European Innovation Partnership (EIP) Wales

Investigation of the effect of contrasting dairy production systems in West Wales on the profile of milk fatty acids (especially omega-3 and 6)

Final Report – January 2023

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1.0 Summary

This project was designed to investigate the fatty acid (FA) profiles of milk, with a particular emphasis on the polyunsaturated FA (PUFA), and specifically omega-3 fatty acids. The overall aim was to evaluate if a specific management practice on dairy farms contributed to the level of Mega 3 and Omega 6 in the milk.

The omega-3 FA α-linolenic acid is an essential component of the human diet. Like many FA present in milk fat, is produced by plants and is passed into the milk of dairy cows from the diet. To compare the effects of milk production system on the FA profile of milk, farms in West Wales that operate using four different farm types took part in this study: A: conventional herds housed winter/grazing summer; B: herds housed all year round; C: organic herds; D: spring block calving herds. In the period May 2018 to April2020, farms were asked to collect and submit monthly bulk-tank samples of milk and forage used in the diets of their herds for FA analysis at Aberystwyth University. In the period May 2021 to April 2022, bulk tank milk samples were analysed by the National Milk Laboratories milk analysis service.

Milk FA concentration tended to change with time of year depending on the calving pattern (and hence stage of lactation of the herd) and the diets offered to the cows (fresh versus conserved forage diets). The relative proportions of individual FA, and groups of FA (saturated, monounsaturated FA and PUFA) were not statistically significantly different between the farm types, when sufficient data was available to carry out statistical analyses. Variation in the diets, cows, calving patterns and other factors that are well known to influence milk FA composition between farms within each farm type, make it impossible to say whether any particular milk production system within this study produces milk with a FA profile that has the potential to be beneficial to human health.

2.0 Introduction

Fatty acids (FA) that appear in milk fat are derived from two sources – *de novo* synthesis by animal tissues, and through ingestion of fatty acids in feed. The majority of short chain FA - those containing between 4 to 8 carbon atoms (C4-C8) – and medium chain FA (C10-C14) are synthesized *de novo*, relatively little, longer chain FA are produced by the animal and are consumed. Rumen microbial processes can alter the amount and quality of dietary FA, which leads to differences in the proportion (or profile) of FA appearing in milk fat compared to the FA profile in feed. The qualities or chemical and physical characteristics of FA are determined by the extent and location of saturation within the FA molecule. A saturated FA (SFA; e.g. palmitic acid – C16:0) has only a single bond between each carbon atom in the molecule. A monounsaturated FA (MUFA; e.g. oleic acid – C18:1 c9) has a single double bond in the molecule, whereas polyunsaturated FA (PUFA) have more than one double bond in the molecule. The presence of double bonds within a FA molecule changes its shape, which is important in terms of the fluidity of cell membranes – unsaturated FA are bent in shape, and this makes lipid membranes they are part of more flexible because the FA do not pack together as closely as molecules of SFA do. A practical example of this is that suet, which contains a high proportion of SFA, is solid at room temperature, compared to sunflower oil, which contains a high proportion of unsaturated FA and is liquid at room temperature.

Fats are important components of the human diet as a source of energy, and as a source of FA for metabolic functions required for healthy life. Consumption of certain omega-3 (n-3) FA is important for human health. α-Linolenic acid (C18:3, n-3; ALA) is an essential PUFA in the mammalian diet, i.e.,it cannot be produced by mammals and must be consumed as part of the diet. Is it produced by plants and is passed into meat and milk of animals that consume them. Two other omega-3 FA that are important in the human diet are eicosapentaenoic acid (C20:5, n-3; EPA), and docosahexaenoic acid (C22:6, n-3; DHA). These can be made by mammals from ALA but are typically found in high abundance in fish and fish oils, having been obtained from the fish's diet of marine algae and phytoplankton.

Despite there being a common belief that increasing the consumption of omega-3 FA is a way of protecting against cardiovascular disease, there is very little scientific evidence for this. A recent systematic review of scientific studies that have investigated the intake of omega-3 FA (Abdelhamid et al, 2020) concluded that there was little or no effect of EPA and DHA on cardiovascular death or deaths from any cause. Increasing the intake of ALA may slightly reduce the risks of cardiovascular events and heart arrhythmia (Abdelhamid et al, 2020). Another systematic review by Brown et al (2019) concluded that there was little or no effect of increasing omega-3, omega-6, or total PUFA intake in food on prevention and treatment of type 2 diabetes mellitus in humans. Simopoulus (2010) argues that the ratio omega-6:omega-3 FA in food is more important on human health than absolute intakes of each FA type, with an ideal target ratio of 1:1 to 2:1. Ratios higher than this are reported to coincide with chronic inflammatory disease in humans (Patterson et al, 2012), although these and similar findings are still the subject of scientific investigation and debate (Zárate et al., 2017).

3.0 Study Aims

The aim of this project was to investigate whether milk from the forage-based production systems in West Wales contains valuable levels of omega-3 fatty acids and to identify management practices that contribute to this.

4.0 Materials and Methods

Milk and feed samples were collected from dairy farms in West Wales comprising four different dairy production systems:

A: Conventional herds - housed winter/grazing summer
B: Herds housed all year round
C: Organic herds
D: Spring block calving herds

The project was divided into two major sampling periods. The first sampling period was planned to last for 24 months, during which samples would be collected by participating farmers and sent to IBERS for analysis. This sampling period occurred from May 2018 to April 2020. Lack of consistent sample collection and analysis during the first sampling period meant that another 12-month sample collection period was subsequently carried out. The second sampling period occurred from August 2021 to July 2022.

Figure 1: Farmer participants, Hannah & Simon with Emily (Landsker) & Owain (EIP).

5.0 First sampling and analysis period, 2018-2020

Farms were provided with sample collection pots containing Lactab milk preservative and were asked to collect bulk tank milk samples each month and send them to IBERS. Similarly, farms were asked to collect representative samples of forages being fed to their cows each month and send those to IBERS. Upon receipt, samples were frozen to -20 °C for later analysis. Table 1 shows which milk samples were received and analysed in the first year of the project. At IBERS, milk fatty acids from thawed milk samples were extracted and methylated using the method of Lee and Tweed (2008). Fatty acids were analysed using a Varian CP3800 gas chromatograph fitted with a flame-ionization detector (Varian Ltd., Oxford, UK) and a CP-Select column (100 m × 0.25 mm × 0.2 μm; Varian Ltd.) as described by Lee et al. (2005).

Table 2 shows which milk samples were received, but not analysed, in the second year of the project. Far fewer milk samples were received in year 2 of the project, but those that were continued to be stored frozen. These samples were sent for analysis by an external laboratory (National Milk Laboratories [NML], Wolverhampton). However, upon arrival and thawing, the samples were deemed to have deteriorated too much to be analysed and the data were therefore lost.

Feed samples were received from most farms during the first year of the project. These were requested to be collected the day before milk samples were taken, on the basis that milk composition is influenced by the feed previously consumed by the dairy cows. Participating farmers were asked to fill in feed questionnaires to indicate the diets offered to their cows and provide information on the level of production each month. The information sheets returned from each farm are indicated in Table 3. The intention had originally been to store feed samples until the end of the first year, and then analyse feeds from farms that had produced milk with the highest concentrations of omega-3 FA. However, retirement of the experienced lab technician that had the skill and expertise to carry outFA analysis at IBERS meant that this part of the work was not completed.

Figure 2: Farmer participant, Tom Harris at his home farm in Boncath.

The COVID pandemic caused further disruption to the project because Aberystwyth University offices and laboratories closed with the imposition of national lockdown procedures, and therefore prolonged efforts to train other technicians in the FA analysis procedure were not successful. To partially compensate for the loss of in-house analysis, some of the forage samples that had been received and stored frozen (see Table 4) were processed (freeze-drying and grinding) at IBERS and then sent for analysis by an external laboratory (Sciantec Analytical Services, Cawood; method number S1152). Samples from one farm from each of the production systems was chosen for FA analysis based on the completeness of the sample sets, these being farms A1, B11, C23 and D32.

Table 1. Milk samples sent and analysed at IBERS in the first year of the project (black boxes). Farms were coded according to the production system and a number within each of those. Farm A6 was a replacement for farm A3 from December 2018, and farm B16 was a replacement for farm B13 in January 2019. White cells indicate no samples were received for this time point.

	2018								**2019**			
Farm	**May**	**Jun**	**Jul**	**Aug**	**Sep**	**Oct**	**Nov**	**Dec**	**Jan**	**Feb**	**Mar**	**Apr**
A1	■	■	■	■	■	■	■	■	■	■	■	■
A2	■	■	■	■	■	■		■	■	■	■	■
A3	■		■									
A4	■	■	■		■		■					
A5		■		■	■	■	■	■	■	■	■	■
A6								■	■	■	■	■
B11	■	■	■	■	■	■	■	■	■	■	■	■
B12		■	■		■							
B13	■	■	■	■								
B14	■	■	■	■	■	■	■		■	■		
B15		■	■	■	■	■		■	■	■	■	■
B16									■		■	■
C21			■	■	■	■	■	■	■	■	■	■
C22	■	■	■	■	■		■			■	■	■
C23	■	■	■	■	■	■	■	■	■	■	■	■
C24	■	■	■	■	■		■			■	■	
C25	■	■	■		■							
D31		■	■	■	■	■	■			■		■
D32	■	■	■	■	■	■	■	■		■	■	■
D33	■		■	■	■	■			■	■		■
D34	■	■	■	■	■	■	■	■			■	■
D35	■	■	■	■		■						

Table 2. Milk samples sent and stored at IBERS in the second year of the project (black boxes). Farm A6 was a replacement for farm A3 from December 2018, and farm B16 was a replacement for farm B13 in January 2019. White cells indicate no samples were received for this time point.

	2019								**2020**			
Farm	**May**	**Jun**	**Jul**	**Aug**	**Sep**	**Oct**	**Nov**	**Dec**	**Jan**	**Feb**	**Mar**	**Apr**
A1		■	■	■	■	■	■	■	■	■	■	■
A2				■						■		
A3												
A4												
A5			■	■	■	■	■	■	■	■	■	
A6												
B11			■	■			■					
B12												
B13												
B14												
B15			■	■	■		■	■	■	■		■
B16				■						■	■	■
C21			■	■	■	■	■	■		■	■	■
C22			■	■	■	■	■	■		■	■	■
C23												
C24			■	■	■	■	■	■		■	■	■
C25							■	■		■		
D31		■	■	■								
D32		■	■	■	■	■	■	■				
D33			■	■	■	■	■	■				
D34			■	■	■	■	■	■				
D35		■						■				

Table 3. Reports received from participating farms (black cells) on diets offered and herd management parameters. White cells indicate no reports were received for this time point.

	2018								2019												2020			
Farm	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr
A1	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■		■	■	■	■	■			
A2	■	■	■	■	■			■	■	■	■													
A3																								
A4	■	■																						
A5		■	■																					
A6																								
B11	■	■	■	■	■	■																		
B12																								
B13	■	■	■																					
B14	■	■	■	■	■	■																		
B15	■	■	■			■	■							■										
B16																								
C21		■	■	■	■																			
C22	■	■																						
C23	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■	■				
C24																								
C25	■	■	■																					
D31			■			■																		
D32	■	■	■	■	■					■	■	■	■		■	■	■							
D33				■		■																		
D34	■	■	■	■	■	■	■	■																
D35						■																		

Table 4. Forage samples sent and stored at IBERS in the second year of the project (coloured boxes). Green boxes signify fresh grass received, brown boxes signify silage samples received. In some months, both fresh and ensiled samples were received. Samples of whole-crop silage, rape, and concentrates received are not shown. White cells indicate no samples were received for this time point. Cells with a tick mark indicate those samples analysed for fatty acid composition. Farm A6 was a replacement for farm A3 from December 2018, and farm B16 was a replacement for farm B13 in January 2019.

	2018								2019			
Farm	**May**	**Jun**	**Jul**	**Aug**	**Sep**	**Oct**	**Nov**	**Dec**	**Jan**	**Feb**	**Mar**	**Apr**
A1	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓
A2												
A3												
A4												
A5												
A6												
B11	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓
B12												
B13												
B14												
B15												
B16												
C21												
C22												
C23	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓
C24												
C25												
D31												
D32	✓	✓	✓	✓	✓	✓	✓	✓		✓	✓	✓
D33												
D34												
D35												

6.0 Second sampling and analysis period, 2021-2022

Lack of data caused by staff leaving IBERS and disruptions due to COVID meant that a second sampling period was started. This utilised the relatively new service of the prediction of milk fatty acids by National Milk Laboratories, with bulk tank milk samples being collected by the collection tanker drivers, as is routinely done for standard milk quality analysis. This simplified the sample collection and analysis process, although the fatty acid data provided in this way was much less detailed than was provided by the full gas chromatography methods used previously.

Milk data were collected from farms using the same production systems (farm types) as previously. Some farms were the same as before, with others being enrolled to replace those who had exited the project. In this round of sampling, there were 5 farms in category A (conventional winter house/summer grazed) and 4 in each of categories B, C and D (housed year round, organic, and spring block calving respectively). Sample data received from NML is summarised in Table 5. It is not clear why, in some months, e.g., November 2021, two sets of data were received for almost all farms. The data for some farms appears to have been duplicated, because all the values were the same (including the stated sampling date), whereas for other farms (e.g., C23*), additional (different) data from consecutive (or even the same) sampling days were received. For the data analysis including all farms, data from a single sampling were used where more than one was supplied for a particular month.

Some forage samples were collected during the second sampling period and analysed for standard nutritional composition by Sciantec; the exact analyses carried out depended on the forage sampled.

Table 5. Summary of the number of NML-analysed milk sample data received from May 2021 to April 2022. The farm codes listed here are do not correspond to the same farms in the first sampling period, which is signified by the * against each code. However, the letters A-D in each farm code do correspond to the same production systems.

	2021								2022			
Farm	**May**	**Jun**	**Jul**	**Aug**	**Sep**	**Oct**	**Nov**	**Dec**	**Jan**	**Feb**	**Mar**	**Apr**
A1*	1	1	1	1	1	1	2	1	1	1	1	1
A2*	1	1	1	1	1	1	2	1	1	1	1	1
A3*	1	1	1	1	1	1	2	1	1	1	1	1
A4*	1	1	1	1	1	1	2	1	1	1	1	1
A5*	1	1	1	1	1	1	2	1	1	1	1	1
B11*	1	1	1	1	1	1	2	1	1	1	1	1
B12*	1	1	1	1	1	1	2	1	1	1	1	1
B13*	1	3	1	1	1	1	2	1	1	1	3	3
B14*	1	1	1	1	1	1	2	1	1	1	1	1
C21*	1	1	1	1	1	1	2	1	1	1	1	1
C22*	1	1	1	1	1	1	2	1	1	1	1	1
C23*	1	3	3	3	3	1	4	3	3	3	3	3
C24*	1	0	1	1	1	1	2	1	1	1	1	0
D31*	1	1	1	1	1	1	2	1	1	1	1	1
D32*	0	1	0	1	1	1	2	1	1	1	1	1
D33*	1	1	1	1	1	1	2	1	1	0	1	1
D34*	1	1	1	1	1	1	2	1	1	1	1	1

7.0 Data analysis

Insufficient milk sample data were generated to carry out statistical analyses on them in the 2018-2019 sampling year. Therefore, mean and standard error data for each farm type (A to D) were calculated and plotted according to sampling month. Even fewer forage samples were received from participating farms in the 2018-2019 sampling year, and the data from a single farm from each farming system were plotted according to sampling month.

Data from samples collected during the second phase of the work, 2021-2022, were statistically analysed using linear mixed models (REML), using farm type and sampling month as fixed (treatment) components, and farm identification number as the random component. Samples were collected on different days from different farms, and were collated according to collection date. This, samples collected at the end of May 2021 were given a May 2021 collection date, samples collected at the end of June/early July were given a June 2021 collection date, and so on. To be able to compare FA data from the two sampling periods, milk FA profiles were calculated as a percentage of milk fat. This is not exactly the same as the FA profiles previously calculated as a percentage of total FA, because milkfat contains compounds other than FA, making the fat concentration slightly higher than the total FA concentration of a sample of the same milk.

8.0 Results and Discussion

8.1 First sampling and analysis period, 2018-2020

Four omega-3 (n-3) FA were measured in the milk samples received, which were α-linolenic acid (C18:3, n-3; ALA), eicosapentaenoic acid (C20:5, n-3; EPA), docosapentaenoic acid (C22:5, n-3; DPA), and docosahexaenoic acid (C22:6, n-3; DHA). These accounted for grand means of 0.72%, 0.07%, 0.10% and 0.01% of total fatty acids (TFA) respectively. α-Linolenic acid accounted for 77% of the sum of omega-3 FA overall and was therefore considered the most important for the purposes of this study. Milk ALA percentage of TFA was consistently higher in milk from organic system (system C) cows (Figure 1) over the spring and summer months and was lowest in the milk of year-round housed (system B) cows. There was little apparent difference between the farm types during the winter months (December to February), when most farms were feeding silage (Table 4). There are no milk data for January 2019 for the spring block calving herds, as expected.

Milk linoleic acid (LA) proportions were highest in the milk of year-round housed cows (system B) during the summer and autumn months (Figure 2), with less difference between farm types in the winter and early spring months. This led to the highest ratios of omega-6 to omega-3 (LA:ALA) being found in the milk of system B cows (Figure 3). Although it is not statistically valid to compare the FA composition of forage samples from one farm from each farming system with the mean FA composition of milk samples received from all farms of the same farming system, particularly since the forage data for the B farm was mostly from maize silage, compared to grass for other farms, Figure 11 shows that as the forage ALA (omega-3) percentage of TFA increased, milk ALA proportion also increased, although only marginally. A clearer relationship is seen between forage and milk LA (omega-6) profiles (Figure 12). Farm type B farms were those that housed cows all year round, and the forages sent for analysis were all silages, apart from fresh grass sampled in July 2018 (Table 4); thesilage analysed from farm B11 was maize silage, which has a much higher proportion of LA and oleic acid than grass. This compares with a mixture of fresh and ensiled grass fed on other farm types. Previous work has shown that the FA profile of grass tends not to be affected by the ensiling process,although the absolute concentration of FA can increase as other components of grass are lost (Alves et al, 2011).

Oleic acid (C18:1, c9) is a MUFA that accounted for between 15% and 20% of TFA in the milk of cows in this study (Figure 4), clearly far higher than the combined percentages of the PUFAs ALA and LA (between 1% and 2% of TFA). Oleic acid may contribute to the perceived healthiness of milk fat as an important part of the complement of unsaturated FA. In this study, milk from cows on farm type A tended to have higher proportions of oleic acid in the autumn months, and the proportion of TFA dropped slightly over the winter on all farming systems. Despite the much higher proportion of oleic acid found in the maize silage fed on the B farm (Figure 9) the proportion of oleic acid in the milk from cows on this farm was not substantially different to other B system farms (as shown by the size of the error bars on Figure 4).

Palmitic acid (C16:0) is a very common SFA that can be synthesised in the body or derived from dietary sources. In dairy cow diets, for example, Megalac is a rumen-protected fat source made from palm oil, which is rich in palmitic acid. In this study, palmitic acid accounted for approximately a third of allFA in milk (Figure 5), and milk from year-round housed cows tended to have a higher proportion of palmitic acid in their milk, possibly as a result of the mixed diet they received. The differences between farm types were reduced in the winter months, presumably when cows were housed and offered mixed diets based on conserved forages and concentrates, as evidenced by an increase in the proportion of palmitic acid in TFA in late autumn onwards.

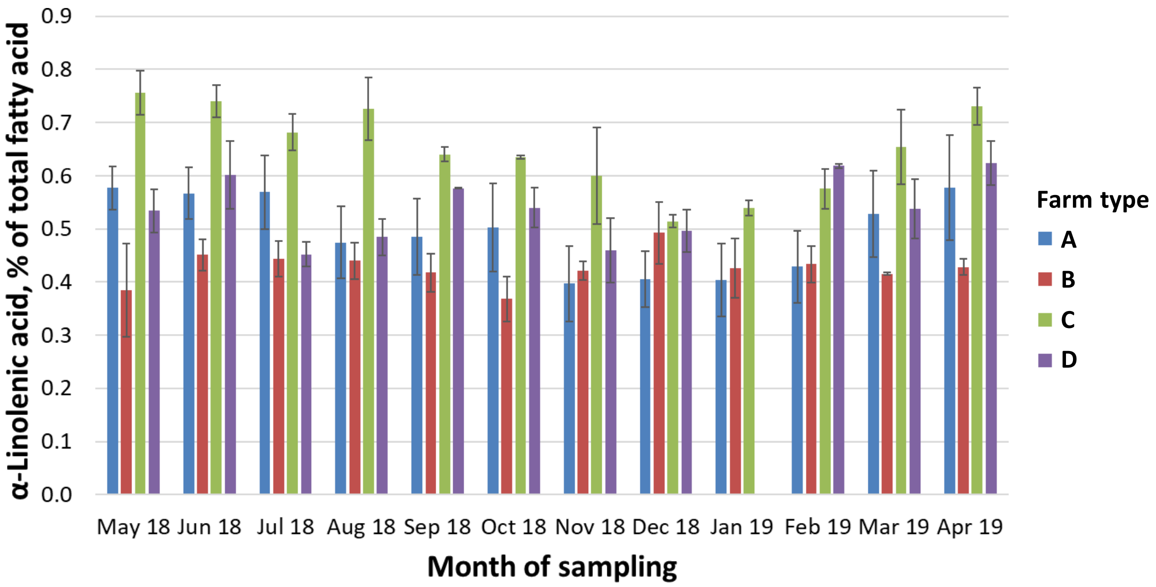

Figure 1. Mean (± standard errors) profile of milk α-linolenic acid (omega-3; ALA) during the sampling period May 2018 to April 2019 for each farm type. A: Conventional herds housed winter/grazing summer; B: Herds housed all year round; C: Organic herds; D: Spring block calving herds.

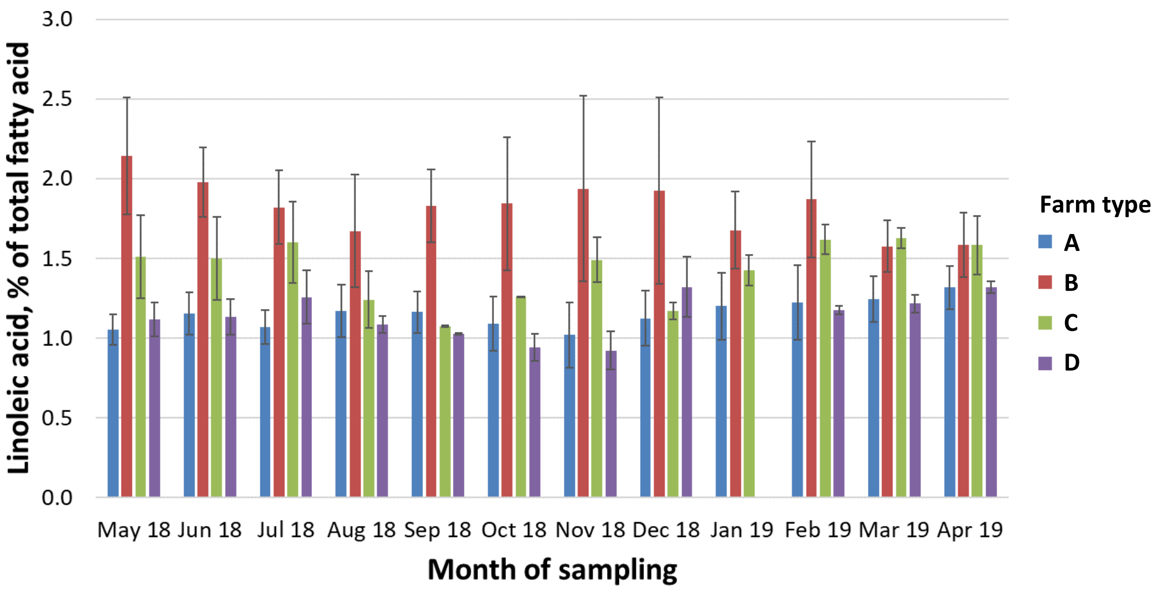

Figure 2. Mean (± standard errors) profile of milk linoleic acid (omega-6; LA) during the sampling period May 2018 to April 2019 for each farm type. A: Conventional herds housed winter/grazing summer; B: Herds housed all year round; C: Organic herds; D: Spring block calving herds.

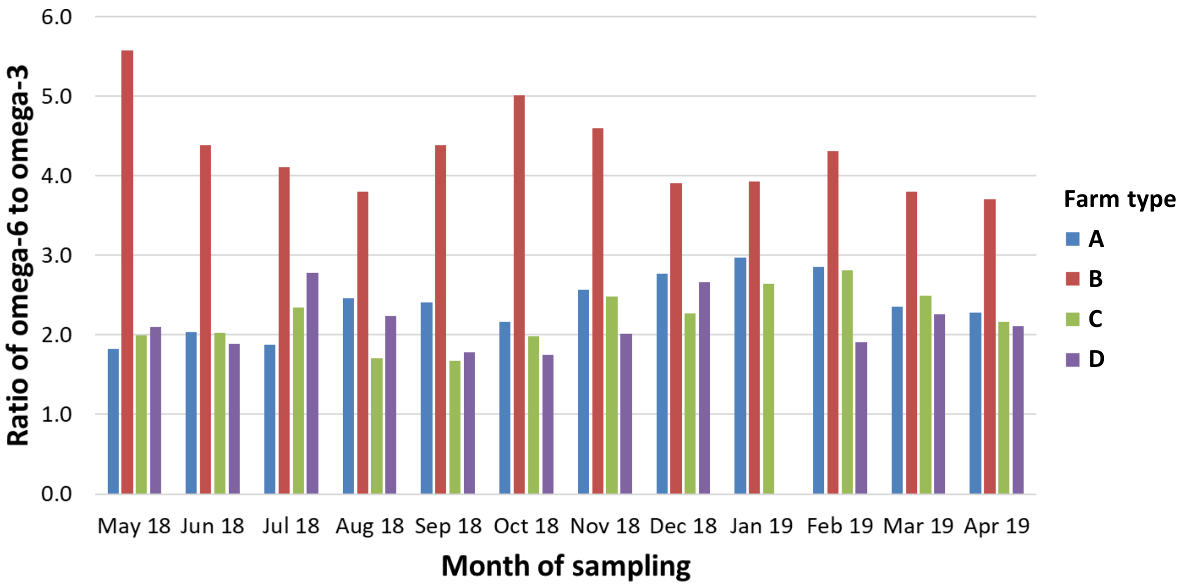

Figure 3. Mean ratios of milk linoleic acid (omega-6) to α-linolenic acid (omega-3) during the sampling period May 2018 to April 2019 for each farm type. A: Conventional herds housed winter/grazing summer; B: Herds housed all year round; C: Organic herds; D: Spring block calving herds.

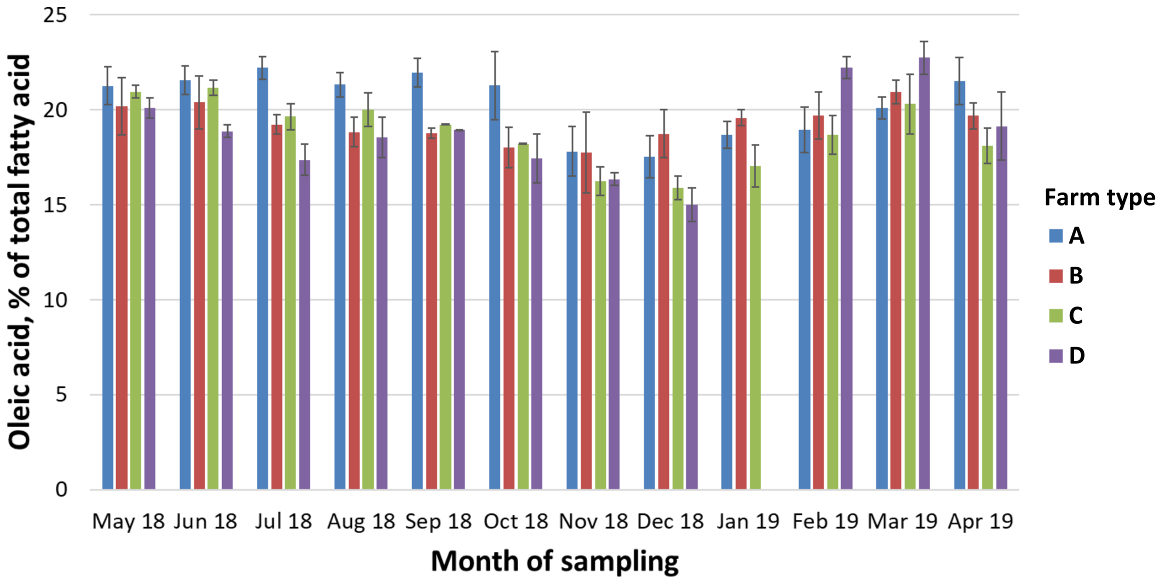

Figure 4. Mean (± standard errors) profile of milk oleic acid (C18:1, c9) during the sampling period May 2018 to April 2019 for each farm type. A: Conventional herds housed winter/grazing summer; B:Herds housed all year round; C: Organic herds; D: Spring block calving herds.

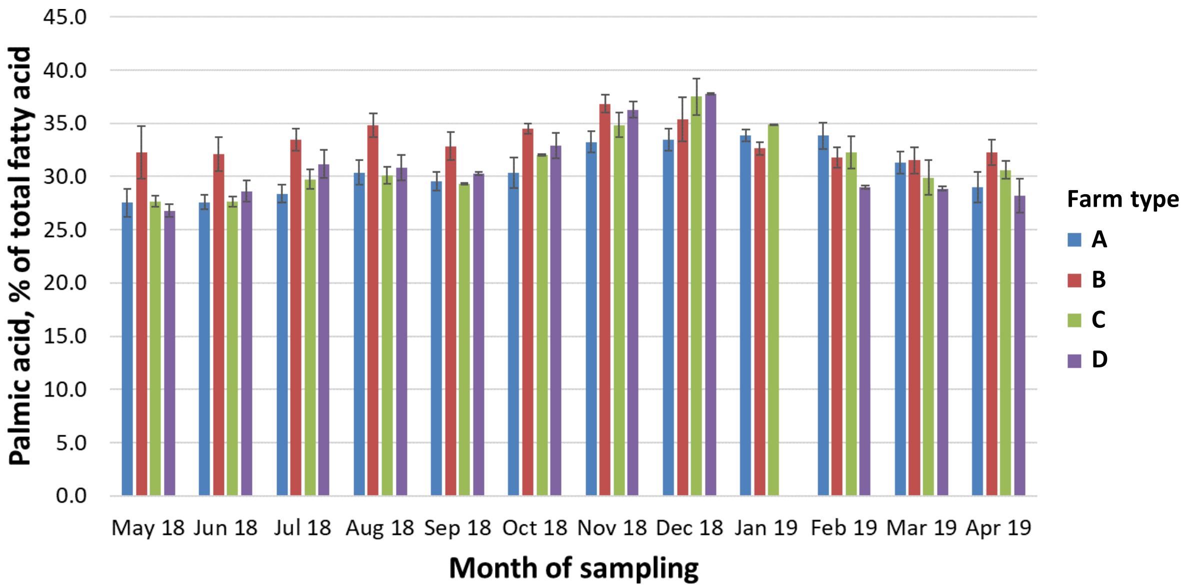

Figure 5. Mean (± standard errors) profile of milk palmitic acid (C16:0) during the sampling period May 2018 to April 2019 for each farm type. A: Conventional herds housed winter/grazing summer; B:Herds housed all year round; C: Organic herds; D: Spring block calving herds.

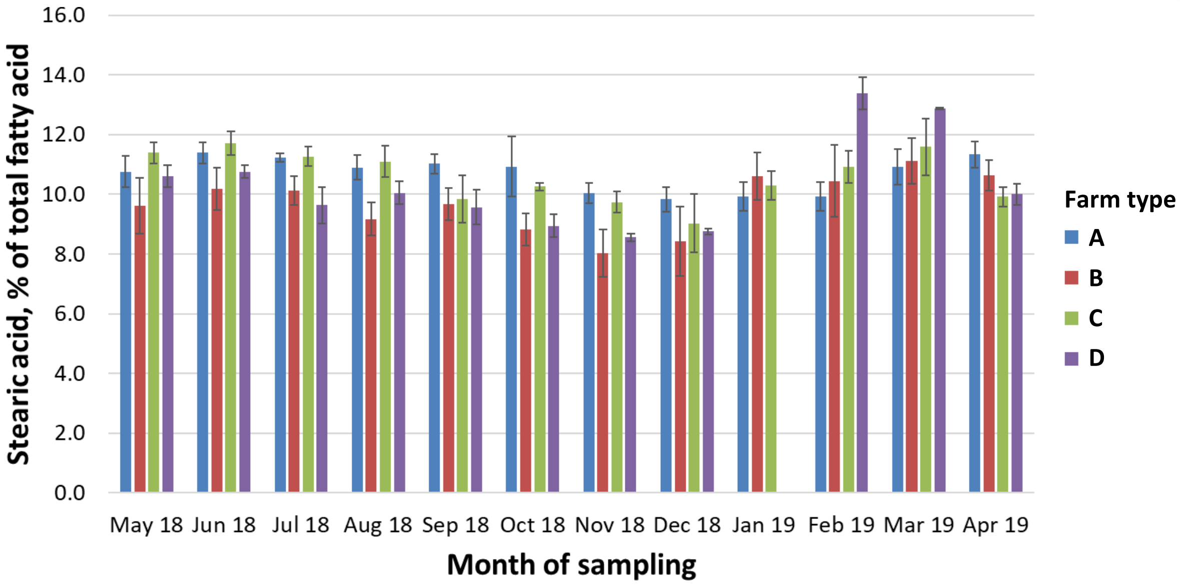

Figure 6. Mean (± standard errors) profile of milk stearic acid (C18:0) during the sampling period May 2018 to April 2019 for each farm type. A: Conventional herds housed winter/grazing summer; B: Herds housed all year round; C: Organic herds; D: Spring block calving herds.

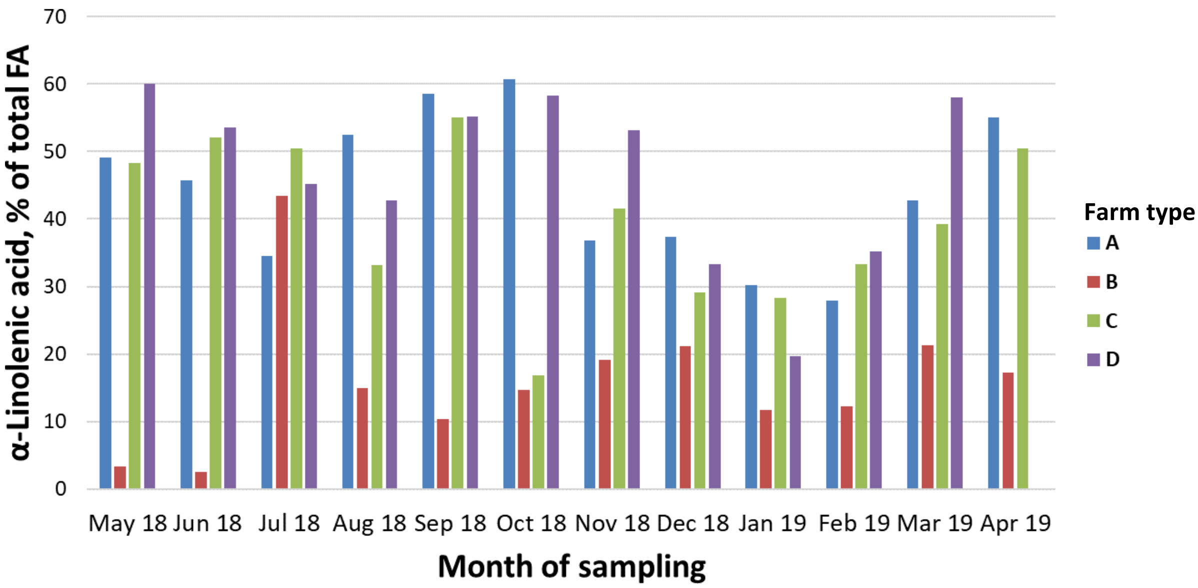

Figure 7. Profile of forage α-linolenic acid (omega-3) during the sampling period May 2018 to April 2019 for one farm from each farm type. A: Conventional herd housed winter/grazing summer; B: Herd housed all year round; C: Organic herd; D: Spring block calving herd.

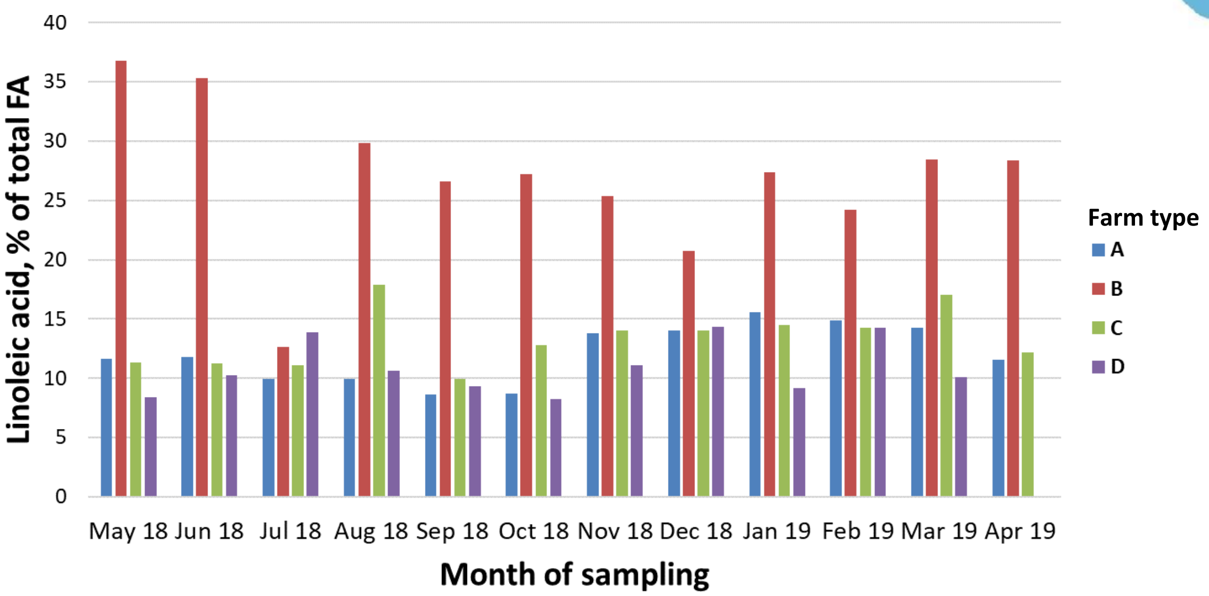

Figure 8. Profile of forage linoleic acid (omega-6) during the sampling period May 2018 to April 2019 for one farm from each farm type. A: Conventional herd housed winter/grazing summer; B: Herd housed all year round; C: Organic herd; D: Spring block calving herd.

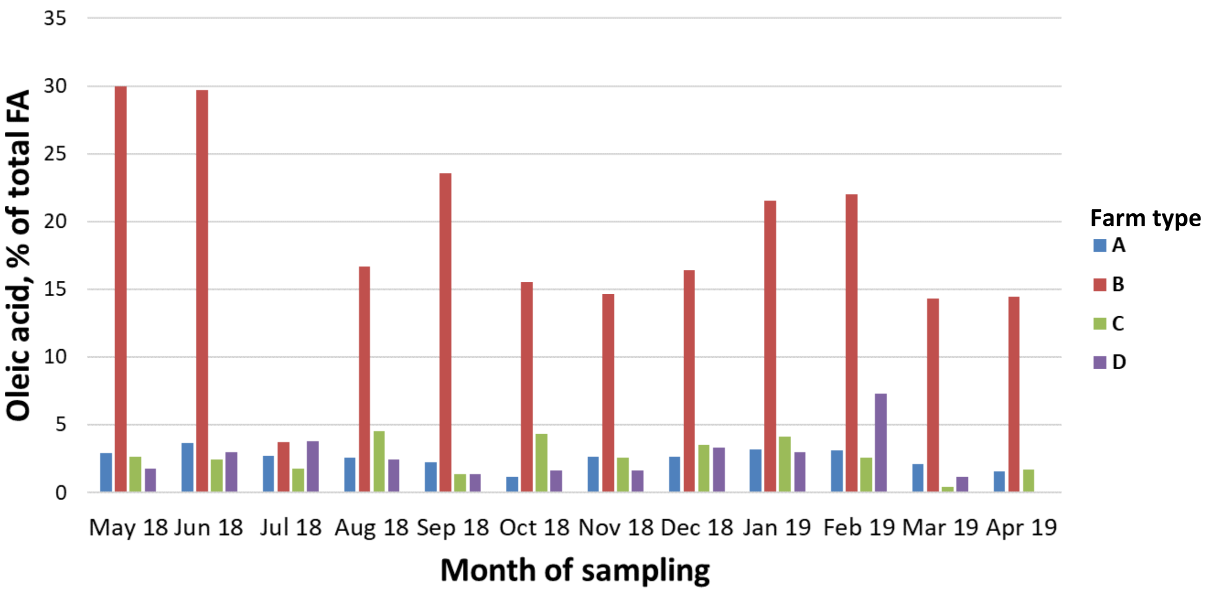

Figure 9. Profile of forage oleic acid (C18:1, c9) during the sampling period May 2018 to April 2019 for one farm from each farm type. A: Conventional herd housed winter/grazing summer; B: Herd housed all year round; C: Organic herd; D: Spring block calving herd.

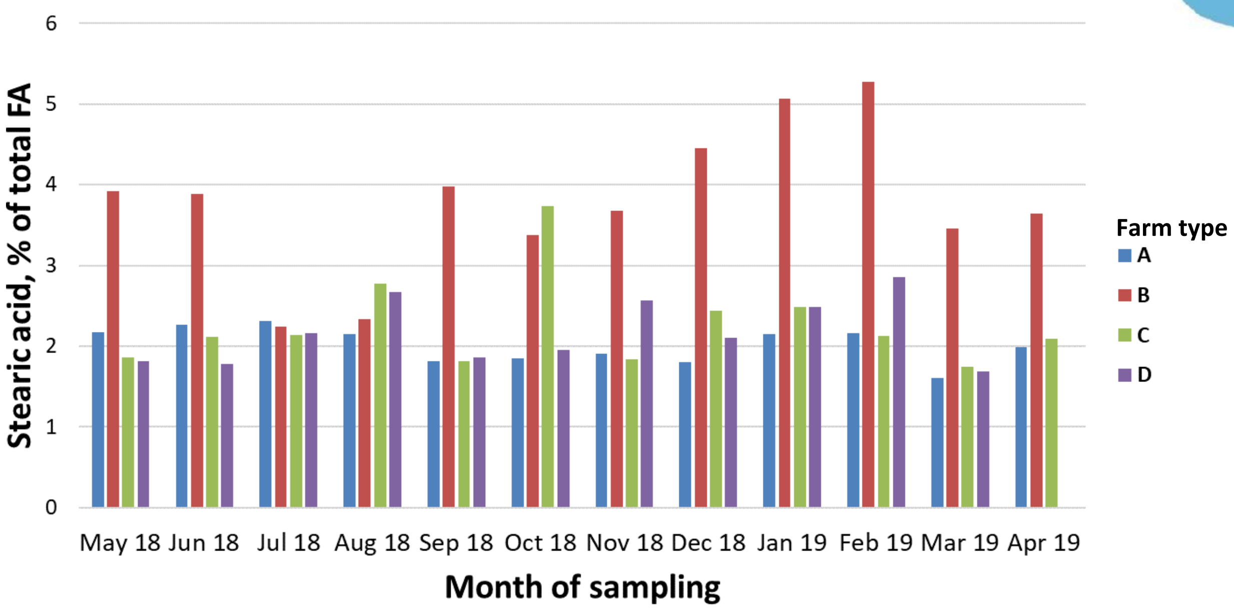

Figure 10. Profile of forage stearic acid (C18:0) during the sampling period May 2018 to April 2019 for one farm from each farm type. A: Conventional herd housed winter/grazing summer; B: Herd housed all year round; C: Organic herd; D: Spring block calving herd.

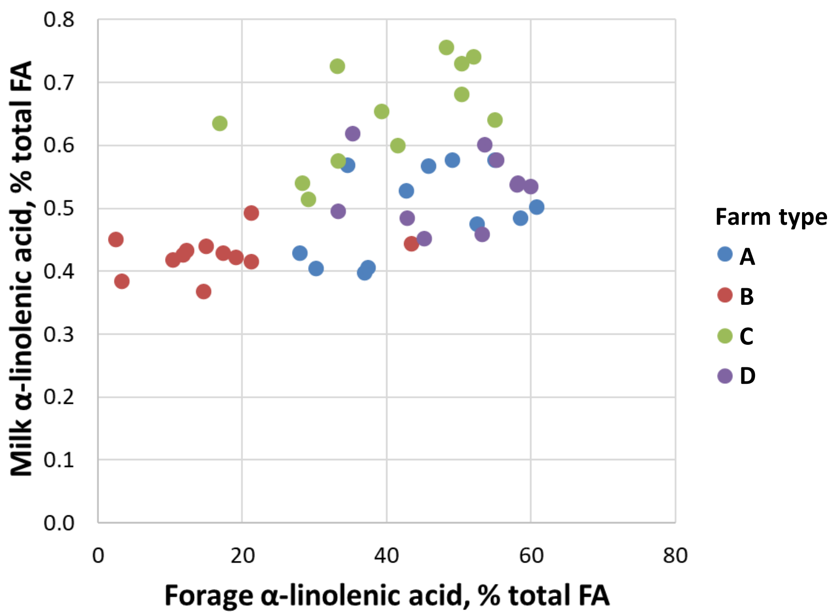

Figure 11. Plot of forage α-linolenic acid (omega-3; ALA) profiles during the sampling period May 2018 to April 2019 against mean profiles of milk ALA from the same sampling months. Forage data were analysed in samples collected from one farm of each farm type, whereas milk data are mean values for all milk samples received from farms of that type for each sampling time. A: Conventional herd housed winter/grazing summer; B: Herd housed all year round; C: Organic herd; D: Spring block calving herd.

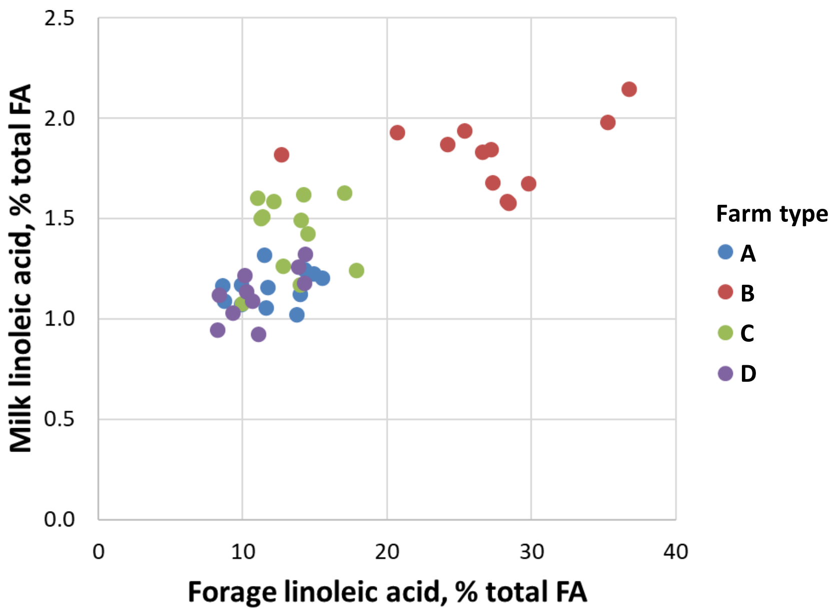

Figure 12. Plot of forage linoleic acid (omega-6; LA) profiles during the sampling period May 2018 to April 2019 against mean profiles of milk LA from the same sampling months. Forage data were analysed in samples collected from one farm of each farm type, whereas milk data are mean values for all milk samples received from farms of that type for each sampling time. A: Conventional herd housed winter/grazing summer; B: Herd housed all year round; C: Organic herd; D: Spring block calving herd.

8.2 Second sampling and analysis period, 2021-2022

Mean basic milk quality (fat, protein and urea percentages) and FA data as estimated by NML are presented in Table 6. It should be noted that NML estimates and reports those milk FA that are present in greatest amounts, i.e. palmitic, oleic and stearic acids. However, it also reported estimated concentrations of SFA and unsaturated FA, but does not provide specific estimates of omega-3 FA – these are part of the PUFA content of milk fat.

Mean concentrations of milk fat, protein and urea were significantly affected by farming system, and by sampling month (Table 6). This is likely to have been caused by differences in stage of lactation curves of the cows on the different production systems at different times of the year.

Table 6. Mean milk composition and fatty acid concentrations for the four farming systems: A: Conventional herd housed winter/grazing summer; B: Herd housed all year round; C: Organic herd; D: Spring block calving herd.

	Farm Type					Significance of effect[2]	
	A	**B**	**C**	**D**	**SED[1]**	**Farming System**	**Sampling Month**
Milk fat %	4.37	4.04	4.43	4.88	0.154	***	***
Milk protein %	3.40	3.18	3.43	3.81	0.071	***	***
Milk urea %	0.026	0.022	0.020	0.019	0.0020	*	***
SFA[3], %	2.79	2.55	2.89	3.20	0.127	**	***
SFA, % TFA[4]	67.1	66.4	68.7	68.5	1.14	ns	***
UnSFA[5], %	1.37	1.29	1.32	1.45	0.047	*	***
UnSFA, % fat	31.4	32.0	29.8	30.0	1.09	ns	***
MUFA[6], %	1.06	0.99	1.01	1.15	0.042	**	***
MUFA, % fat	24.3	24.5	22.9	23.8	0.94	ns	***
PUFA[7], %	0.16	0.14	0.16	0.17	0.009	*	***
PUFA, % fat	3.63	3.50	3.53	3.62	0.250	ns	***
Palmitic acid, %	1.27	1.19	1.33	1.44	0.080	*	***
Palmitic acid, % fat	29.1	29.4	29.9	29.0	1.12	ns	***
Oleic acid, %	0.91	0.84	0.87	0.98	0.037	*	***
Oleic acid, % fat	20.8	20.9	19.6	20.4	0.87	ns	***
Stearic acid, %	0.40	0.38	0.40	0.43	0.017	*	***
Stearic acid, % fat	9.12	9.39	9.03	9.01	0.283	ns	***

[1]Maximum standard error of the differences of the means
[2]ns = not significant; * = P < 0.05; ** = P < 0.01; *** = P < 0.001
[3]SFA = saturated fatty acids (FA)
[4]TFA = total FA
[5]UnSFA = unsaturated FA
[6]MUFA = monounsaturated FA
[7]PUFA = polyunsaturated FA

Milk fat and protein concentrations are inversely correlated with milk yield, being highest at either end of lactation and lowest at peak lactation. This is clearly seen in Figures 13 and 14, with block calved cows (system D) having the highest milk fat and protein concentrations (respectively) in December and January, which coincides with the end of one lactation and the start of the next. Meanmilk fat and protein concentrations from cows on other systems, which were more likely to have beencalving at all times of the year, were much less variable.

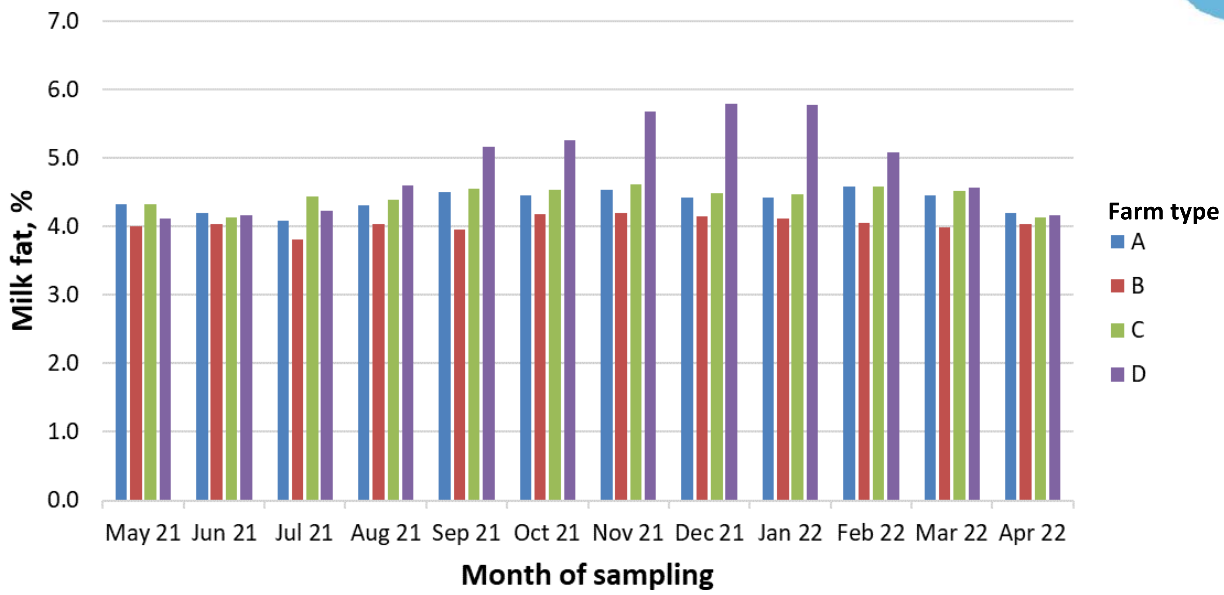

Figure 13. Mean concentrations of milk fat during the sampling period May 2021 to April 2022 for each farm type. A: Conventional herds housed winter/grazing summer; B: Herds housed all year round;C: Organic herds; D: Spring block calving herds.

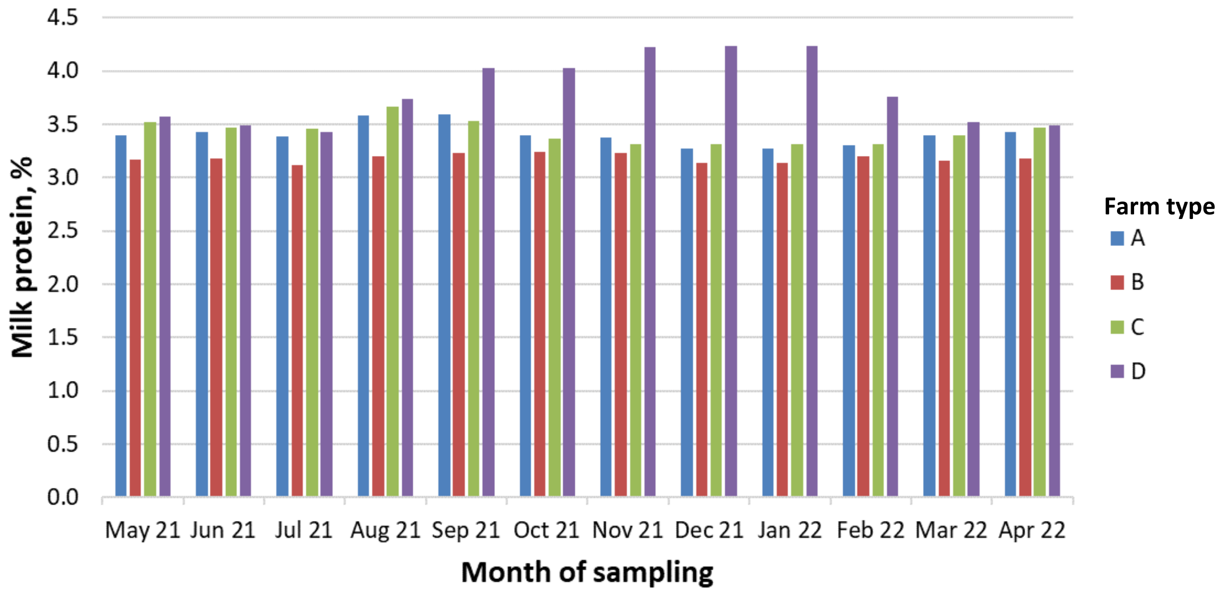

Figure 14. Mean concentrations of milk protein during the sampling period May 2021 to April 2022 for each farm type. A: Conventional herds housed winter/grazing summer; B: Herds housed all year round; C: Organic herds; D: Spring block calving herds.

Milk urea concentration is largely determined by diet, and was significantly affected by farming system and by month of the sampling period (Table 6). However, it is beyond the scope of this report to discuss this any further. Analyses of forage samples, which comprised part of the diets offered to thecows on the participating farms, are presented in full in Appendix 1. The analyses did not include FA composition of the diets, and therefore offer no value in the interpretation of milk FA data in the second sampling period.

The concentrations of individual FA estimated by NML, i.e., palmitic, oleic and stearic acids, together with the different groups of FA, i.e., SFA (Figure 15), unsaturated FA, MUFA and PUFA, were all significantly affected by farm type (Table 6). However, this is likely to have been caused by changes in milk fat concentration (Figure 13). Saturated FA as a percentage of TFA were reported by NML, and these were not significantly different between farm types (Figure 16). Similarly, when the individually-estimated FA and other FA groups were expressed as a percentage of milk fat, there were no significant differences between farm types.

Individual milk omega-6 and omega-3 FA were not reported by NML, presumably because the concentrations of these FA are very low in milk and cannot be accurately predicted. However, the milk concentration of PUFA, which ALA and LA contribute to, was reported. When expressed as a percentage of milk fat concentration, there was significant variation throughout the sampling period (Figure 17), but were not affected by farm type, with the lowest percentages tending to be found in the winter months. This is likely to have been caused by in changes in the diet from summer grazing to winter feeding of conserved forages, with the exception of year-round housed herds, which showed the least variation throughout the year.

Milk profiles of oleic, palmitic and stearic acid, expressed as a percentage of milk fat concentration, are presented in Figures 18, 19 and 20, respectively. The highest percentages of palmitic acid tended to occur over the winter, while the opposite was found for oleic and stearic acids. These patterns were very similar to those seen during the first sampling period, confirming the original data.

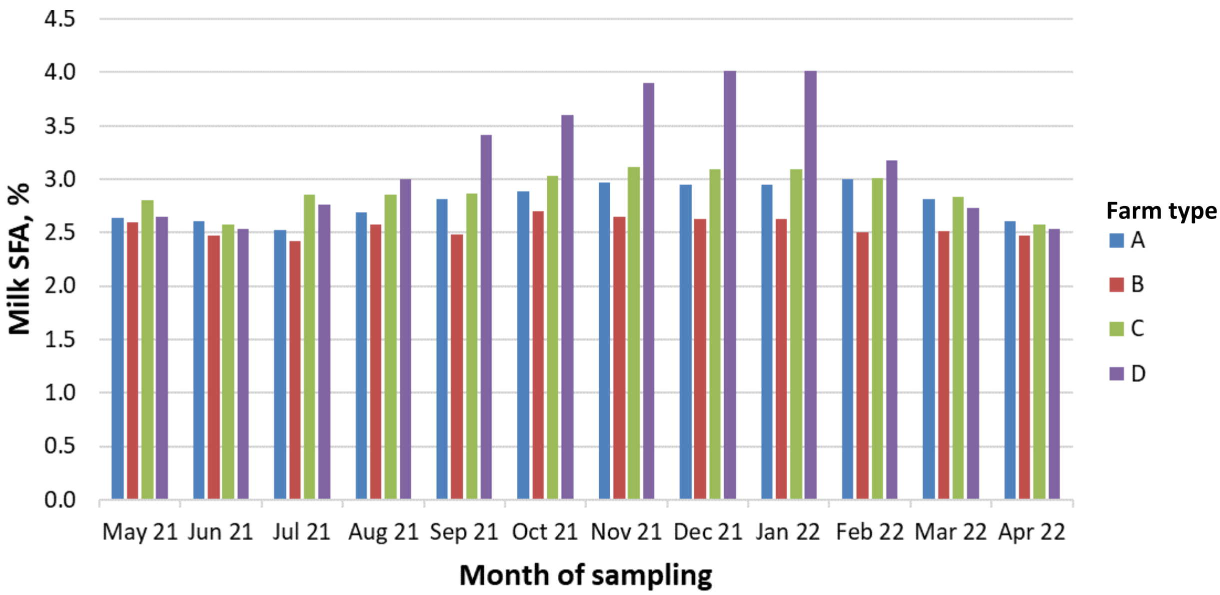

Figure 15. Mean concentrations of milk saturated FA (SFA) during the sampling period May 2021 to April 2022 for each farm type. A: Conventional herds housed winter/grazing summer; B: Herds housed all year round; C: Organic herds; D: Spring block calving herds.

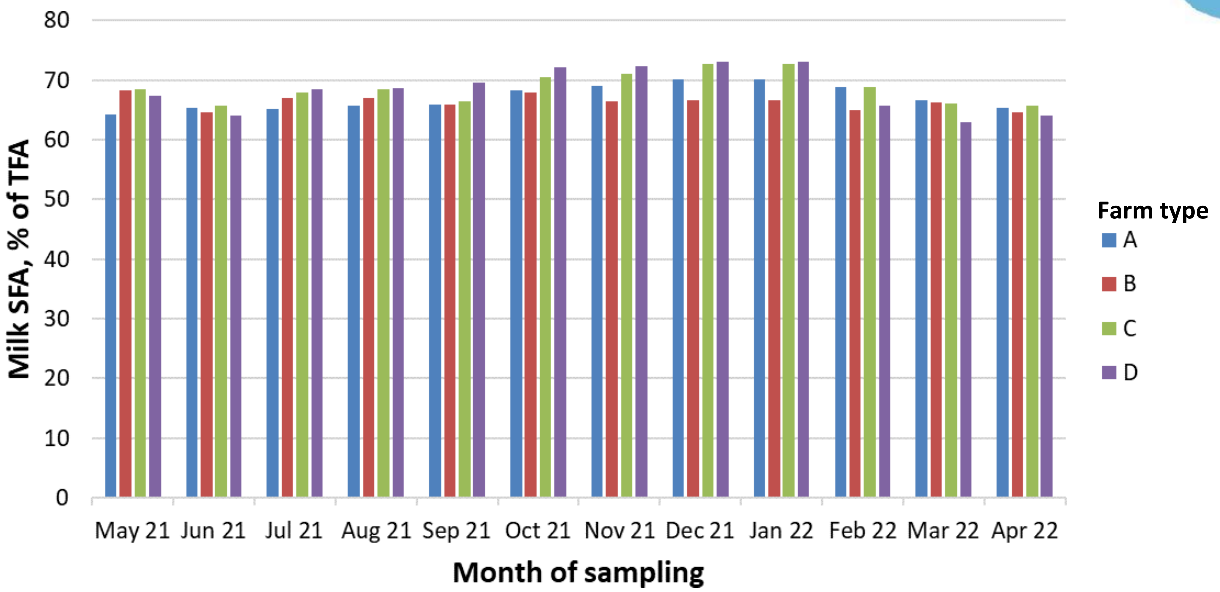

Figure 16. Mean profile of milk saturated FA (SFA, as % of total FA; TFA) during the sampling period May 2021 to April 2022 for each farm type. A: Conventional herds housed winter/grazing summer; B: Herds housed all year round; C: Organic herds; D: Spring block calving herds.

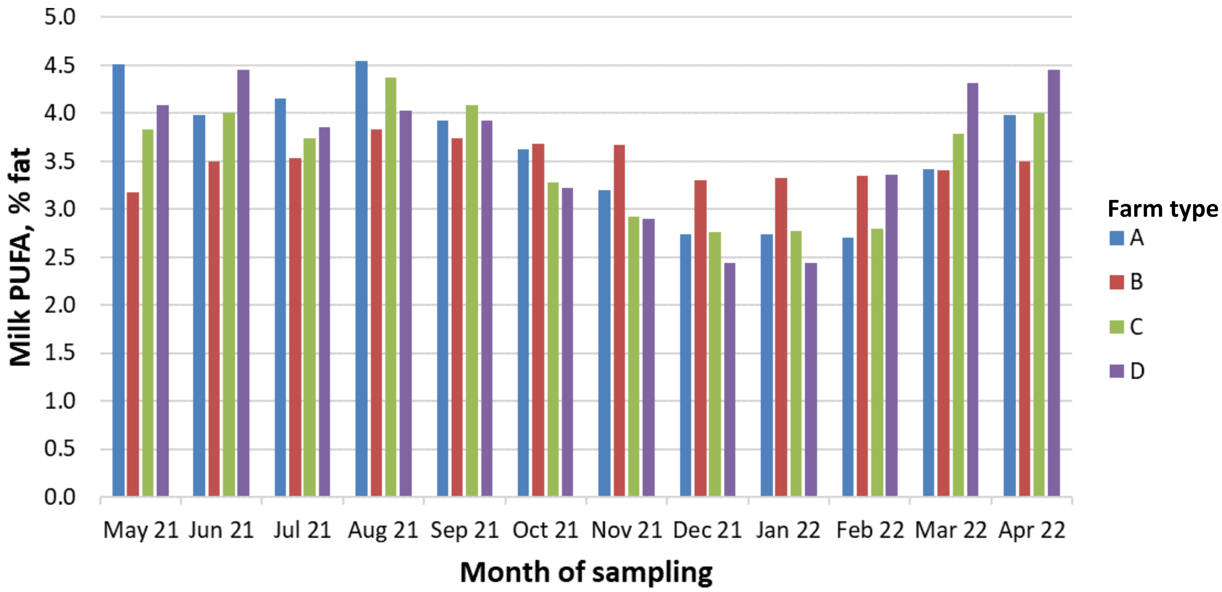

Figure 17. Mean profile of milk PUFA (as % of milk fat) during the sampling period May 2021 to April 2022 for each farm type. A: Conventional herds housed winter/grazing summer; B: Herds housed all year round; C: Organic herds; D: Spring block calving herds.

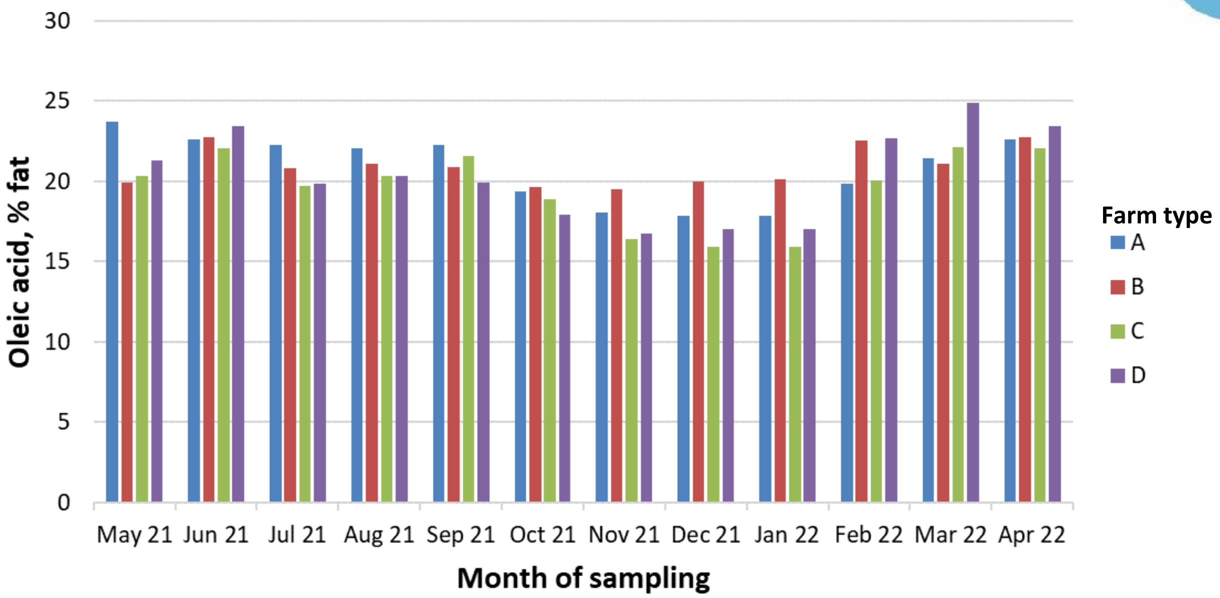

Figure 18. Mean profile of milk oleic acid (as % of milk fat) during the sampling period May 2021 to April 2022 for each farm type. A: Conventional herds housed winter/grazing summer; B: Herds housed all year round; C: Organic herds; D: Spring block calving herds.

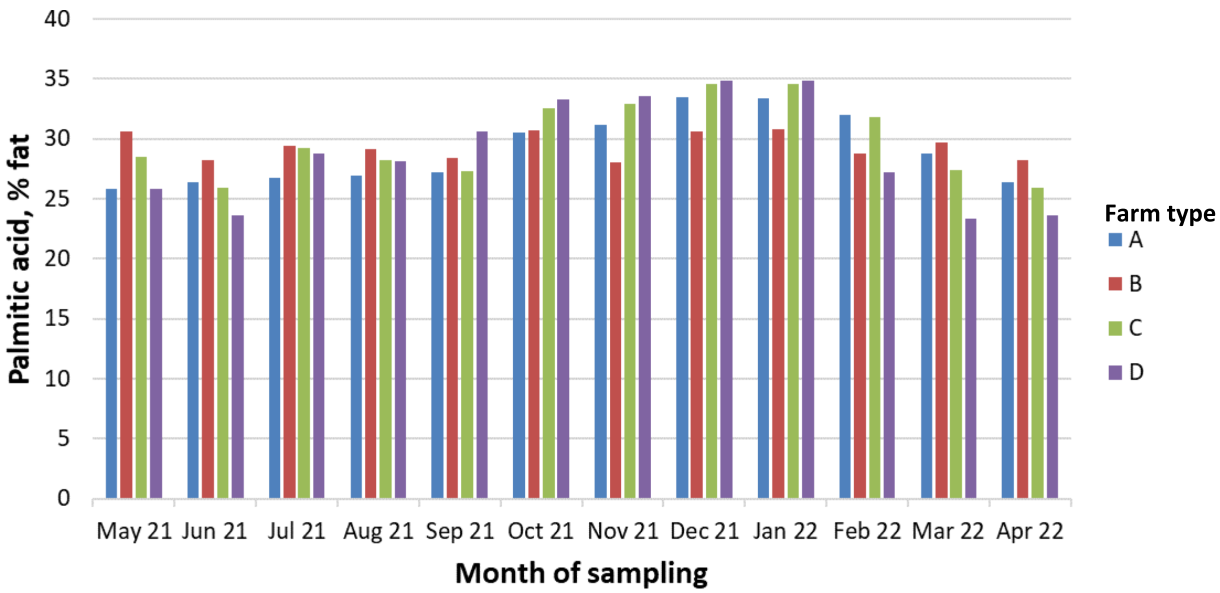

Figure 19. Mean profile of milk palmitic acid (as % of milk fat) during the sampling period May 2021 to April 2022 for each farm type. A: Conventional herds housed winter/grazing summer; B: Herds housed all year round; C: Organic herds; D: Spring block calving herds.

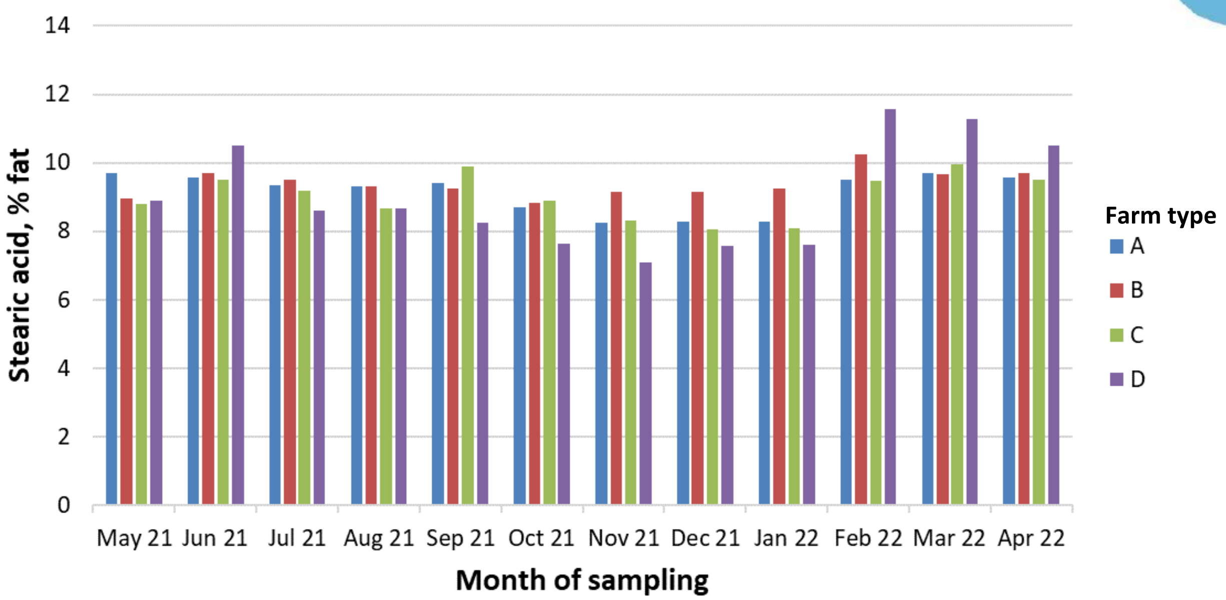

Figure 20. Mean profile of milk stearic acid (as % of milk fat) during the sampling period May 2021 to April 2022 for each farm type. A: Conventional herds housed winter/grazing summer; B: Herds housed all year round; C: Organic herds; D: Spring block calving herds.

9.0 Conclusions

It is well known that the fatty acid profile of milk fat is significantly influenced by the diet offered to the cow. The results found in this study are consistent with that. The FA data obtained from the first sampling period was more detailed, but limited in the numbers of samples received and therefore analysed than the milk composition data obtained in the second sampling period. Differences in the forage profiles of FA were somewhat reflected in milk FA profiles during the first sampling period, but forage was only part of the diets offered – the rest being concentrate feeds of various types. Differences between the FA content of maize silage and grass (fresh and conserved) were very large, and probably confounded the results. Changes in milk FA concentration reflect changes in milk fat concentration, while the profiles of milk FA, expressed as a percentage of total milk FA or milk fat, were not significantly affected by farm type in this study.

10.0 References

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11.0 Summary report provided to participants

Investigation of the effect of contrasting dairy production systems in West Wales on the profile of milk fatty acids (especially omega-3 and 6)

Executive Summary

- This study tracked the fatty acid composition of milk across multiple seasons in West Wales farms using differing dairy production systems. Participating farms represented either conventional (housed and grazed, typically for around six months), housed herds, organic herds and spring calving units.

- Forage samples were taken from a representative farm from each system and analysed for fatty acid composition. Forage samples varied in their fatty acid composition by type of forage and by season, with the biggest differences between systems evident during the summer grazing period.

- Milk fatty acid composition varied between units and farming system (in particular organic units), with greater omega-3 fatty composition evident in spring. There appeared to be a relationship between the fatty acid composition of representative forage samples and milk fatty acid composition, as might be expected. .

- Various studies have shown beneficial effects of the fatty acid content of grass on cow health, human health and methane production. West Wales is a strong grass growing area, and there may be opportunities promote milk produced by dairy units in the area farming, particularly linked to the quality & fatty acid profile of fresh grass.

What are fatty acids?

Milk fat is predominantly (~95-98%) made up of triglycerides which are molecules of glycerol each attached to three fatty acids. These fatty acids are made up of carbon chains of variable lengths, with each carbon atom normally having two hydrogen atoms bonded to it.

Occasionally, a carbon atom only has one hydrogen atom attached so uses its 'spare' bond on the next carbon atom in the chain (forming a double-bond). A fatty acid with one or more of these carbon double bonds is called 'unsaturated'. If a carbon has one double bond, it is called 'mono-unsaturated' (mono meaning 'one'). If it has more than one double bond, it is called 'poly-unsaturated' (poly meaning 'many'). Unsaturated fatty acids have a 'kink' in their chain at the double bond(s) rather than the straight line of a saturated fatty acid. This changes the way these fatty acids (and the fats they make up) behave, and their effects within the body.

Fats with more unsaturated fatty acids are more likely to be liquid at room temperature; butter made from milk with higher unsaturated fatty acids / fat is easier to melt and spread.

Where the unsaturated bond occurs along the fatty acid chain can also influence how it behaves in the body. The 'omega number' tells us how far the double bond is away from the tail of the fatty acid. Omega-3 (or n-3) fatty acids have a double bond three carbon atoms away from the end of the chain; Omega-6 (or n-6) fatty acids have the double bond 6 carbon atoms from the end. Omega-3 and Omega-6 are important fatty acids to humans, as our bodies cannot make them, and instead rely on absorbing them from our food. They are sometimes called 'essential fatty acids' for this reason.

Why are fatty acids of interest to us?

Fatty acids (FAs) are of interest to us for four main reasons:

1) The type of fatty acids we feed our cows may influence their health, fertility and GHG output.
2) The fatty acid profile of milk can alter the properties of the milk (consistency, taste)
3) The fatty acids consumed by humans may influence their health
4) Ideas and science surrounding FAs may alter consumer demand and market opportunities

How do fatty acids affect cow health and sustainability?

The type of fatty acids we feed cows influences what fatty acids they absorb and the composition of fat in their body, as well as their milk. Some fatty acids, particularly unsaturated ones, are toxic to some of the rumen microbes, and in particular can inhibit some of the methane-producing species, reducing greenhouse gas (GHG) output of cattle.

The rumen microbes tend to saturate (add hydrogen atoms to) unsaturated fatty acids, which is why milk tends to be relatively high (65-70%) in saturated fat. However, this process takes time, and diets containing high levels of unsaturated fatty acids usually result in enough escaping the rumen and then being absorbed in the intestines. Under some circumstances, certain partially-saturated fatty acids leaving the rumen are absorbed and suppress butterfat production by the udder; one of the reasons why milk fat often drops on Spring grass.

Some research studies have suggested that higher omega-3 fatty acid intake may improve fertility in cattle. Fatty acids are the building blocks of several types of hormones, including prostaglandins, and the types of fatty acids used can influence which hormones are produced. Cattle fed higher omega-3 diets appear to have higher levels in their ovum (eggs) and follicles, and often have a better chance of pregnancy at service. The role of omega-6 fatty acids is less well understood, but may exacerbate inflammatory responses in animals and possibly be beneficial around calving, although excess inflammation around calving is also known to be detrimental. More research is still needed in this area.

Animals fed diets higher in unsaturated fats typically produce milk with a higher proportion of unsaturated fat; equally, cattle fed higher proportions of omega-3 and omega-6 have these at higher levels in their milk. The fatty acid profile of milk and milk products can influence their physical properties. Of particular interest is the improved 'spreadability' and melting point of butter with higher unsaturated fats, and softer cheeses. There may be potential marketing opportunities for such products.

How do fatty acids affect human health and vitality?

Although still an active area of research and debate, there have been a number of studies that have linked omega-3 intakes with positive human health, including reducing risk of cardiac disease, obesity, and reducing inflammation in people with diabetes, rheumatoid arthritis and pre-existing cardiac diseases. Some studies have also indicated potential benefits to mood and cognition, but further work is being undertaken in this area.

While omega-6 is an essential fatty acid and a needed dietary component of humans, there is some evidence that the ratio of omega-6:omega-3 is increasing in modern diets, and that an excessively high ratio may have detrimental effects on human health. The 'healthy' ratio of omega-6:omega-3 currently suggested ranges between 1:1 to 4:1, depending on the study.

What influences the fatty acid profile of milk?

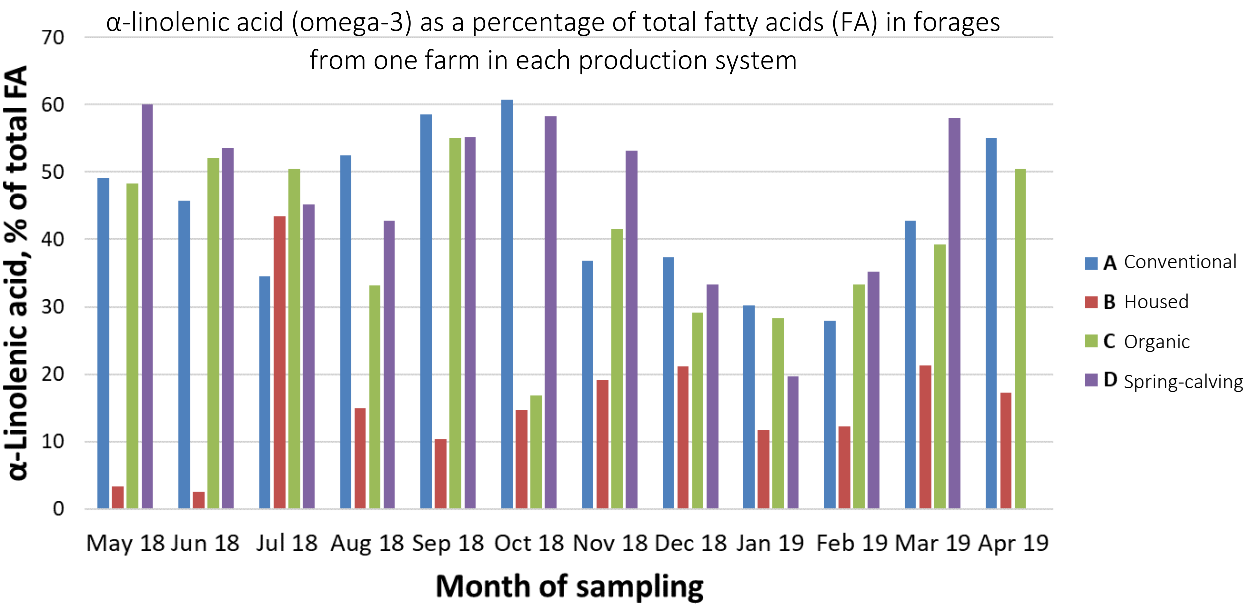
The fatty acid profile of the cows' diets has the major influence on milk fatty acid profile. Feeds high in unsaturated fatty acids (including omega-3 and omega-6 FAs) tend to be reflected in the milk. Forage usually has the greatest influence, as it represents the greatest proportion of the diet in cattle. The fatty acid profile of forage is influenced by:

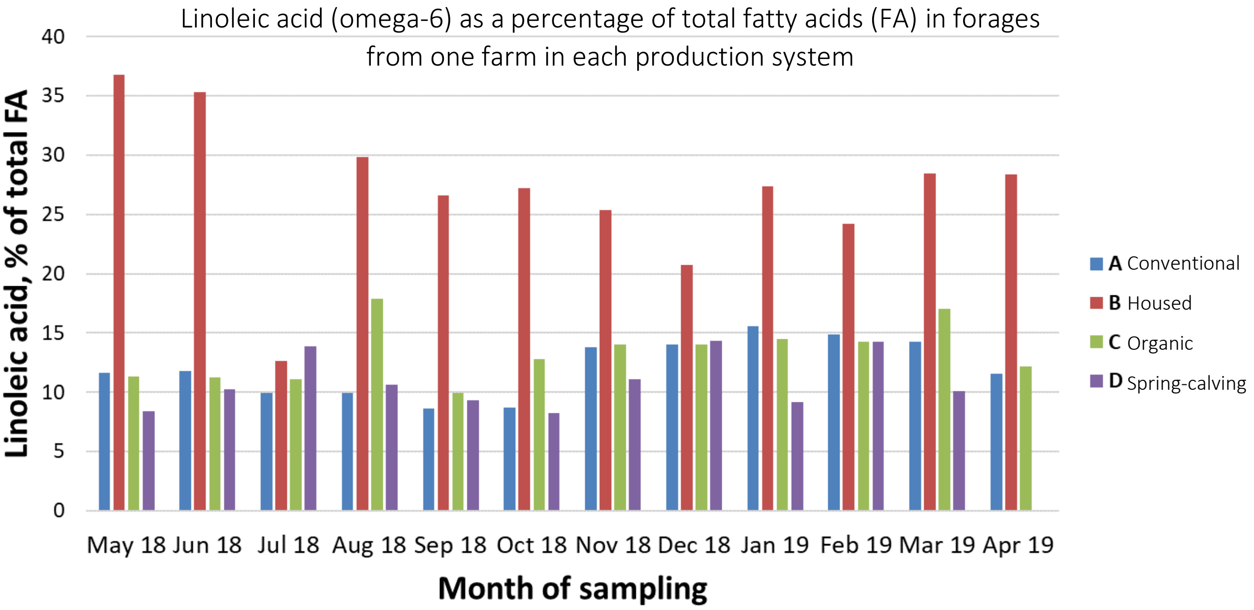
- The type of forage. E.g. maize silage has greater omega-6 FA relative to omega-3 cf. grass.
- The time of year. Total fatty acids in grass are higher in spring and autumn, with higher levels of unsaturated fatty acids, including omega-3 FAs, in the spring period.
- Forage management. Longer grazing intervals (e.g. >38 days) decrease unsaturated fatty acids, including omega-3 and omega-6 levels.

Other supplementary feeds we give to cattle have their own fatty acid profile, and this influences the total dietary intake of fatty acids, the proportion that are unsaturated and the relative levels of omega-3 and omega-6 FAs. For example, grains such as wheat have little total fat (around 2% of dry matter), which is largely unsaturated (84% of total fat) but have very high levels of omega 6 relative to omega 3 (20:1). C16 fats that are fed to increase butterfat, are 99% fat, high in saturated fat (~94%) and have low levels of omega 3 and 6.

Within this study, measurements of the fatty acid composition of forages from one farm in each type of farming system was made. While useful, it is not possible to assume that forages from other farms within that system would have been similar, although it provided a good indication. The proportion of this forage in the ration for herds would obviously have varied considerably across different systems and time of year.

There was a marked seasonal variation in forage omega-3 (α-linolenic) FA in all systems, with concentrations lower in the winter, reflecting silage intakes instead of grazed grass. While silage fatty acids are not radically changed by ensiling, crops are harvested at a greater stage of maturity and therefore tend to be lower; equally cuts made mid-summer are also likely to be lower still.

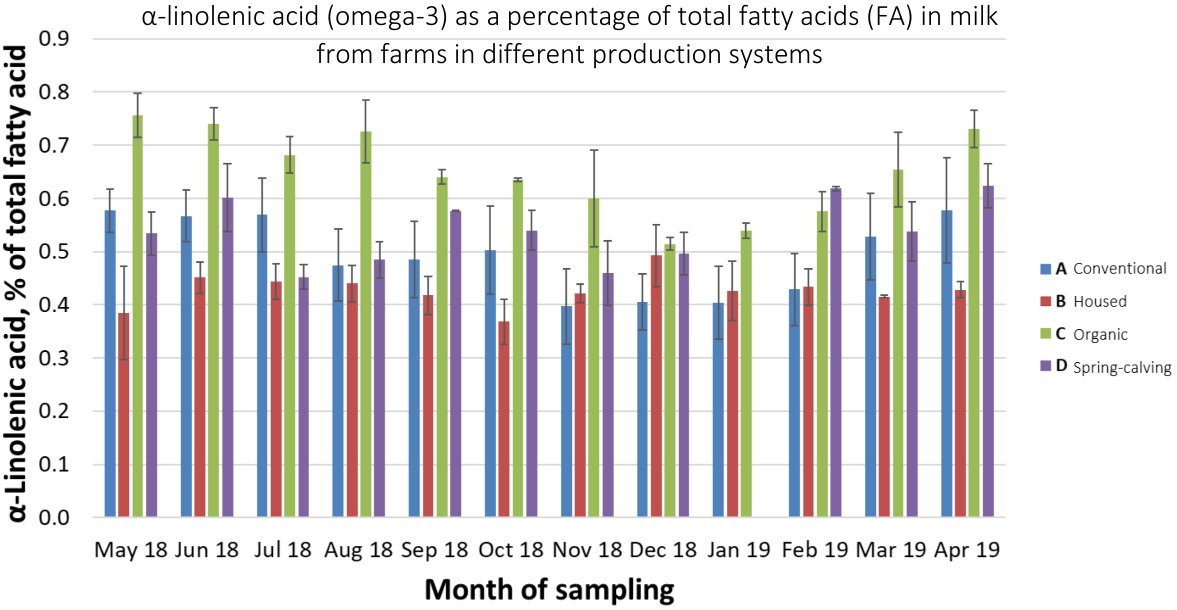




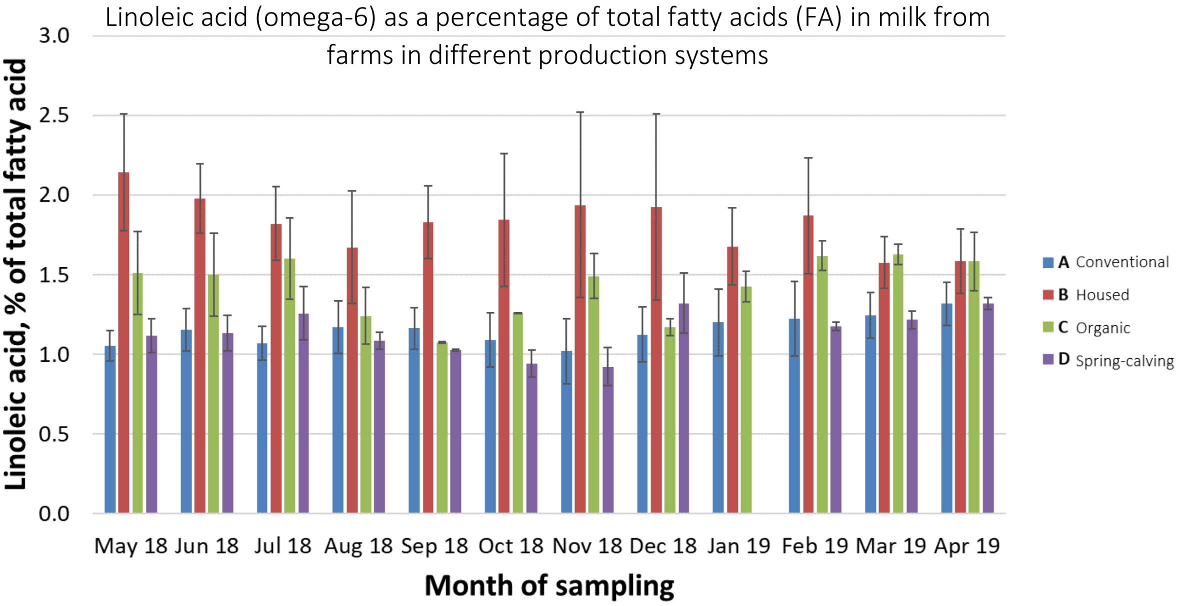
Omega-6 content of forages (using linoleic acid) was higher in winter months in conventional, organic and spring calving herds, likely reflecting silage feeding. The high level of omega 6 in the housed herd reflects the feeding of maize silage, which tends to be higher in this type of fatty acid.

Milk fatty acids

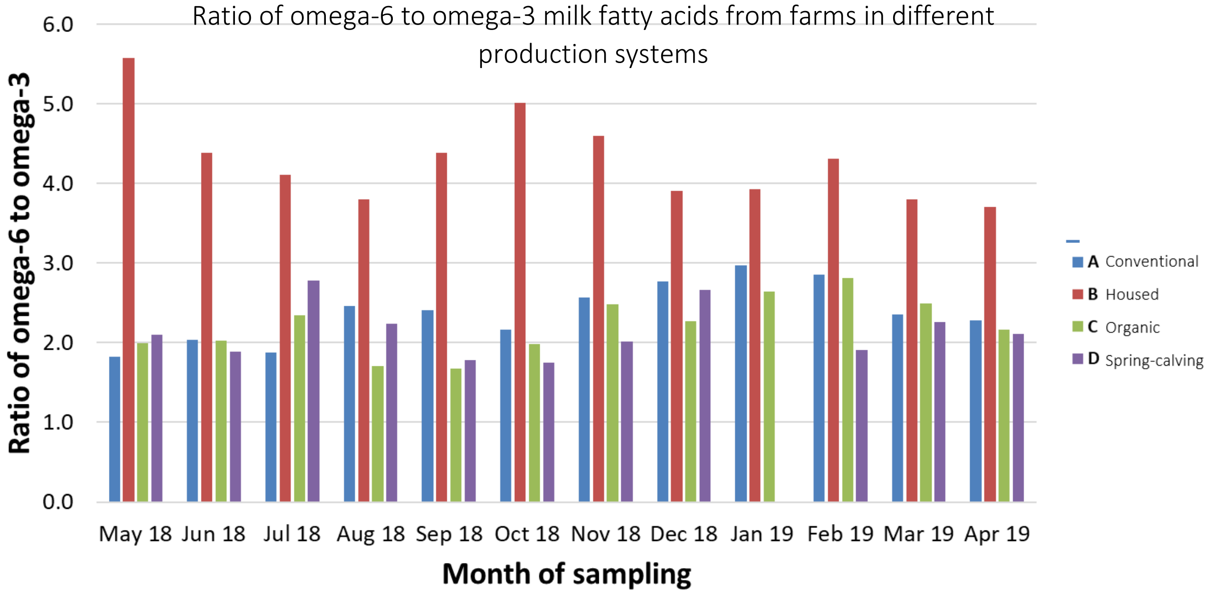
The amount of α-linolenic acid (omega-3) varied across the season and between different farming systems. In broad terms, during the summer months more variation was seen in omega-3 content of the milk, with organic unit's average being highest overall. Conventional and spring calving herd averages were in general higher than housed herds early and later in the grazing season, likely reflecting grass omega-3 content, but showed less difference mid-summer and during the winter where silage predominated.



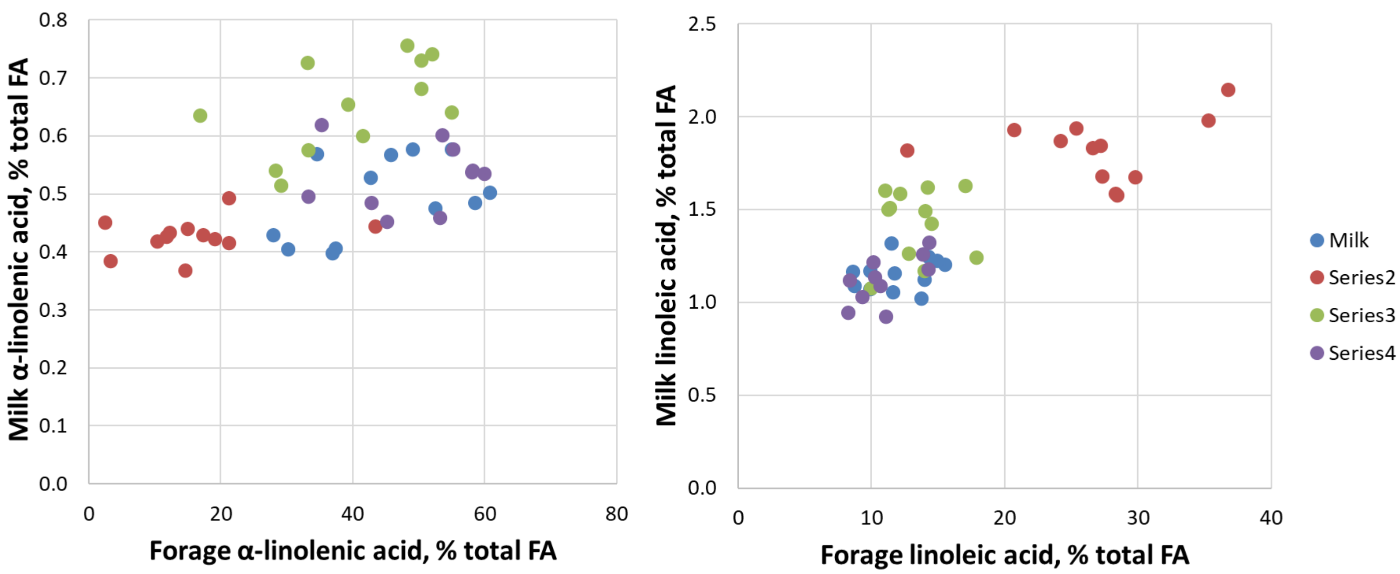
Linoleic acid (Omega-6) average was higher in housed herds than conventional and spring-calving systems during the grazing period, likely reflecting maize feeding in housed systems, although there was variability between the farms. Organic farms were a little higher than conventional and spring calving systems across the grazing season, perhaps reflecting different forage species, or possibly the oil profile in the common organic supplementary feeds.



Average ratios of milk linoleic acid (omega-6) to α-linolenic acid (omega-3) all lay within what is thought to be the 'healthy' range for a human diet, with the exception of a small number months for housed systems. There was variation between different farms, however, and it is likely that many would still have sat within that range at an individual level.



The relationship between forage linoleic acid (omega-6) to α-linolenic acid (omega-3) showed some correlation between milk content of the respective fatty acids (acknowledging forage samples were from one farm per group). This is nice confirmation that forage is a key area to focus on if increasing milk omega-3 and omega-6 levels were desired.



What can we take away from this trial?

The way in which we feed cows can significantly affect the fatty acid composition of their milk. Giving cows feeds that are naturally high in omega-3 or omega-6 fatty acids can increase the amount of those

fatty acids in their milk. These, and other unsaturated fatty acids, can influence different physical properties such as the spreadability of butter, and the nutritional value to consumers.
The fatty acid profile of bulk milk is easily and cheaply tested and can be used as a basis for calculating the methane output per litre of milk produced. Various studies have shown that cows consuming significant amounts of spring grass have lower methane emissions per litre due to the FA content of the grass.

Farms in West Wales are particularly well-placed to produce milk higher in omega-3, given that fresh grass is naturally high in these fatty acids, and that West Wales is one of the prime grass growing regions of the country. Farms unable to graze still have options to increase the omega-3 content of their milk through techniques such as zero grazing, or use of feeds naturally high in omega-3 such as extruded linseed. It was also encouraging to see that the ratio of omega 3 : omega 6 in milk remained within the current recommended threshold for the majority of farming systems throughout the year.

There are strict labelling requirements relating to claims about fatty acids. A food may be labelled as a source of omega-3 product if it contains at least 0.3g α-linolenic acid per 100g and per 100kcal. In this respect, on a fresh-weight basis milk it would be difficult to make a label claim with on a fresh milk product. However, there is potential scope to look at other marketing avenues, as exemplified by the French Bleu Blanc Coeur initiative.

In summary, this study successfully demonstrated:

- The variance in forage fatty acid composition across different points of the season in different farming systems.
- That variations in fatty acid composition did vary between farming systems, but that such differences were largely restricted to the summer months, and appear linked to grass intakes.
- Potential opportunities to promote the brand of West Wales dairy farming, particularly linked to the quality and fatty acid profile of fresh grass.