supplement pasture availability in the spring for 12-weeks. Supplementation of 4 kg/day per cow was offered in the autumn period for 12 weeks.
- Low grass allowance: Similar to control with a lower post-grazing residual of 3.5–4.0 cm in spring and autumn.

A total of 1035 milk samples (combined morning + evening milk), approx. 150 mL each, were collected during this period and each of the samples were tested for compositional profile and whole milk density. The evening samples were collected once every two weeks and stored in a standard refrigerator at 4–5 °C overnight to prevent spoilage, while morning samples collected the next morning were then mixed with these to create a representative sample for analysis. The samples were proportionately mixed based on milk yield for the morning and evening milking to ensure that a representative sample was prepared, which was then properly agitated to ensure thorough mixing of constituents and to remove errors due to settling. Sampling requirements were in accordance with ISO 707:2008 (Milk and Milk Products: Guidance on sampling).

2.2 Sample Analysis

The following parameters were tested during the process: milk fat, protein and lactose content and raw milk density. A sample of approximately 30 mL was required for testing on the Dairyspec infrared manual FT model (Make-Bentley systems, Chaska, MN, USA) calibrated for raw whole milk

compositional analysis. Milk density (measured at 20 °C, for all three equipment) was determined using three different pieces of equipment, i.e., DMA 35 portable density meter, DMA 4500 desktop density meter (Make-Anton Paar GmbH, Graz, Austria) and 100-mL calibrated glass pycnometers (Make-BRAND GMBH + CO KG, Wertheim,, Germany), following the procedure described by AOAC standard 925.22.

Before analysis, the density meters were calibrated using distilled water. The measured density of water on DMA 35 was 0.9974 g/cm^3 and, for DMA 4500, it was 0.99826 g/cm^3. The values fall under permissible limits of the theoretical value of 0.9982 g/cm^3 for water at 20 °C. DMA 35 is commonly used for density measurement across industry due to its easier handling and manoeuvrability. DMA 35 works on the FTIR (Fourier transform infrared spectroscopy) principle of a hollow oscillating U-tube technology; the principle of operation is based on changing frequency of a hydrogen-filled hollow oscillator when filled with different liquids. The mass and density of the liquid changes the natural frequency of the oscillator due to overall change in mass of the oscillator when a liquid is added into the tube. The DMA 4500 also works on the similar principle of FTIR as described above. DMA 4500 has an operational range of temperature 0–100 °C and takes only 1–2 mL of sample for density measurement. The equipment is capable of automated cleansing and introduces immediate temperature equilibrium. The measurement principle and method of operation makes it robust and independent of manual interference, thus, reducing risk of errors in measurement. The sample was tested on the DMA 35 with approx. 1–2 mL sample drawn directly from the sample container, and density was noted from the display screen of the equipment. Syringes (2 mL) were used to inject the samples into the oscillating tubes of the DMA 4500 equipment, preventing the flow of air into the sample. Additional sample could be injected into the equipment if air bubbles were noticed on the display, which enabled optimization of the sample measurement to eliminate any errors.

The third method of measuring density was the AOAC 925.22 official method for determining the specific gravity of a liquid using pycnometer. The densities of liquids attained from the pycnometer method are obtained against water. In this method, firstly, an empty glass pycnometer was weighed and noted. The glass pycnometer was then filled with distilled water and wiped dry to remove any water molecules on the outer surface of the pycnometer. This filled weight was then measured and noted, after which the pycnometer was emptied completely. The pycnometer was then filled with liquid (milk) and the outer surface was wiped dry and weighed again. Excess liquid or water from the pycnometer was removed from the pycnometer through a capillary action of the pycnometer lid. The density of the liquid against water was measured using the formula

$$Density = \frac{WS - WE}{WW - WE}$$

where *WS* is the weight of the sample-filled pycnometer, *WE* is the weight of the empty pycnometer, and, *WW* is the weight of the water-filled pycnometer.

2.3. Statistical Analysis

The data for each sampling run were collected and collated for profile and density values for each season. The collected data were firstly analyzed to estimate the distribution of composition throughout the monitored period. Descriptive statistics (mean, standard deviation, minimum and maximum values) for density and milk compositional profile were determined using the MEANS procedure of Statistical Analysis Software (SAS) 9.4 (SAS Institute, Cary, NC, USA). Analyses of variance of the dependent variables (contents of fat, protein and lactose and density) were performed with a linear mixed model using the MIXED procedure of SAS 9.4 (SAS Institute, Cary, NC, USA). The model included the fixed effects of the genetic group, the feeding treatment, parity, the analytical approach for density measurement, days in milk with the linear and quadratic effect as the covariate and random effects of the cow and residual error.

A prediction model was developed using the linear mixed model for estimating density values considering the feeding treatment, the season, the measurement instrument, the genetic group, parity, the interaction between genetic group and the season, the linear effects of percentages of fat, protein and lactose, the linear and quadratic effects of days in milk, and random effects of the cow.

3. Results

A total of 1035 samples (combined morning + evening) were collected, and analyzed to obtain the descriptive statistics results shown in Table 1. The average fat content in milk samples was 4.72 ± 1.30%, and protein, casein, total solids and lactose contents were 3.85 ± 0.61%, 2.88 ± 0.58%, 14.02 ± 2.65% and 4.69 ± 0.30%, respectively, while average density for the study period was estimated at 1.0308 ± 0.0021 g/cm^3. Table 1 also shows the somatic cell count (SCC), calculated as somatic cell score (SCS = log10 (SCC)), which is a marker for hygienic quality of milk samples. The somatic cell score (SCS) average was estimated at 4.66 ± 0.48, while the average somatic cell count was estimated at ~93,300 cells/mL. The somatic cell score calculated for the period of study had no significant impact on milk density found during analysis ($p > 0.05$). Table 2 shows the variations in the composition of milk constituents along with the standard error of the mean with fat contents; there was no significant difference between the seasons of spring (5.00 ± 0.14%) and autumn (5.13 ± 0.14%), while a significantly lower fat content ($p < 0.05$) was obtained in summer (4.71 ± 0.11%). On the other hand, protein content for each season was not significantly different ($p > 0.05$) (3.93 ± 0.05% protein in spring, 3.86 ± 0.04% protein in summer and 3.92 ± 0.05% protein in autumn) and lactose content varied significantly in autumn ($p < 0.05$) compared to the seasons of summer and spring (4.59 ± 0.26% in spring, 4.62 ± 0.17% in summer and 4.68 ± 0.31% in autumn). There was a significant difference in casein content in summer and spring season ($p < 0.05$), while no significant difference was found in casein content for autumn compared to spring and summer (3.00 ± 0.06% in spring, 2.91 ± 0.04% in summer, and 2.93 ± 0.05% in autumn). The total solids content with standard error of mean was significantly different ($p < 0.05$) for autumn when compared to spring and summer (13.95 ± 0.37% in spring, 13.68 ± 0.32% in summer, and 14.72 ± 0.37% in autumn). Descriptive statistics for the complete dataset showed that the minimum density was observed in April, at 1.0298 ± 0.0016 g/cm^3, while maximum density was observed in the autumn period (November at 1.0316 ± 0.0022 g/cm^3).

Trait	Mean	SD	Minimum	Maximum
Fat, %	4.72	1.30	2.14	14.86
Protein, %	3.85	0.61	1.76	5.95
Lactose, %	4.69	0.30	2.45	5.61
Casein, %	2.88	0.58	0.61	5.00
Total Solids, %	14.02	2.65	8.66	22.48
SCS (SCC × '000) [1]	4.66 (93.3)	0.48 (3.35)	3.00 (1)	6.39 (2452)
Density, g/cm^3	1.0308	0.0021	1.0153	1.0378

Table 1. Descriptive statistics of milk composition, somatic cell score and density in milk in samples ($n = 1035$) collected from Jersey ($n = 20$) and Elite ($n = 20$) and National Average ($n = 20$) Holstein–Friesian cows over a period of 9 months (March–November 2018).

[1] Somatic cell score (SCS) calculated as = $\log_{10}$(SCC), SCC = somatic cell count measured in '000 cells/mL.

As shown in Table 3, the highest density value was obtained for the summer season (1.0314 ± 0.00005 g/cm^3) while the lowest density value was estimated for the spring season (1.0304 ± 0.00008 g/cm^3) and autumn had an intermediate density value of 1.0309 ± 0.00007 g/cm^3. There were significant differences in density values for all the seasons ($p < 0.05$), with greatest difference being between spring and summer season (0.001 g/cm^3). All the parameters, i.e., the season, the feeding treatment, the instrument, the genetic group of the animal, parity, the days in milk, and the days in milk squared as well as milk constituents, i.e., fat, lactose and protein, had a significant effect on the variation in milk density ($p < 0.05$), as also shown by the probability values estimated for the factors during analysis

(Table 4). The interactive effect of genetic group and season was the only factor which was not significant ($p > 0.05$), while parity of the animal was also a significant factor and could be included as a parameter in the model. Further analysis of results from the linear mixed model procedure showed significant differences ($p < 0.05$) between measurement techniques (pycnometers and DMA4500, pycnometers and DMA35) but no significant difference between the results for DMA35 and DMA4500. Table 4 also shows the parameters of a linear model to predict milk density, including the season, the feeding treatment, the measurement instrument, the genetic group, parity, the

interaction between the genetic group and the season, the linear effects of percentages of fat, protein, lactose, the linear and quadratic effects of days in milk, and random effects of the cow.

Table 2. Least squares means and standard error of the mean (SEM) of milk composition in samples (n = 1035) collected from Jersey (n = 20) and Elite (n = 20) and National Average (n = 20) Holstein–Friesian cows over a period of 9 months (March–November 2018).

Trait	Season	Mean	SEM
Fat, %	Spring	5.00 [a]	0.14
	Summer	4.71 [b]	0.11
	Autumn	5.13 [a]	0.14
Protein, %	Spring	3.93 [a]	0.05
	Summer	3.86 [a]	0.04
	Autumn	3.92 [a]	0.05
Lactose, %	Spring	4.59 [a]	0.26
	Summer	4.62 [a]	0.17
	Autumn	4.68 [b]	0.31
Total Solids, %	Spring	13.95 [a]	0.37
	Summer	13.68 [a]	0.32
	Autumn	14.72 [b]	0.37
Casein, %	Spring	3.00 [a]	0.06
	Summer	2.91 [b]	0.04
	Autumn	2.93 [a]	0.05

[a,b,c] Means with different superscript within each milk component are significantly different (p-value < 0.05).

Table 3. Least squares means and standard error of the mean (SEM) of milk density in samples (n = 1035) collected from Jersey (n = 20) and Elite (n = 20) and National Average (n = 20) Holstein–Friesian cows over a period of 9 months (March–November 2018).

Season	Mean	SEM
Autumn	1.0309 [b]	0.00007
Spring	1.0304 [a]	0.00008
Summer	1.0314 [c]	0.00005

[a,b,c] Means with different superscript are significantly different (p-value < 0.05).

Table 4. Estimates of parameters and p-values of a linear model to predicted milk density, including the season, the feeding treatment, the measurement instrument, the genetic group, parity, the interaction between the genetic group and the season, the linear effects of percentages of fat, protein, lactose, the linear and quadratic effects of days in milk, and random effects of the cow, in Jersey (n = 20) and Elite (n = 20) and National Average (n = 20) Holstein–Friesian cows.

Effect	Genetic Group	FT	Season	Instrument	Parity	Estimate	p-Value
Intercept						1.00700	
FT							0.024
		HC				0.00012	
		HGA				9.26×10^{-6}	
		LGA				0.00000	
Season							<0.0001
			Autumn			−0.00054	
			Spring			−0.00097	
			Summer			0.00000	

Table 4. *Cont.*

Effect	Genetic Group	FT	Season	Instrument	Parity	Estimate	*p*-Value
Instrument							<0.0001
				Pycnometer		0.00205	
				DMA 35		−0.00006	
				DMA 4500		0.00000	
Genetic Group							<0.0001
	Elite HF					0.00009	
	Jersey					0.00036	
	NA HF					0.00000	
Parity							0.0057
					1	0.00035	
					2	0.00032	
					3	0.00044	
					4	0.00041	
					5	0.00023	
					6	0.00053	
					8	0.00000	
Genetic group × season							0.5545
	Elite HF		Autumn			−0.00002	
	Elite HF		Spring			−0.00015	
	Elite HF		Summer			0.00000	
	Jersey		Autumn			−0.00003	
	Jersey		Spring			−0.00016	
	Jersey		Summer			0.00000	
	NA HF		Autumn			0.00000	
	NA HF		Spring			0.00000	
	NA HF		Summer			0.00000	
dim						−0.00002	<0.0001
dim * dim						6.713×10^{-8}	<0.0001
Fat						−0.00066	<0.0001
Protein						0.00305	<0.0001
Lactose						0.00342	<0.0001

(Elite HF = Elite Holstein–Friesian, NA HF = National Average Holstein–Friesian; FT = feeding treatment, HC = high concentrate feeding, HGA = high grass allowance, LGA = low grass allowance; dim = days in milk).

4. Discussion

4.1 The Effect of Seasonal Variation and Photoperiod on Milk Composition

The effect of seasonal variation and other factors on milk compositional profile has been extensively studied in the literature in the past [2–4,11,38,39]. However, the most important parameters that affect milk composition are diet/feed and the stage of lactation [4,29]. The lactation period significantly affected the milk composition, with late-lactation milk having higher fat and protein content as compared to mid-lactation [29]. The results of this study also align with [29], wherein the fat and protein contents were higher during the later phase of lactation, lowest in the spring period and highest in the autumn period. The density of milk has previously been shown to be dependent on fat and solids-non-fat (SNF) content in milk, and is normally measured at 20 °C [32]. The results from our study show the variation in milk density with season and compositional changes, where the density values in the summer season (lowest fat content) were highest and comparatively lower (1.0309 g/cm^3) in the autumn samples (with higher fat content). Microbiological factors such as somatic cell count were not exclusively included in our analysis. However, somatic cell count (SCC) and somatic cell score (SCS) of milk samples were determined for the study period. The average somatic cell count over the period of study was ~93,000 cells/mL, while the average SCS was estimated at 4.66. In the literature, SCC has been shown to impact milk composition, especially the lactose content of milk due to decreased synthesis of lactose [2]. However, in our study, SCC was within acceptable limits and, thus, no significant impact of SCC was found on milk composition ($p > 0.05$). The total solids content was also higher in the autumn period compared to the summer and spring periods, but there was no significant variation between the summer and spring periods. This is in line with other studies in the UK and Ireland where the total solids content decreased during the January to April and July to August periods [20,24]. As stated earlier, milk yield and compositional characteristics are affected

by the stage of lactation and diet. Milk density is dependent on milk fat and SNF content; therefore, the variation in total solids content also impacts milk density, increasing in the autumn season with increasing lactose and total solids contents of milk. The impact of variation in different constituents, i.e., protein and lactose, is also shown in Table 4 and was statistically significant. Fat content showed the highest variation when compared with protein and total solids, which is in line with the general observation that fat is the most sensitive to dietary changes [5,40]. The density results were determined for major constituents, i.e., milk, total protein and lactose, not segregated for casein (and whey) and/or total solids, to avoid multicollinearity errors in the analysis.

Diet plays a significant role in the variations observed in milk composition [2]. During the grazing season in Ireland, cows graze outdoors, and their diet is comprised mostly of fresh grass. Fatty acids form a significant component of milk fat and variation in fatty acid composition has been mainly attributed to the supply of fatty acids through diet and rumen microbial activity [5]. The main precursors of milk fat, i.e., acetic and butyric fatty acids—derived from rumen fermentation, can be affected by diet through changes in rumen fermentation or the addition of fats for direct absorption and inclusion into milk fat [2]. It has also been shown that the grass consumed by cows during grazing is less mature, and this less mature grass has lower levels of polyunsaturated fatty acids [41]. Oxidative losses in fatty acids due to the wilting and ensiling of grass have also been observed [42]. This reduces the amount of fatty acids from fresh grass and, thus, causes fluctuations in the fatty acid composition of milk, affecting the total fat content and milk density. Therefore, a combination of these factors and seasonal variation impacted the feed quality for grazing cows, which in turn affected the milk composition and milk density, respectively, as shown in results of this study.

Photoperiod is also known to have a significant impact on the milk production and compositional changes in milk. Photoperiod refers to the length of day or the period of daylight received by an organism [43], and the importance of photoperiod on the variations in milk composition has also been highlighted [44]. In dairy cattle, photoperiod influences a series of hormonal changes which affect the milk yield, composition and feed behaviour, among other parameters. Milk yield and dilution of fat and protein content have been reported to vary considerably with the increase in photoperiod or the length of the daylight period [44–46]. Photoperiod, as a factor, was not studied in this analysis but may contribute to the variation in milk composition and milk density and may thus require further analysis and exploration.

4.2. The Effect of Seasonal Variations on Milk Density, Mass Balances and Milk Payment Systems

It is evident from past research and the results of this study that seasonal variations introduce significant fluctuations in fat and protein content, increasing towards the autumn season. The variations in density values can be estimated using the model developed in this study. Variations in different parameters introduce differences in density values and, therefore, the use of a single density conversion factor is not representative of seasonal variations, including compositional changes, climatic conditions and feed practices.

The method of density analysis is also another important factor that can affect the accuracy of measurements. The results shown in this study indicate a significant impact of the measuring technique on the raw milk density for all the samples studied (Table 4, analytical method, $p < 0.001$). The differences in desntiy results between different analytical methods were observed. The pycnometer method was found to have statistically signficant differences with both DMA 35 and DMA 4500 ($p < 0.001$); however, DMA 35 and DMA 4500 results were not significantly different from each other ($p > 0.05$) over the period of study. DMA 35 is used in industry for quick analysis of density (Source: interactions with industry personnel), while DMA 4500 and pycnometer methods are comparatively time-consuming. The results of the pycnometer method were higher than the other two methods, and this may be attributed to different factors, such as accuracy and tolerance limits of the measuring equipment, foreign matter in samples like sediment and particulate matter, entrapped air and bubble formation, viscosity and homogenity of samples, and temperature and temperature history of samples.

In this study, the analysis was carried out in a controlled environment using strong experimental protocols to remove errors or bias.

A mass balance may be defined as the consideration of the input, output and distribution of a product/ingredient between streams in a process. For a butter manufacture process, it may be presented as follows [47]:

$$Fat\ intake = Fat\ in\ products + losses + recycled\ fat$$

The use of a density factor is paramount in terms of a mass balance calculation that can help identify different loss-making points in a process, estimate losses in the fat conversion process and, subsequently, make important process-related and investment-related decisions. Milk payment systems across different regions follow the a multiple component pricing model (A + B - C system), where the value of protein (A) and fat (B) in kg supplied by the farmer to the processor are calculated and the cost of collection and processing (C) in cents per litre, related to the volume of milk supplied by the farmer, is deducted [47]. Milk volume is converted to weight using the density conversion factor by multiplying the volume collected in litres on each farm by the density factor to obtain the weight of milk in kg.

As stated earlier, the profile of milk in Ireland has considerably changed and a single density conversion factor is not representative of the variations in milk profile due to composition and seasonality. To put this in perspective, a hypothetical example is discussed here. The annual supply of milk in Ireland for the year 2019 was 7990 million L of milk [48] with the seasonal profile as supplied, corresponding to a peak milk supply of 13.4% in the month of May and trough of 2.2% in January. Milk distribution for the year 2019 varied between a maximum of 1072.2 million L in May, with the lowest supply observed in December (243.7 million L) and January (175.3 million L). Thus, using season-based density factors, milk weight was determined, giving a peak of 1105.33 million kg (using a density value of 1.0309 g/cm^3) in May, while the minimum weight of milk was calculated for the December (251.38 million kg) and January (180.72 million kg) period using a density factor of 1.0314 g/cm^3. Peak values of milk weight were obtained towards the end of spring and the beginning of the summer period when the milk supply was also at its highest (May–July). When an average density factor (1.0297 g/cm^3, current industry standard) was used to calculate milk weight as compared to the density factors determined in this study, there was a total difference of 9.39 million kg/year in milk kg produced, with monthly differences as high as up to 1.3 million kg.

The model defined in this study can be a useful tool to predict the milk density value that can be used to estimate weight–volume calculations, based on different parameters such as the season, days in milk etc. Milk weight estimated using the predicted density may then be used to determine the fat and protein (in kg) available for processing. This variation in milk weight and constituents estimated from the use of new density factors will require appropriate planning. With proper planning and capacity appropriation, the processors can therefore have better operational control in terms of product mix and capacities, as well as a better understanding of their overall mass balance, while also presenting a more accurate financial picture by having the seasonal density factors calculated appropriately.

5. Conclusions

The density of milk is dependent upon seasonal variations observed in milk composition throughout the year. This is evident from the results of the present study, with density varying significantly with changes in the constituents' content of the milk. Variations in the composition and ultimately density could be attributed to various factors, such as the stage of lactation, climatic conditions (including microclimatic pattern), the feeding pattern during the period of study, housing conditions in autumn and winter seasons, the genetic group, and temperature, amongst other parameters. Seasonal and annual factors for density conversion used in weight–volume relationships were determined, with an emphasis on usage of a periodic, rather than an average, conversion factor evident from the strength of linear regression models. The distribution of density and individual constituents of milk over the different seasons showed a similar trend, with higher fat and protein content observed in

the autumn and winter seasons and the lowest content of these observed during summer. Monthly and season-based density factors were determined, which are relevant for milk-processing planning. Milk density is an important factor in milk processing to estimate the individual milk constituents (weight–volume calculations). The constituent contents thus calculated significantly influence the product portfolio, in conjunction with operating capacities and market demand. The use of season-based density factors, therefore, may improve upon the estimation of individual milk constituents, as shown from this study and, thus, it is vital for the processing industry to plan and control their product mix and operations more effectively. The estimation of new density factors may also enable improvements in the milk payment systems for the production and processing industry.

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The effects of cow genetic group on the density of raw whole milk

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Abstract

The density of milk is dependent upon various factors including temperature, processing conditions, and animal breed. This study evaluated the effect of different cow genetic groups, Jersey, elite Holstein Friesians (EHF), and national average Holstein Friesians (NAHF) on the compositional and physicochemical properties of milk. Approximately 1,040 representative (morning and evening) milk samples (~115 per month during 9 mo) were collected once every 2 wk. Milk composition was determined with a Bentley Dairyspec instrument. Data were analysed with a mixed linear model that included the fixed effects of sampling month, genetic group, interaction between month and genetic group and the random effects of cow to account for repeated measures on the same animal. Milk density was determined using three different analytical approaches – a portable and a standard desktop density meter and 100-cm3 calibrated glass pycnometers. Milk density was analysed with the same mixed model as for milk composition but including the analytical method as a fixed effect. Jersey cows had the greatest mean for fat content (5.69 ± 0.13%), followed by EHF (4.81 ± 0.16%) and NAHF (4.30 ± 0.15%). Milk density was significantly higher (1.0313 g/cm³ ± 0.00026, P < 0.05) for the milk of Jersey breed when compared to the EHF (1.0304 ± 0.00026 g/cm³) and NAHF (1.0303 ± 0.00024 g/cm³) genetic groups. The results from this study can be used by farmers and dairy processors alike to enhance accuracy when calculating the quantity and value of milk solids depending upon the genetic merit of the animal/herd, and may also improve milk payment systems through relating milk solids content and density.

Keywords

Composition • density • genetic group • raw milk • whole milk

Introduction

Composition is an important determinant of the processability and nutritive value of milk (Lindmark-Månsson *et al*., 2003) and also affects the quality of final products (Amenu & Deeth, 2007). Differences in milk composition have been observed among different cow breeds and also in individual cows within the same breed, partially attributed to the genetic variations between cows (McLean *et al*., 1984; Tyrisevä *et al*., 2004; Wedholm *et al*., 2006; Gustavsson *et al*., 2014; Penasa *et al*., 2014; Bland *et al*., 2015; Stocco *et al*., 2017). Past experiments have shown that Jersey cows yield higher concentrations of fat and protein as compared to Friesian cows (Mackle *et al*., 1996; Auldist *et al.*, 2004). These variances in fat and protein are attributed to differences in fatty acid and individual protein profiles within the milk and, according to literature, are influenced by breed (Peterson *et al*., 2002; Soyeurt *et al.*, 2007; Bobe *et al*., 2008; Maurice-Van Eijndhoven *et al*., 2011, 2013). This correlation indicates that genetic selection for milk production affects the composition of milk protein and content of milk fatty acids (McLean *et al*., 1984; DePeters *et al.*, 1995). Similar effects of breed on milk fat and fatty acid composition have also been reported for Irish milk (Lawless *et al*., 1999; Dillon *et al.*, 2003; Walsh *et al*., 2008; O'Callaghan *et al*., 2016).

The density of milk is an important physical characteristic that is widely used for weight–volume calculations, product mix management and profitability calculations. The density

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of milk is used to convert the volume of milk entering a processing environment to weight/mass of milk. The weight of individual constitutents in milk can then be determined by multiplying the mass of milk entering the processing system by the constituents' percentage. There is a direct correlation between the content of fat and milk solids and milk density (Ueda, 1999). Milk density is correlated with the size of fat globules (Ueda, 1999) and fat globule size is dependent upon characteristics like feeding treatment, seasonal and compositional changes, breed, physiology of the animal and stage of lactation (Walstra, 1969; Walstra & Mulder, 1974; Huhtanen & Rinne, 2007; Heck *et al*., 2009; Kljajevic *et al*., 2018; Parmar *et al*., 2020). Breed and genetic characterisitics of the animal significantly affect the concentration and ratio of fatty acids in milk fat and affect the processability, i.e., its hardness or softness (MacGibbon, 1996). Processes such as homogenisation lead to smaller fat globules with a larger surface area, and also a higher density (Truong *et al.*, 2016). It has been noted through past research that the content of fatty acids such as stearic, palmitic, and oleic acids is positively correlated with the size of milk fat globules (Wiking *et al.*, 2004). Milk from different breeds of cows has varying fatty acid content, which affects the overall fat concentration and also affects the fat globule size (Marín *et al*., 2018). This has been attributed to the genetic merit and breed characteristics influencing the milk composition (White *et al*., 2001; Auldist *et al*., 2004; Larsen *et al*., 2012).

Breed variations also impact the protein content in milk as observed from various studies (Ng-Kwai-Hang *et al*., 1986; Malacarne *et al*., 2006; De Marchi *et al*., 2008). Malacarne *et al*. (2006) found that protein content (3.49%) and subsequent cheese yield was markedly higher for Italian Brown cows compared to Friesian cows (3.07%). A comparison of Danish Jersey, Swedish red and Danish Holstein cows also showed that there was a significant difference in concentration of protein in milk (Gustavsson *et al*., 2014). Individual protein concentration and overall content of protein are affected by the genetic variations and influence processing capabilities, including coagulation properties. However, milk density is largely dependent upon factors such as milk fat content, fat globule size and ratio of solid:liquid fat (Ueda, 1999).

Peak season for milk production and supply in Ireland is the period between March and May/June when milk production increases steadily, hitting a peak in May/June, plateauing in July–August and begins falling (off-peak) from the autumn/winter period (as grass growth begins to decline). This is evident from the data available for milk production and intake of creameries in Ireland for 2018 (CSO, 2018). While the effect of breed on milk composition has been well established through numerous research studies, the effect of genetic group on raw whole milk density has not been studied and is unavailable in the literature. The current study was designed to investigate the interaction between cow genetic group and milk density, measured through different analytical approaches, and observed for one complete season (March–November 2018), including peak and off-peak. The composition of milk samples obtained from different cow genetic groups was also measured and results were evaluated to determine the interaction between genetic group, density and analytical approach.

Materials and methods

Experimental design and sampling

The research was carried out over a period of 9 mo from March 2018 to November 2018. Season was defined as spring (March, April, May), summer (June, July, August) and autumn (September, October, November). Raw whole milk samples from the combined evening and morning milking were obtained from a Teagasc Research farm, Kilworth, Co. Cork (latitude 50°07′N, longitude 08°16′W). The genetic groups and breeds assessed in this study included Jersey, and two genetic groups of Holstein-Friesian breed, i.e., elite Holstein-Friesian (EHF) and national average genetic merit Holstein-Friesian (NAHF) cows. FHF and NAHF cows were chosen on the basis of the economic breeding index (EBI). The EBI is a profit index aimed at providing information to farmers regarding the selection of cows for breeding herd replacements (Berry *et al*., 2005). All the cows (n = 54) included in the study were milked twice a day. The cows were segregated into three groups on the basis of feed (three different feed patterns explained below) given to each genetic group and six cows were selected for each feed pattern (6 × 3 = 18 cows per genetic group). Indicative feeding treatments were as follows:

(a) Control system: Stocking rate (SR) of 2.75 cows/ha, 250 kg N/ha. Concentrate (3 kg) was offered per cow per day immediately post calving to supplement pasture availability in the spring (12 wk). Pasture was allocated in accordance with best management practice in mid-season (approx. 4.5 cm post grazing residual; 18 wk). A grass only diet was offered in the autumn period (12 wk). Post grazing residual was managed at 4.5 cm in the spring and autumn.

(b) High concentrate system: Concentrate (7 kg) was offered per cow per day immediately post calving to supplement pasture availability in the spring (12 wk). A 4 kg/cow per day supplementation was offered in the autumn period (12 wk). Pasture allocation, stocking rate and post grazing residual was similar to the control.

(c) Lower grass residual: Concentrate (3 kg) was offered per cow per day immediately post calving to supplement pasture availability in the spring (12 wk). A grass only diet was offered in the autumn period (12 wk). Post grazing

residual was 3.5–4 cm in the spring and autumn. Pasture allocation and stocking rate was similar to the control.

A total of 1,040 samples of approx. 150 mL each were collected during this period and each of the samples were tested for compositional profile and whole milk density. The following parameters were measured: fat, protein, total solids content, while raw milk density was evaluated using three different analytical approaches. The milk composition was determined using a Dairyspec FT manual model (Bentley Dairy Systems, Chaska, MN, USA) while the milk density was determined using three different analytical approaches – using a DMA 35 portable density meter (Anton Paar, Graz, Austria), a DMA 4500 desktop density meter (Anton Paar, Graz, Austria) and 100-cm^3 calibrated glass pycnometers (Blaubrand, Wertheim, Germany). Sampling procedures were in accordance with ISO 707:2008 (Milk and Milk Products: Guidance on sampling) (ISO, 2008). The number of samples collected for each genetic group throughout is shown in Table 1.

Evening samples were collected and stored under refrigerated conditions at 5°C for 18 h to prevent microbial growth and enzymatic activities. Morning samples were collected the next day and mixed with the evening samples to prepare a representative sample. The samples were then tested for composition and density immediately after morning milk recording to prevent alteration to composition or spoilage. Therefore, the analysis was always completed within 24 h of the earliest milk collection.

Methodology

The raw milk density was determined using three different methods, i.e., a DMA35 portable density meter, a standard desktop density meter DMA4500 and the results from these two methods were then compared against the results obtained from measurements using 100-cm^3 glass calibrated pycnometers. The samples collected were properly agitated before analysis to ensure thorough mixing of constituents and to remove any errors due to settling. Before analysis, the density meters were also calibrated using distilled water. Once calibrated, one sample at a time was analysed from start to finish on all three analytical methods, while maintaining sample temperature at approx. 20°C. After completing density measurement for all samples, the samples were then analysed on the Dairyspec infrared manual (Fourier transform (FT) model for milk composition.

DMA4500 and DMA35

DMA35 is used as a method for density measurement across industry due to rapid results, easier handling and manoeuvrability. It works on the principle of hollow oscillating U-tube technology. The principle of operation in the two different pieces of equipment (DMA35 and DMA4500) is based on the principle of changing frequency of a hydrogen filled hollow oscillator when filled with different liquids. The mass and density of the liquid changes the natural frequency of the oscillator due to overall change in mass of the oscillator when a liquid is added into the tube. The DMA4500 is capable of evaluating density with precision of 0.00005 g/cm^3 and 0.02°C with a working temperature range of 0–100°C and requires only 1–2 mL of sample, requires no viscosity-related standards and eliminates temperature-related fluctuations. The DMA4500 can be calibrated at one temperature and all samples for density can be measured at the set temperature. The equipment is also capable of automated cleansing and introduces immediate temperature equilibrium. The measured density of water at 20°C using DMA35 was 0.9974 g/cm^3 and, for DMA4500, it was noted to be 0.99826 g/cm^3, close to the theoretical value of 0.99820 g/cm^3 for water at 20°C.

AOAC standard method using glass pycnometers

The third method used to measure density was the Association of Official Agricultural Chemists (AOAC) 925.22 official method for determining the specific gravity of a liquid using pycnometry. Calibrated 100-cm^3 glass density pycnometers (Make Blaubrand BR43338, Wertheim, Germany) were used to determine the density of the milk samples. The densities of liquids attained from pycnometry method are compared against water. In this method, firstly, the empty glass bottle was weighed and noted. The glass bottle was then filled with distilled water and wiped dry to remove any water on the outer surface of the bottle. This filled mass was then measured and noted, after which the bottle was emptied completely. The bottle was then filled with liquid (milk) and the outer surface was wiped dry and weighed again. Excess liquid or water from the bottle was removed from the bottle through a capillary action of the bottle lid. The density of the liquid against that of water was measured using the formula

$$\text{Density} = \frac{\text{WS} - \text{WE}}{\text{WW} - \text{WE}}$$

Where WS is the weight of a sample-filled bottle, WE is the weight of an empty bottle and WW is the weight of a water-filled bottle.

The sample was firstly tested on the DMA35 with approx. 1–2 mL of sample drawn directly from the sample container and the density was noted from the display screen of the equipment. Secondly, 2 mL syringes were used to inject the samples into the oscillating tubes of the DMA4500 equipment, preventing the flow of air into the sample. The desktop model DMA4500 was adjusted to note the density of milk samples at

20°C for all samples using the temperature settings available on the panel. The milk density of samples was then noted using the glass bottles from the standard AOAC 905.22 method and formula. The same procedure was applied to measure the density of all the samples collected during every run (18 samples for each genetic group each month). The glass pycnometer method requires a minimum of 100-cm^3 sample for density measurement and thus needs to account for insufficient milk produced and collected at the farm, spillage and/or wastage. The number of sample points for the pycnometry method in this study are therefore less (approx. 740), compared to the other two methods (approx. 1,040 for the other two approaches).

After analysis of density was completed, the milk compositional profile, i.e., milk fat, protein and total solids content, was assessed by infrared spectrophotometry. An approx. volume of 30 mL sample was required to be tested on the Dairyspec infrared manual FT model (Bentley Instruments Inc.) calibrated for raw whole milk compositional analysis. The Dairyspec machine is based on the FTIR (Fourier transform infra-red spectroscopy) principle.

Statistical analysis

All dependent variables were analysed using the statistical package SAS version 9.4 (SAS Institute Inc., Cary, NC, USA). Descriptive statistics were obtained using the means procedure. Least square means (LSMeans) and s.e. for factors affecting milk composition and density were obtained using the mixed procedure. The model for milk composition traits included the fixed effects of genetic group, feeding treatment, parity, days in milk with linear and quadratic effect as covariates, and random effects of cow and residual error. Milk density was analysed with the same mixed model as for milk composition with the addition of analytical method (DMA4500, DMA35 and glass pycnometers) as fixed effects. LSMeans were used for multiple mean comparisons using the Fisher's least significant difference test and was implemented in the option LSMEANS and significant differences were defined at $P < 0.05$. Variance components for cow (σ^2_{cow}) and residual (σ^2_e) were used to estimate repeatability of the trait, calculated as $\text{rep} = \sigma^2_{cow} / \sigma^2_{total}$ where $\sigma^2_{total} = \sigma^2_{cow} + \sigma^2_e$.

Results

Descriptive statistics for milk composition for all samples collected during the period of study were determined. The average fat, protein, lactose, total solids, casein and weighted average density values are presented in Table 1. The coefficient of variation was also determined for each of the constituents analysed and are shown in Table 1. Table 2 presents the LSMean values along with the s.e. for the constituents and density based on the genetic groups analysed. The fat content was highest and significantly different for Jersey cows, compared to FHF and NAHF cows ($P < 0.05$). The fat content for Jersey milk was approx. 30% higher compared to NAHF cow milk. Overall milk density LSMean value for Jersey milk was determined to be significantly higher ($P < 0.05$) from EHF

Table 1: Mean, SD, CV and minimum and maximum values of milk composition (n = 1,044) and density – samples (n = 2,836) collected from three cow genetic groups (averaged results)

Trait	Mean	SD	CV	Minimum	Maximum
Fat, %	4.73	1.30	27	2.14	14.86
Protein, %	3.85	0.56	16	1.76	5.95
Total solids, %	14.03	2.21	19	8.57	22.48
Casein, %	2.88	0.58	20	0.61	5.00
Lactose, %	4.70	0.30	6	2.45	5.61
Density, g/cm^3	1.0308	0.002	0.20	1.0153	1.0378

Table 2: LSMEans and s.e. of milk composition and density from three cow genetic groups

Trait	Genetic group[1]	n	LSMean	SE
Fat, %	EHF	357	4.81^b	0.165
	Jersey	341	5.69^c	0.131
	NAHF	346	4.30^a	0.154
Protein, %	EHF	357	3.82^a	0.063
	Jersey	341	4.18^b	0.050
	NHF	346	3.73^a	0.058
Total solids, %	EHF	357	14.11^b	0.242
	Jersey	341	15.36^c	0.185
	NHF	346	13.34^a	0.227
Lactose, %	EHF	357	4.63^a	0.031
	Jersey	341	4.67^a	0.037
	NAHF	346	4.61^a	0.026
Casein, %	EHF	357	2.89^a	0.065
	Jersey	341	3.15^b	0.052
	NAHF	346	2.82^a	0.060
Density, g/cm^3	EHF	330	1.0304^a	0.00026
	Jersey	301	1.0313^b	0.00021
	NAHF	314	1.0303^a	0.00024

[1]Elite HF = Elite Holstein-Friesian, NAHF = national average Holstein-Friesian.

[a,b,c]LSMeans with different superscript within each milk component are significantly different ($P < 0.05$). DMA 4500 method was used to measure density shown in this table. LSMeans, least square means.

and NAHF cows. The difference between density for Jersey cow milk and Holstein-Friesian cow milk was observed to be statistically significant ($P < 0.05$). Table 3 presents the density values for each of the genetic groups estimated with all three measurement techniques. The density values obtained for the pycnometer method were significantly different for all three genetic groups while density values obtained from DMA35 and DMA4500 methods (for all genetic groups) were not significantly different ($P > 0.05$). Table 4 presents the analysis for density when the same number of samples (n) was used for estimation of density for all three measurement techniques. The pycnometer method showed the highest estimate of density and pycnometer density results were significantly higher ($P < 0.05$) from those of the other two methods [DMA 35 and DMA4500, no significant difference between DMA 35 and DMA 4500 ($P > 0.05$)]. Table 5 shows the Pearson's correlation coefficient determining the relationships between the three methods. The pycnometer method was established as the gold standard and the other two methods were compared against it. Lastly, in Table 6, covariance parameters were determined to test the repeatability of the effect of the cow on density variation over the sampling period. Random cow effects on density accounted for 20.45% of between-cow effects and 79.54% for within-cow effects, which could be attributed to genetic merit and inter-genetic group differences.

Table 3: LSMEANS and s.e. of genetic group-wise milk density determined by three analytical methods

Genetic group	Method	LSMean	SE
EHF	Pycnometer	1.0319[a]	0.00024
	DMA35	1.0296[b]	0.00024
	DMA4500	1.0296[b]	0.00024
Jersey	Pycnometer	1.0327[a]	0.00021
	DMA35	1.0308[b]	0.00021
	DMA4500	1.0308[b]	0.00021
NAHF	Pycnometer	1.0318[a]	0.00023
	DMA35	1.0295[b]	0.00023
	DMA4500	1.0296[b]	0.00023

EHF = Elite Holstein-Friesian, NAHF = national average Holstein-Friesian.
[a,b]LSMeans with different superscript within each genetic group are significantly different ($P < 0.05$). LSMeans, least square means.

Table 4: LSMeans, least square means. and s.e. of milk density determined by three analytical methods (to assess the effect of each measurement technique)

Method[1]	N	LSMean	SE
Pycnometer	744	1.0321[b]	0.0001
DMA35	744	1.0300[a]	0.0001
DMA4500	744	1.0300[a]	0.0001

[a,b]LSMeans with different superscript are significantly different ($P < 0.05$).
[1]Analytical methods used for measurement of milk density, discussed in detail in Materials and methods. LSMeans, least square means.

Table 5: Pearson correlation coefficients determined to compare the relationship between the three measurement techniques – pycnometer method as a gold standard; DMA35 and DMA4500 compared with the standard

Pearson correlation coefficients			
	Pycnometer	**DMA35**	**DMA4500**
Pycnometer	1.00	0.82	0.83
Pycnometer		<0.0001	<0.0001
DMA35	0.82	1.00	0.92
DMA35	<0.0001		<0.0001
DMA4500	0.83	0.92	1.00
DMA4500	<0.0001	<0.0001	

Table 6: Estimates of variance components and repeatability of milk density for three cow genetic groups

Trait	Between cows	Within cow	Total	Repeatability (%)
Fat	0.24	0.69	0.93	26.22
Protein	0.04	0.08	0.12	30.26
Density	6.779E-7	2.636E-6	3.31E-6	20.45

Discussion

Effect of genetic group on raw milk density

The impact of breed on different characteristics of milk such as composition profile, fatty acid profile, processability, etc. has been well established in the literature (Malossini *et al.*, 1996; Kelsey *et al*., 2003; Lock & Bauman, 2004; Tyrisevä *et al*., 2004; Yang *et al.*, 2013; Penasa *et al*., 2014; Bland *et al*., 2015; Stocco *et al*., 2017). However, the impact of breed and the use of different types of analytical approaches to measure raw milk density have not been widely addressed, to the best of our knowledge. The effect of genetic group on milk composition, for example, fat and protein levels, fatty acid composition and protein polymorphisms has been discussed widely (Ng-Kwai-Hang *et al*., 1986; Malacarne *et al*., 2006;

De Marchi *et al*., 2008; Heck *et al*., 2009; Kljajevic *et al*., 2018). Because of genetic background and traits, milk samples collected from different cattle genetic groups have diverse compositional profile. A similar trend was observed in the results of this study, where fat, protein and total solids content varied across different genetic groups over the period of study. Sample-related factors include temperature history of the sample, inclusion of air and concentration of fat and solids-non-fat. Other factors affecting physical characteristics and composition of milk may be the genetic merit of the cow, feeding treatment, lactation cycle and period and inter- and intra-herd variations (McLean *et al*., 1984; Wedholm *et al*., 2006; Huhtanen & Rinne, 2007; Gustavsson *et al*., 2014). Sample-related factors such as temperature and temperature history of the sample have been described (Richmond *et al*., 1953; Short, 1955; Hlaváč & Božiková, 2011). The results for milk density from this study show the highest density value for Jersey milk, compared to EHF and NAHF cows. This may be attributed to genetic merit of the animal and variations in milk fat concentration due to genetic group effects.

Genetic merit and its impact on milk composition has been extensively studied in the literature. Milk fat is mainly present in globule form as an oil-in-water emulsion (MacGibbon, 1996) and fat is comprised of approx. 400 different types of fatty acids, out of which approx. 70% are saturated fatty acids and the remaining 30% are unsaturated (Lindmark Månsson, 2008). The fatty acid profile of milk is dependent upon different factors: animal breed, stage of lactation, feed, and microbial activity in the rumen of the animal (Lindmark Månsson, 2008). The main pre-cursors of milk fat, i.e., acetic and butyric fatty acids – derived from rumen fermentation, can be affected by diet through changes in rumen fermentation, directly dependent upon the genetic variations in cows (Lindmark Månsson, 2008). The impact of genetic variations and background significantly affects the fatty acid composition in individual breeds, for example, a higher content of short chain fatty acids and to some extent, medium chain fatty acids were observed in Danish Holstein cows compared to the Danish Jersey breed (Poulsen *et al*., 2012). It has been noted through past research that the content of fatty acids such as stearic, palmitic, and oleic acid is positively correlated to the size of milk fat globule (Wiking *et al.*, 2004). Walstra & Mulder (1974) suggested that the majority (94%) of fat globules are sized between 2 and 8 µm and the fat globule size is dependent upon characteristics like breed, physiology of the animal and lactation period. Milk fat globule size directly impacts the milk density and the size of globules increase with an increase in fat content of milk, due to limited membrane production (Wiking *et al.*, 2004). However, the size of milk fat globule was not measured in this study but it is clear that the changes in milk fat globule size and subsequent milk density are directly correlated to the genetic merit of the animal, as shown from the results of this study (Table 2). This outcome was also corroborated by other studies available in literature (White *et al.*, 2001; Larsen *et al*., 2012) and is independent of dietary effects on composition and only due to genetic traits and breed differences (Beaulieu & Palmquist, 1995). Thus, the size of milk fat globules critically affects the stability, technological and physical properties of milk, such as density, and is reliant on characteristics like breed and physiology of cows (Heck *et al*., 2009; Kljajevic *et al*., 2018). Disintegration of fat globules during proccessing also impacts the size of milk globule and, therefore affects the milk density.

A related assessment of the effect of breed on protein profile and individual protein content was conducted by Gustavsson *et al*. (2014). The results from their study showed a significant impact of breed on the relative overall concentrations of proteins (as shown in the results of this study, milk of Jersey cows have highest protein content, compared to EHF and NAHF strains of Holstein Friesian). Protein content, as well as its composition, is known to impact the processability of milk (Malossini *et al*., 1996; Tyrisevä *et al*., 2004; Wedholm *et al*., 2006; Ketto *et al*., 2017; Poulsen *et al*., 2017), however, its impact on milk density is not clearly established. The impact of seasonal and compositional variation on milk density has been assessed in a study by the same authors (Parmar *et al*., 2020), which showed that variation in milk constituents including protein over different seasons significantly impacted milk density ($P < 0.05$).

Other studies in the literature have observed an inverse relationship between milk fat content and milk density values (Czerniewicz *et al*., 2006). However, Parmar *et al*. (2020) stated that fat content was the most important contributor to density value, other intrinsic (protein, lactose, genetic traits and parity) and extrinsic factors (days in milk, season, feeding treatment, and measurement technique) have statistically significant impacts on milk density.

Effect of analytical technique on the measurement of raw milk density

The results from this study indicate a significant impact of the measuring technique on the raw milk density for all the samples studied. The results were significantly affected by measurement method ($P < 0.05$) with 100-cm^3 glass pycnometers recording the highest values of density for all genetic groups. The results of density measured from 100-cm^3 glass pycnometers, as per the AOAC method, revealed a higher value of density as compared to the results of the DMA 35 and DMA 4500 with all samples undergoing the same treatment and also thoroughly mixed to mix constituents and to avoid any settlement issues (storage conditions). This may be attributed to the precision and tolerance limits of the measurement technique, along with variations in

density introduced due to temperature history of the samples and Recknagel's phenomenon. Recknagel's phenomenon refers to the density of sample measured immediately after milking being lower compared to milk stored for longer periods of time especially at lower temperatures. This is observed due to the increase in hydration of protein at lower temperatures instead of the escape of air bubbles (IASRI, 2012). Another critical factor affecting density measurement using different equipment is the temperature history of the samples. The samples collected in the evening were stored in a refrigerator overnight at 5°C and were mixed with freshly collected samples from the morning milking. This affected the temperature of the representative sample subsequently used for density measurement. The temperature of measurement for the DMA 4500 was standardised at 20°C for all samples while temperature variations could have been introduced into density measurement when assessed on the DMA 35 and 100-cm^3 glass bottles. This may be attributed to the temperature sensitivity of the DMA 35 and no temperature control was used during the use of pycnometers for density measurement. Past research has highlighted the need to determine the controlled temperature history necessary for high precision and accurate density measurement (Sharp & Hart, 1936; Vanstone, 1960; Hilker & Caldwell, 1961). Other factors affecting the density measurement using bottles may include the possible presence of foreign particles in the sample, entrapped air, bubble formation, temperature influence and/or viscosity-related errors.

Conclusions

Genetic traits and merit of the animal significantly impacts on whole milk density, in conjunction with other factors like composition, feed treatment, seasonality, processing environment and temperature. To the best of our knowledge, this is the first of its kind of research, especially for the Irish dairy sector, wherein the breed of the animal has been studied to analyse its impact on milk density, which is an integral parameter in weight–volume calculations in a dairy processing environment. Milk density factors established for different genetic groups in this study may be helpful in estimating weight–volume relationships based on milk supplied from different herds (genetic groups). This will also help in calculating the weight of milk constituents received for processing. The relationship between genetic group and density, thus, established, may enable the inclusion of breed as a support parameter in decision-making for milk payments. Also, the determination of density using different analytical methods presents a new perspective and can influence density values as seen in this study. It was shown that genetic groups producing higher fat content of milk tended towards a higher density value which could be important decision-support information for the milk payment schemes.

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Original article

Effect of temperature on raw whole milk density and its potential impact on milk payment in the dairy industry

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Summary The objective of this study was to determine the effect of temperature on whole milk density measured at four different temperatures: 5, 10, 15, and 20 °C. A total of ninety-three individual milk samples were collected from morning milking of thirty-two Holstein Friesian dairy cows, of national average genetic merit, once every two weeks over a period of 4 weeks and were assessed by Fourier transform infrared spectroscopy for milk composition analysis. Density of the milk was evaluated using two different analytical methods: a portable density meter DMA35 and a standard desktop model DMA4500M (Anton Paar GmbH, UK). Milk density was analysed with a linear mixed model with the fixed effects of sampling period, temperature and analysis method; triple interaction of sampling period x analysis method x temperature; and the random effect of cow to account for repeated measures. The effect of temperature on milk density (ρ) was also evaluated including temperature (t) as covariate with linear and quadratic effects within each analytic method. The regression equation describing the curvature and density–temperature relationship for the DMA35 instrument was $\rho = 1.0338 - 0.00017T - 0.0000122T^2$ ($R^2 = 0.64$), while it was $\rho = 1.0334 + 0.000057T - 0.00001T^2$ ($R^2 = 0.61$) for DMA4500 instrument. The mean density determined with DMA4500 at 5 °C was 1.0334 g cm^{-3}, with corresponding figures of 1.0330, 1.0320 and 1.0305 g cm^{-3} at 10, 15 and 20 °C, respectively. The milk density values obtained in this study at specific temperatures will help to address any bias in weight–volume calculations and thus may also improve the financial and operational control for the dairy processors in Ireland and internationally.

Keywords Density, payment, raw milk, temperature, whole milk.

Introduction

The dairy processing sector contributes significantly to the economy of many countries such as Ireland, Netherlands, New Zealand, Denmark and the United States. For example, in 2017, Irish dairy's economic contribution accounted for approx. one-third, or €4.02 billion, of the total €12.6 billion exports from the food and drink sector, rising by approx. 19% compared to 2016 (Cornall, 2018). In view of this, milk composition is considered as an important parameter for process-ability and quality of final products (Amenu & Deeth, 2007), as well as the yield of products produced from the milk. The composition of raw whole milk procured by dairy processing companies plays a vital role in the profitability of the business and is a key determinant of the value of milk (Lindmark-Månsson *et al.*, 2003). A significant amount of research has been conducted globally to study the physico-chemical properties and variations in milk composition during the course of the year. Variations in composition of milk are dependent on a number of factors, such as season, lactation stage, health of cow, feeding regime and cow genetics (Heck *et al.*, 2009; Kljajevic *et al.*, 2018). As a result, the composition of milk and its associated functional properties can vary significantly throughout the year (Chen *et al.*, 2014). This is particularly true where pasture-based feeding is practised, that is in New Zealand, Australia and Ireland. The associated changes in feeding pattern affect

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the yield and composition of milk throughout the year (Grimley *et al.*, 2009).

Milk density is a function of composition at a given temperature and pressure (Walstra & Jenness, 1984). Density is particularly important in milk processing, where milk intake is typically measured on volume units (L); however, process and final product yields are typically calculated based on weight (kg). Thus, density is calculated as mass = volume × density. Changes in density are closely related to solids-not-fat content, fat content and temperature of milk (Short, 1955). Past research suggests that density of milk fluctuates between 1.025 and 1.035 g cm^{-3} (Scott *et al.*, 1998). Milk density is also dependent upon external factors like processing, agitation, animal genotype, stage of lactation, seasonal variation and homogenisation of milk (Rutz *et al.*, 1955; Short, 1956; Heck *et al.*, 2009; Kljajevic *et al.*, 2018; Parmar *et al.*, 2020). The effect of temperature on milk density has also been studied, and it has been previously shown that milk density decreased as the temperature is increased up to 40 °C (Short, 1955, 1956). Past research also found that pasteurisation affected the milk density negligibly, but that pasteurisation of milk at high temperature 95 °C decreased the density for both whole and skim milk (Short, 1956). Thermal treatment of milk affects the size of fat globules by impacting the crystallisation of fat, which directly impacts on density (Mulder & Walstra, 1974; Van Boekel & Walstra, 1995; Huppertz & Kelly, 2006).

To the best of our knowledge, milk density–temperature relationships have not been analysed for the dairy industry recently, and the past research on this relationship has been completed many years ago (Short, 1955, 1956). The compositional profile of milk has altered considerably since then, due to improvements in animal genetics, health and physiology, management practices, feeding regimes and other factors, thus requiring the current density factors to be evaluated. This study also enables to establish a link between milk density, variations in milk density due to temperature and its usage and impact on milk payment systems.

The current study was designed to assess the impact of temperature on whole milk density for the milk production and processing sector. The temperatures identified to conduct density trials are important during milk processing within a dairy plant and, therefore, can be used to establish weight–volume relationships and to estimate the variations in yield of products and profitability of the milk conversion processes. It is also worthy to note that, in practice, most density measurements are completed at 20 °C at the dairy plant sites, while milk is collected from farms at 4–5 °C. This difference in temperature (between collection and processing) leads to variance in milk density estimation. This study, therefore, aimed to establish density factors at different temperatures, that is 5, 10, 15 and 20 °C, for use in weight–volume calculations.

Materials and methods

Milk samples

Data were available from the 'Next Generation Herd' project at the Teagasc research farm in Kilworth (Co. Cork, Ireland) in 2018. A detailed description of this study has been published previously (O'Sullivan *et al.*, 2019). The farm comprised of an effective area of 93 ha, with a capacity of 200–250 spring-calving cows. For this study, thirty-two Holstein Friesian (HF) individual cows of national average genetic merit were selected for sampling and were chosen on the basis of economic breeding index (EBI), which is a profit index aimed at providing helpful information to farmers regarding selection of cows for breeding herd replacements (Berry & Amer, 2005). EBI is derived from the breeding values of milk production traits, that is milk, fat and protein yields along with two functional traits, that is measure calving interval or fertility, and survival rate of the herd, weighted by the respective economic value. Individual raw milk samples (100 mL each) were collected from Teagasc Kilworth Research Farm, Kilworth, Co. Cork, Ireland (Latitude 50°07′N, Longitude 08°16′W).

Morning samples were collected from the thirty-two HF cows once every 2 weeks over a 4-week period. A total of ninety-three samples were collected for a period of approx. 4 weeks (August 2018) to assess the variations in density associated with temperature. The composition and physical properties were measured every two weeks. The following parameters were measured: fat, protein, total solids, temperature and milk density. Approx. 100 mL samples were collected from each of the selected cows milked using a 20-unit herringbone parlour (Make-DairyMaster, Cincinnati, OH, USA) with daily electronic milk weighing and sampling. Milk samples collected were stored overnight at 4–5 °C to prevent spoilage and bacterial growth before each analysis.

Sample analysis

The compositional characteristics of whole milk samples, that is fat, protein and total solids were determined at 5 °C by Fourier transform infrared spectroscopy using a Dairyspec FT manual model (Bentley systems, Chaska, MN, USA) to determine the variation in fat, protein and total solids content over the monitored period. However, there were no statistically significant differences noted in the constituents for the three sampling periods. The temperature of samples was adjusted using a cooling circulator waterbath, (CC K-6, Make- Huber Kältemaschinenbau AG, Offenburg, Germany). The

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sampling chamber (20 mL) was heated to the required temperature (first measurement done at 5 °C and then heated up to the temperature, 10, 15 and 20 °C) by circulating water though the surrounding jacket for 1 min using a Huber water bath CC-K6 (cooling circulator) (Make- Huber Kältemaschinenbau). A screw nut arrangement at the bottom of the sampling chamber allowed for drainage of each sample, and the chamber was cleaned after every sample.

Density of the samples was determined using two different methods: DMA35 portable density meter (Make- Anton Paar, Hertfordshire, UK) and DMA4500 desktop density meter, (Make- Anton Paar). The DMA35 has a working temperature range of 0–40 °C and density tolerance limit of 0.001 g cm^{-3}. Current industry practice includes the use of a portable handheld density meter (DMA35) (for quicker results, source: interaction with industry personnel). The DMA4500 has a temperature range of 0–100 °C and density tolerance limits of 0.00005 g cm^{-3}. The DMA4500 is capable of automated cleansing, introduces immediate temperature equilibrium and there are no temperature-related ageing effects on the measuring cell. All measurements were made at 5, 10, 15 and 20 °C after storing samples at 4–5 °C for 24 h. Both instruments were calibrated using distilled water to ensure that the measured density of water was within the permitted range (1.0000 at 4 °C – 0.9980 g cm^{-3} at 20 °C) (USGS, 2018).

For the portable density meter, the calibrated density value for water was 0.9971 g cm^{-3} and, for the desktop density meter, it was 0.9988 g cm^{-3}. For the first batch of samples tested at 5 °C, the samples were maintained at the treatment temperature in the water bath and density was measured using the two measurement approaches. After measuring density at 5 °C, the samples were heated to 10 °C by adjusting the temperature of water bath (an equilibration time of approx. 90 s) and density was again measured using DMA35 and DMA4500. The sample remained in the water bath chamber for the duration of density measurement.

New milk samples were collected once every 2 weeks, and the process was repeated for the other temperature combinations, that is 5 and 15 °C and 5 and 20 °C, giving a set of measurements for every batch. The analysis provided a set of three readings for density at 5 °C and one set of readings for each of the temperatures monitored, that is 10, 15 and 20 °C across different samples. The three sets of readings obtained at 5 °C were then statistically analysed.

Statistical analysis

The data were analysed using SAS 9.4 (SAS Institute Inc., Cary, NC, USA) to determine the effect of temperature on the density of the milk. Least squares and standard errors for factors affecting density were obtained using the MIXED procedure. The mixed linear model included the fixed effect of sampling period (early, mid and late Aug), temperature (5, 10, 15 and 20 °C), analytical method (DMA35 and DMA4500) and the triple interaction of sampling period × analysis method × temperature, as well as the random effect of cow to account for repeated measures on the same cow. Least squares means were obtained for the fixed effects and used for multiple mean comparisons using the Fisher's least significant difference test as implemented in the option LSMEANS. Significant differences were defined at $P < 0.05$. Variance components for cow (σ^2_{cow}) and residual (σ^2_e) were used to estimate repeatability of the trait calculated as rep = $\sigma^2_{cow}/\sigma^2_{total}$ where $\sigma^2_{total} = \sigma^2_{cow} + \sigma^2_e$. The effect of temperature on milk density was also determined considering temperature as a covariate in the model described above with linear and quadratic effects within each analytic method. From the model estimates of the regression coefficients, standard errors and *P*-values were obtained to model milk density on temperature.

Results

The milk composition was analysed to determine the mean fat, protein and total solids content. Sampling periods 1, 2 and 3 were defined as the period of sampling milk, that is every 2 weeks during July–August 2018. The samples were analysed separately for three temperature combinations, that is 5–10, 5–15 and 5–20 °C. The changes in density value increased as the temperature increased from 5 to 10 °C and beyond. Table 1 depicts the least squares means of milk density for combinations between sampling period, analytical method and temperature. Least squares mean of milk density for the DMA35 instrument at 5 °C was 1.0330 g cm^{-3}, at 10 °C was 1.0322 g cm^{-3}, at 15 °C was 1.0311 g cm^{-3} and at 20 °C was 1.0296 g cm^{-3}.

The least squares mean milk density values were comparatively higher for DMA4500 for similar test conditions. Table 2 shows the least squares means of milk density at the different temperatures measured with the DMA4500 instrument; density values were 1.0334 g cm^{-3} at 5 °C and 1.0305 g cm^{-3} at 20 °C.

Table 3 shows the estimates of regression coefficients of milk density on temperature with linear (β_1) and quadratic (β_2) effects in each of the analytic method. The two equations corresponding to each of method are shown below.

For DMA35, the equation was $\rho = 1.0338 - 0.0001726T - 0.0000122T^2$ (1) ($R^2 = 0.64$)

And for DMA4500, the equation was $\rho = 1.0334 + 0.000057T - 0.00001T^2$ (2) ($R^2 = 0.61$) where, ρ = milk density in g cm^{-3} and T = temperature in °C.

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Table 1 Least squares means (LSMean) and standard errors of mean (SEM) of milk density ($n = 93$) determined by 2 analytical methods, adjusted for interactions between different sampling periods and temperatures of milk samples

Analytical method	Sampling point (2018)	Temperature	Density LSMeans (g cm^{-3})	SEM
DMA35	2nd Aug	5	1.0330^{a}	0.0001
		10	1.0322^{b}	0.0001
	15th Aug	5	1.0331^{a}	0.0002
		15	1.0311^{b}	0.0002
	30th Aug	5	1.0328^{a}	0.0002
		20	1.0296^{b}	0.0002
DMA4500	2nd Aug	5	1.0339^{a}	0.0001
		10	1.0334^{b}	0.0001
	15th Aug	5	1.0335^{a}	0.0002
		15	1.0319^{b}	0.0002
	30th Aug	5	1.0330^{a}	0.0002
		20	1.0303^{b}	0.0002

(a, b) LSMeans within each date for each instrument with different superscripts are significant different ($P < 0.05$).

Table 2 Least squares means (LSMean) and standard errors of mean (SEM) for milk density for different temperature corrected for effect of sampling period, analytical approach and random cow effects

Effect		LSMean (g cm^{-3})	SEM
Temperature	5	1.0334^{a}	0.0001
	10	1.0330^{b}	0.0002
	15	1.0320^{c}	0.0002
	20	1.0305^{d}	0.0002

(a–d) LSMeans within each effect with different superscripts are significant different ($P < 0.05$). Density values for temperature shown here are for the DMA4500 instrument.

Table 3 Estimates of regression coefficients ± standard error (and *P*-value) of milk density on temperature for different analytical methods

	Analytical method	
Regression coefficient	DMA35 Estimate ± SE	DMA4500 Estimate ± SE
β_0	1.03380 ± 0.00033	1.03340 ± 0.00033
β_1	−0.0001726 ± 0.000056 ($P = 0.0024$)	0.000057 ± 0.000056 ($P = 0.3104$)
β_2	−1.22E-06 ± 2.386E-6 ($P = 0.6102$)	−0.00001 ± 2.386E-6 ($P < 0.0001$)
R^2	0.64	0.61

The quadratic effect of temperature on milk density was significant ($P < 0.0001$) only when estimated in the DMA4500 instrument, indicating curvature in the density–temperature relationship (Fig. 1). The figure also highlights the scale of variation in density values for the two analytical devices measured at different temperatures.

Table 4 highlights the significance of density as a conversion factor in weight–volume calculations for the dairy industry. The effect of milk density on the milk payment for the farmers was evaluated considering the estimation of total milk solids at different temperatures. The data shown in Table 4 were obtained from the Irish Central Statistics Office for the year 2018, and the current density value, 1.0297 g cm^{-3}, used for weight–volume calculations, was obtained from interactions with industry personnel. Irish dairy farmers produced approx. 7.576×10^9 L of milk in 2018, which when converted to weight using the current density factor of 1.0297 g cm^{-3} gives approx. 7.801×10^9 kg of milk. The same produced volume multiplied by a density factor of 1.0334 g cm^{-3}, as determined in this study, gives approx. 7.83 billion kg of milk, a difference of approx. 28.03 million kg of milk (Table 4). When this is equated to kilograms of fat and protein across the entire industry as a whole in 2018, it represents just over 1 million kilograms of protein and over 1.1 million kilograms of fat. Table 5 shows the estimates of variance components for cow, residual and total variation; this parameter was analysed to determine the effect of cow on milk density. Variation between cows accounted for 61.1% of the total variation for milk density, and 39% of the total variation was explained by other environmental factors not accounted for in the model (e.g. variations in pasture quality and relative humidity between paddocks creating different microclimates).

Discussion

Changes in milk density on changing temperature are dependent upon its constituents, especially water and fat (Short, 1955; Hlaváč & Božiková, 2011; Parmar *et al.*, 2020), and may be attributed to the thermal expansion characteristics of fat in milk (Richmond & Davis, 1953). The estimate of repeatability for milk density in our study (61%) was similar to the estimates of repeatability for contents of fat, protein and lactose (Costa *et al.*, 2019) meaning that genetic and permanent effects of the cows are important in explaining the phenotypic variation for milk density during the lactation. The analysis of variance indicated that 39% of the phenotypic variance was explained by environmental factors. Research conducted in the past shows that the changes in density and volume of milk are greater than when compared to water when subjected to different temperatures (Short, 1955). A study to determine the density of water (Lewin, 1972) showed that the density of water peaks at 3.98 °C and

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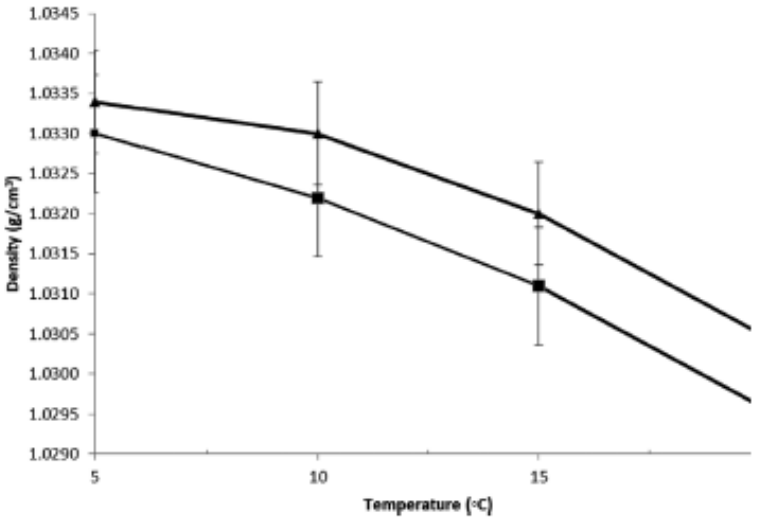


Figure 1 Density as a function of temperature for two different measuring devices, DMA 4500 (▲) and DMA35 (■) adjusted for sampling period, effect of measurement technique and random effects of cow.

Table 4 Weight-volume relationships for Irish milk volumes in 2018, showing differences in fat mass for historical density factor and the new density factor at 5 °C

Description	Density factor (g cm^{-3})	Quantity (in millions)
Estimated volume of milk produced (L)		7576.00
Estimated weight of milk (kg)	1.0297	7801.01
	1.0334	7829.04
Difference in weight estimation (kg)		28.03
Variance in fat at 4.14% (kg)		1.16
Variance in protein at 3.61% (kg)		1.01

Table 5 Estimates of variance components for random animal effects and repeatability of milk density

Cov Parm†	Estimate (×10^{-7})	Repeatability
Cow	6.04	61.13%
Residual	3.84	
Total	9.87	

†Covariance parameter.

maintains a linear relationship with temperature; the density of water does not vary significantly with increasing temperature (1.000 at 4 °C to 0.99802 at 20 °C) (USGS, 2018).

Previous research suggested that the density of milk decreases with increasing temperature up to 40 °C (Short, 1955, 1956). Another study (Hilker & Caldwell, 1961) measured density of milk between 2.2 and 74 °C, and found that minimum density was observed at 74 °C, while the highest value was observed at the lowest temperature. Additionally, it has been reported that the maximum density value for milk was reported between two different temperatures, that is −0.6 and −0.3 °C (Davies, 1939; Olson, 1950), respectively. All the density results from past studies are in line with results from our study, with the highest density being recorded at the lowest temperature and *vice versa*. Watson & Tittsler (1961) assessed the density of raw milk between 1 and 10 °C to replicate a range of milk handling conditions and determined a predictive best-fit equation that could be used to estimate density using fat, solids-not-fat (SNF) and temperature parameters. These authors evaluated density at 4 °C and obtained an average value of 1.0344. However, it was found that most density values were overestimated, and the residual errors became larger as the predicted density increased. This may be attributed to the method used for determining milk density (Ueda, 1999). Further research corroborating this point was shown when the Babcock method and Mojonnier method were compared (Goff & Hill, 1993), where the fat content estimated by the Babcock method produced higher results than the Mojonnier method. Research from the USDA (1965) also pointed out that specific gravity measured by a Lactometer in the method used (Watson & Tittsler, 1961) was lower than that determined with the Babcock bottle method (USDA, 1965).

In addition, solid and liquid fat fractions in milk affect density, and are determined by temperature at the time of measurement and the temperature history of the sample (McCarthy & Singh, 2009; Hlaváč & Božiková, 2011). Milk fat is liquid at temperatures

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above 40 °C and is solidified at −40 °C; it is in intermediate state as a mixture of, crystals and oil at temperatures between 40 and −40 °C (Walstra, 1999). Temperature affects the physical state of fat available in milk and the fat begins to crystallise as the temperature drops. Increasing the fat crystallisation process leads to an increase in milk density. Milk density, as measured in this study, was highest at 5 °C (1.0334 g cm^{-3}) and, as the temperature increased, melting of fats decreased density. It may also be noted that the higher the fat content in milk, the more density varies with increasing temperature, because the volume of fat varies more with temperature compared to water.

Effect of analytical method on density results

Milk density measured for the samples in this study at different temperatures was also impacted by the use of different measuring methods. Referring to the results of milk density for both DMA35 and DMA4500 at the measured temperature, both systems showed a very similar trend, although there were differences in the absolute numbers, with the DMA35 showing a consistently lower density than the DMA4500.

The DMA35 is used regularly in the dairy industry for rapid measurement for milk density (personal communications from industry personnel) and measures the density based on oscillating U-tube technology. The frequency of the oscillator changes due to introduction of liquids, and this variation in natural frequency of the oscillator enables density measurement (Paar, 2009). The effect of instrument was assessed calculating the density of milk at different temperatures after adjusting for any variations introduced due to sampling period, instrument and random effects of cow. Other factors that may also affect the density of samples using different equipment include temperature history of the sample, which introduces small variations in density, Recknagel's phenomenon, which refers to the increased density of stored cold milk, and the level of trapped air. The amount of entrapped air in fresh milk could be as high as 6%, and this entrapped air may influence the milk density measurement and lead to errors in measuring results and poor repeatability (Hyfoma, 2019). Past research has shown that entrapped air does not significantly impact milk density directly but needs to be removed to improve measurement accuracy (Sharp & Hart, 1936; Bouvier *et al.*, 2013). For this study, air bubbles on the oscillating tube were visible on the display screen of DMA4500 during density measurement and can be removed by pushing in more sample using a 2-mL syringe as described earlier. This, therefore, enables more accurate measurement of density without any air-induced errors.

Several researchers have determined the controlled temperature history necessary for high precision and accurate density measurement (Sharp & Hart, 1936; Vanstone & Dougall, 1960; Hilker & Caldwell, 1961). However, for this study, the temperature history did not affect the results because all the samples were subjected to the same procedure and temperature history (equilibrated at each temperature for same time duration).

Implications of milk density measured at temperature (5 °C) on milk payment

Previous research (Shalloo, Dillon & Wallace, 2008) suggested that milk procured from dairy farmers should be paid for based on a multi-component pricing system, that is A+B-C system, which has been used in many countries around the world (e.g. Denmark, Australia, Holland and New Zealand), including Ireland, for approximately 10 years. This system works by putting a value on the kg of protein (A) and fat (B) supplied by farmer to the processor and deducts the cost of collection and processing (C) related to the volume of milk supplied by the farmer. Currently, milk is collected at the farm at ~4–5 °C and, presently, the processors' payment system quantifies the amount of fat and protein using milk volume in litres, milk fat and protein concentration and a density factor of 1.0297 g cm^{-3} for the weight–volume relationship. The reduction in the density of milk with increasing temperature has been noted. Furthermore, it has been found that as the fat content of milk increases, there are larger density changes with temperature variations (Paar, 2009). The density factor is used to convert the volume of milk from litres to weight (kilos) by multiplying the volume of milk with the density factor, that is 1 L of milk at density factor 1.0297 g cm^{-3} weighs 1.0297 kg. The density factor is also used when calculating the amount of fat and protein in milk by multiplying the volume of milk in litres to estimate the weight of milk and multiplying by the concentrations of fat and protein (%) in milk, which generates the mass of fat and protein in milk, respectively. As revealed by the results of this study, milk density varies at different temperatures (reducing with increasing temperature) and significantly impacts the weight–volume calculations.

Density may also be used to calculate the amount of milk solids as depicted by Fleischmann's formula (Ullmann *et al.*, 1985):

$$TS = 1.2 \times F + 266.5 \times \frac{(S-1)}{S}$$

where TS is total milk solids, F is the fat content in milk (both in %) and S is the density.

The above formula shows the importance of milk density and thus implies that total milk solids content

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estimated at lower temperature (5 °C) will be higher than total milk solids estimated at higher temperatures of approx. 20 °C. The results of density estimated in this study were based on a mass per mass basis. The new density factor of 1.0334 g cm^{-3} may be used for volume-weight conversion, that is 1 L of milk with the new factor will weigh 1.0334 kg. This may enable a more precise estimation of fat and protein quantity in milk. An example of the use of the density conversion at the same temperature (5 °C) in weight–volume relationships is shown in Table 4. For total milk produced in Ireland in the year 2018, a significant difference in mass estimation of individual constituents (1.16 million kg in fat and 1.011 million kg in protein) is observed between the use of historical factor, 1.0297 g cm^{-3}, and the new factor, 1.0334 g cm^{-3}. While saying this, it is important to note that while there may have been more kilograms of fat and protein in the milk (at milk density 1.0334 g cm^{-3}) than the conversion factor of 1.0297 g cm^{-3}, in reality, this does not mean that there will be more money to pay out in milk price, but will mean that allocation of payment is aligned with increased levels of milk solids.

However, over time, improving milk payment systems is one of the key areas in developing better communication mechanisms between the farmer and the processor. Ensuring the accuracy of this communication is key to ensuring thrust on both sides. Within the processing plant, accurate measurement of incoming milk constituents, process control and monitoring allocation for product mix under different processing conditions will ensure that any issues that become apparent are identified early and appropriate remedies are put in place in an efficient manner. Accurate monitoring and measurement of temperature and its effect on raw milk density through the quadratic model suggested earlier will enable improvement in milk payment models and impact on the appropriate product mix for processors and profitability of both dairy farmers and processors.

The model developed may enable farmers to estimate density changes based on changes in temperature of milk samples, and the density factor thus estimated can help in measuring the total solids content in milk. The volume of milk produced and supplied from Irish dairy farms has significantly increased since the removal of EU milk quotas, and this research aligns with the current trend, enabling accurate measurement of milk solids and directly impacting the profitability of both dairy farmers and processing industries. The results of this study can be effectively utilised by processors during weight–volume calculations to accurately record the amount of total solids incoming at the plant gates and also monitor and control the milk constituents' conversion process with better efficiency. The temperatures observed in the study were in line with prevalent processing conditions observed at dairy plants (personal communication with dairy plant managers and professionals).

Conclusion

The intake temperature of milk on farm significantly affects whole milk density, along with other external factors such as composition and processing conditions. There is an inverse relationship between temperature and density, that is density of milk decreases with increasing temperature, and there is also a quadratic effect of temperature on milk density. To the best of our knowledge, this study is the first of its kind for the Irish dairy sector and generates a new density conversion factor to be used, for example in the A+B-C milk payment system currently followed in the Irish dairy sector. The results from this study for measurement of density at specific temperatures will help to address any bias in weight–volume calculations and thus may also improve the financial and operational control for the dairy processors in Ireland and internationally.

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Conflicts of interests

The authors declare no conflict of interests.

Peer review

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Data availability statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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